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In vitro screening for drought tolerance of some chickpea varieties in Bangladesh

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Abstract

The aim of the present study was to study physiological response of seven chickpea varieties viz. Binachola-2, Binachola-3, Binachola-4, Binachola-5, Binachola-6, Binachola-7 and Binachola-8 at different levels of drought stress induced by polyethylene glycol (PEG). Five different concentrations (0, 20, 35, 50, 60 g/L) of PEG 6000 was added in MS medium to create five different levels of drought stress for *in vitro* screening of drought tolerant chick pea varieties. Data were recorded on germination percentage, fresh weight, shoot length, root length, dry weight, turgid weight, relative water content (RWC) and proline content. The seven chickpea varieties differed significantly for different parameters in response to the drought stress. Binachola-2 and Binachola-7 showed the best performance for all the parameters studied. At the highest dose of PEG (60 g/l), in the most water deficient condition, fresh weight was recorded 0.59 g and 0.84 g, Shoot lengths of 2.10 cm and 3.75 cm, root lengths of 1.15 cm and 1.00 cm, turgid weight of 0.960 g and 0.970 g, dry weight of 0.13 g and 0.21 g, relative water contents (RWC) of 85.71% and 83.33% were recorded in Binachola-2 and Binachola-7, respectively. Increased proline content was recorded with the increasing level of PEG concentration. Proline content 0.533g/100g FW and 0.598g/100g FW were observed in Binachola-2 and Binachola-7, respectively under the influence of PEG at 60 g/l. Data recorded for Binachola-3, Binachola-4, Binachola-5, Binachola-6 and Binachola-8 for the studied parameters revealed that they showed susceptible response against higher drought stress level generated by PEG. The data of this experiment revealed that, the accumulation of proline was significantly greater under drought stress. Thus, it is evident that Binachola-2 and Binachola-7 performed better against drought stress condition. These results of physiological and biochemical parameters may be utilized as a selection indicator for breeding program and used as a baseline for improvement of chickpea varieties in Bangladesh.

Key words: Chick pea, drought stress, polyethylene glycol, relative water content, proline content

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Introduction

Chickpea (*Cicer arietinum* L.) is one of the most important pulse crops under the family Fabaceae and subfamily Faboideae. Chickpea ranks third in terms of area contributing around 12% of total pulse production of Bangladesh. The area of chickpea is 8,097 hectares with total production of 7000 metric tons per year (BBS, 2011). Chickpeas have a protein digestibility corrected amino acid score of about 76 percent which is higher than fruits, vegetables, many other legumes, and cereals (Milan *et al.*, 2007). It is able to drive more than 70% of nitrogen from symbiotic dinitrogen fixation, which makes it a promising crop for "alternative agriculture" that is now attracting a considerable attention in the industrialized world. The heavy demand created by the pressure of increasing population in the developing world requires a tremendous scientific effort to meet the requirements of food, fiber, fuel and other necessities of life (Sujatha *et al.*, 2007).

Several factors such as biotic and abiotic stresses reduce yield of chickpea. Among the abiotic stress factors, drought stress is relatively important in chickpea which causes a 40–50% reduction in yield globally (Millan *et al.*, 2006). As the economy of Bangladesh is mainly agriculture oriented, crop failure comes as significant strain to its socioeconomic structure. In northern region of Bangladesh, chickpea is continuously exposed to increasing drought and high temperature during flowering and maturity stages due to insufficient and irregular rainfall which significantly affect chick pea yield every year.

Tissue culture based selection has emerged as a feasible and cost-effective tool for screening stress tolerant crop plants. Chemical agents such as NaCl for salt tolerance, PEG or mannitol or sorbitol can be used to screen for abiotic stress tolerant crop plants (Errabii *et al.*, 2008). In most of the cases PEG is used to induce drought stress. PEG of high molecular weight has long been used as osmoticum lowering the water potential of nutrient solutions without being taken up or being phytotoxic (Hassan *et al.*, 2004). Plants can partly protect themselves against mild drought stress by accumulating osmolytes. Proline is one of the most common compatible osmolytes in drought stressed plants. The accumulation of proline in plant tissues is also a clear marker for environmental stress, particularly in plants under drought stress. Proline accumulation may also be part of the stress signal influencing adaptive responses (Maggio *et al.*, 2002).

In the present study, seven Bangladeshi chickpea varieties were studied *in vitro* under different concentrations of PEG generated drought stress and plant responses were observed on some physiological (germination percentage, fresh weight, root length, shoot length, dry weight, turgid weight, relative water content) and biochemical (proline accumulation) parameters to study the physiological and biochemical response of chickpea varieties at different levels of water shortage at seedling stage, and to develop a rapid *in vitro* screening method for drought tolerant chickpea varieties in Bangladesh.

Materials and Methods

The experiment was conducted during the period from September, 2014 to May, 2015 in the USDA Biotech Lab of Department of Biotechnology, Post-graduate Biochemistry Laboratory of Department of Biochemistry and Molecular Biotechnology and Central Laboratory of Bangladesh Agricultural University (BAU), Mymensingh. Seven chickpea varieties viz. Binachola-2, Binachola-3, Binachola-4, Binachola-5, Binachola-6, Binachola-7 and Binachola-8 were selected to observe their tolerance level against drought stress using MS (Murashige and Skoog, 1962) medium containing five different concentration levels (0, 20, 35, 50, 60 g/l) of PEG. Mature seeds of chickpea were inoculated to glass vial (70mm) containing MS medium supplemented with different concentrations of PEG. The incubation room was maintained at 25 ± 1^0C and a light intensity of 2000-3000lux from fluorescent tube. The photoperiod was maintained at 16 hours light and 8 hours dark (16L/8D) and the relative humidity was 60-70%. The experiment replicated four times. After 30 days of seed inoculation, data were recorded to investigate the effect of different treatment on germination percentage, fresh weight (g), shoot and root length (cm), turgid and dry weight (gm), relative water content (RWC) and proline content (g g^{-1} fresh weight).

To calculate the percentage of germination, the seeds were observed one day interval. The mean values of the germinated seeds from each of the replication were collected and statistically analyzed using following formula:

$$\text{Percent germination} = \frac{\text{Number of seeds germinated and became seedlings}}{\text{Number of seeds set for germination}} \times 100$$

Fresh weight (gm) of plantlets was measured by using an electric balance at 30 days after inoculation (DAI). Shoot length (cm) was measured from the base to the apex of the shoots and root length (cm) was measured from the base of the root to the root tip using a scale after 30 days of inoculation (DAI). Turgid weight of plantlets was measured in gram by using an electric balance. For this purpose, the plantlets were taken out from vial and were soaked in distilled water for 14 hours. Then the weight was measured. Dry weights (gm) of plantlets were measured by incubating them at 60^0C for 72 hours. Then the dry weight was measured. The mean for all the parameters was calculated for further analysis.

To calculate the relative water content (RWC), the leaves were cut and the relative water content (RWC) of leaf was determined as follows:

$$RWC = \frac{(\text{fresh weight} - \text{dried weight})}{(\text{fully turgid weight} - \text{dried weight})} \times 100$$

To determine the fully turgid weight, the leaves were kept in distilled water in the darkness at 4^0C to minimize respiration losses until they reached a constant weight (full turgor, typically after 12h). The leaf dry weight was obtained after 48 h at 70^0C in an oven. Four replications per treatment were maintained.

Proline accumulation in fresh leaves was determined according to the method of Bates *et al*. (1973). Free proline was extracted from the leaves of plants using aqueous sulfosalicylic acid. The filtrate (1 ml) was mixed with equal volumes of glacial acetic acid and ninhydrin reagent (1.25 g ninhydrin, 30 ml of glacial acetic acid, 20 ml 6 NH_3PO_4) and incubated for 1 h at 100°C. The reaction was stopped by placing the test tubes in cold water. The samples were vigorously mixed with 4ml of toluene. The light absorption of toluene phase was estimated at 520 nm using Pharmacia LKB-Novaspec II model spectrophotometer. Calculation for proline concentration was done using a standard curve. Free proline content was expressed as g g^{-1} fresh weight.

The recorded data were statistically analyzed using Microsoft Statistical (MSTAT) programme and Microsoft Excel wherever applicable. The data for the characters under the present study were statistically analyzed following Completely Randomized Design (CRD) and analysis of variance (ANOVA) technique. The analysis of variance was performed and means were compared by Least Significant Difference (LSD) test at 5% level of probability for results (Gomez and Gomez, 1984) and the mean differences were adjusted by DMRT (Ducan's Multiple Range Test) and the ranking was indicated by letters.

Results and Discussion

Germination Percentage

Germination percentage of seven chickpea varieties under different drought stress were recorded and shown in Table 1. A highly significant variation was observed in seed germination percentage at different (3, 7, 10, 15, 20, 25 and 30) days after inoculation to different levels of polyethylene glycol (PEG) (0, 20, 35, 50, 60 g/l) induced drought levels. According to data presented in Table 1, the highest germination percentage (100%) was observed in V_2T_1, V_2T_2, V_2T_3, V_7T_1, V_7T_2, V_7T_3, V_8T_1 and V_8T_2 whereas in V_3T_4, V_3T_5, V_4T_3, V_4T_5, V_5T_4, V_5T_5, V_6T_5 and V_8T_5 showed the lowest germination percentage (0%). The other values showed intermediate status. The percentage of germination decreased with the increase in PEG concentration. At T_5 (60 g/l of PEG) seed germination was found only in Binachola-2 and Binachola-7. A germination percentage of 66.67% was recorded in V_2T_5 and V_7T_5 at 3 days after inoculation. Other chickpea varieties (Binachola-3, Binachola-4, Binachola-5, Binachola-6 and Binachola-8) did not germinate under the influence of severe osmotic stress generated by the highest dose of PEG (60 g/l) at different (3, 7, 10, 15, 20, 25 and 30) days after inoculation (Figure 1a and Table 1).

Total fresh weight

Total fresh weight is the major growth parameter measuring from the base to apex of the shoot and root of plantlet. Fresh weight of chick pea varieties showed significant variation in culture medium supplemented with different concentrations (0, 20, 35, 50, 60 g/l) of polyethylene glycol at 30 days after inoculation (Table 1). The highest value of fresh weight (1.85g) was recorded in V_8T_2, while the lowest value (0.000g) was recorded in V_3T_4, V_3T_5, V_4T_3, V_4T_5, V_5T_4, V_5T_5, V_6T_5 and V_8T_5. It was observed that under the influence of severe osmotic stress (PEG 60 g/l), only Binachola-2 and Binachola-7 regenerate shoots and roots whereas Binachola-3, Binachola-4, Binachola-5, Binachola-6, and Binachola-8 did not show any growth (Figure 1-b). It might be due to severe water stress that stopped the growth of the susceptible varieties whereas only two varieties showed the potential to resist the higher level of water stress.

Shoot length of chickpea varieties under drought stress

Shoot length showed significant variation under different concentrations of PEG at 30 days after inoculation. The longest shoot was (5.25cm) observed in control treatment, which was statistically different from other values. At T_5 (60 g/l of PEG), shoot formation was observed only in Binachola-2 and Binachola-7 and their shoot lengths were 2.10cm and 3.75cm, respectively (Figure 1C and Table 1). These results indicated that the high level of water stress negatively influences the shoot length of chick pea varieties. The lowest value (0.00cm) was observed in V_3T_4, V_3T_5, V_4T_3, V_4T_5, V_5T_4, V_5T_5, V_6T_5 and V_8T_5 (Table 1). The remaining treatments showed intermediate status.

Root length

Significant difference was observed in root length of plantlet at culture medium supplemented with different concentrations of PEG (Fig. 1D). Root length decreased with the increasing level of PEG concentration (Table 1) in the culture medium. The highest root length (3.00 cm) was recorded in V_8T_4, while the lowest value (0.000cm) was recorded in V_3T_4, V_3T_5, V_4T_1, V_4T_3, V_4T_5, V_5T_4, V_5T_5, V_6T_4, V_6T_5 and V_8T_5. The second highest root length (1.80 cm) was found in V8T2 followed by V2T1 (1.75 cm) and V3T2 (1.75 cm), respectively. At T_5 (60 g/l of PEG) the root was found only in Binachola-2 and Binachola-7 and root lengths of 1.15cm and 1.00cm were recorded after 30 days after inoculation, respectively (Figure 1-d and Table 1).

Dry weight and turgid weight

Dry weight showed significant variation under different concentrations (0, 20, 35, 50, 60 g/l) of polyethylene glycol at 35 days after inoculation. Dry weight decreased with the increasing level of PEG concentration (Table 1). The highest dry weight (0.530g) was recorded in V_3T_2, while the lowest value (0.000g) was recorded in V_3T_4, V_3T_5, V_4T_3, V_4T_5, V_5T_4, V_5T_5, V_6T_5 and V_8T_5. The second highest dry weight (1.80 cm) was found in V5T2 followed by V5T3 (0.250g) and V4T2 (0.226g), respectively. At T_5 (60 g/l of PEG), formation of shoots and roots were observed only in Binachola-2 and Binachola-7 whereas other varieties did not show any response under the influence of severe osmotic stress generated by the highest dose of PEG (60 g/l) (Table 1, Figure 1-e).

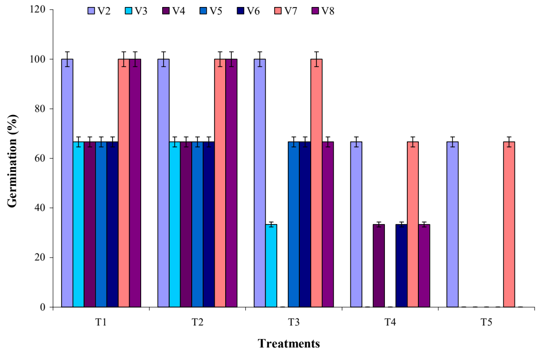
Turgid weight of seven chick pea varieties showed significant variation under different concentrations (0, 20, 35, 50, 60 g/l) of polyethylene glycol at 31 days after inoculation. Turgid weight decreased with the increasing level of PEG concentration. The highest turgid weight (1.81g) was recorded in V_8T_2, while the lowest value (0.000g) was recorded in V_3T_4, V_3T_5, V_4T_3, V_4T_5, V_5T_4, V_5T_5, V_6T_5 and V_8T_5. The second highest turgid weight (1.80g) was found in V2T1 followed by V8T1 (1.75 g) and V6T1 (1.62 g), respectively (Figure 1-e and Plate. 1-a). Under the influence of severe osmotic stress generated by the highest dose of PEG (T_5, 60 g/l), significant turgid weight were recorded only in Binachola-2 (0.670g) and Binachola-7 (0.970g) (Table 1, Figure 1f).

Relative water content

Relative water content showed significant variation at different levels of drought stress induced by different concentrations (0, 20, 35, 50, 60 g/l) of polyethylene glycol on seven chickpea varieties (Table 1, Figure 2a). The highest relative water content (98.81%) was recorded in V_8T_2, while the lowest value (0.000%) was recorded in V_3T_4, V_3T_5, V_4T_3, V_4T_5, V_5T_4, V_5T_5, V_6T_5 and V_8T_5. The second highest relative water content (98.04%) was found in V8T4 followed by V2T2 (96.81%) and V4T4 (96.30%), respectively. At T_5 (60 g/l of PEG), only in Binachola-2 (85.71%) and Binachola-7 (83.33%) showed significant response for relative water content.

Proline content

The proline accumulation showed significant variation under different osmotic stress (Table 1). At the highest level of drought stress (T_5 = 60 g/l of PEG) the significant amount of proline was found only in Binachola-2 and Binachola-7.



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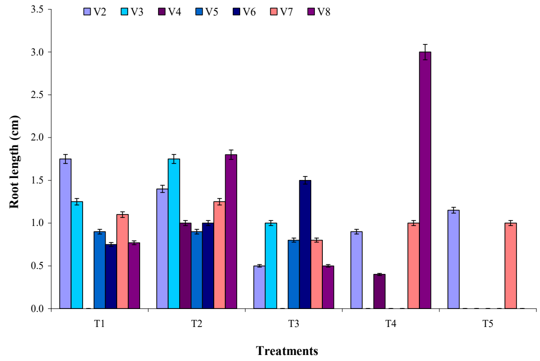

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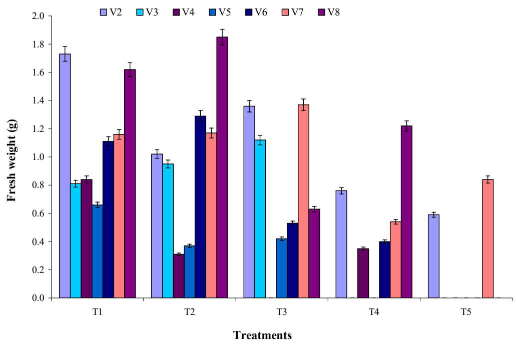

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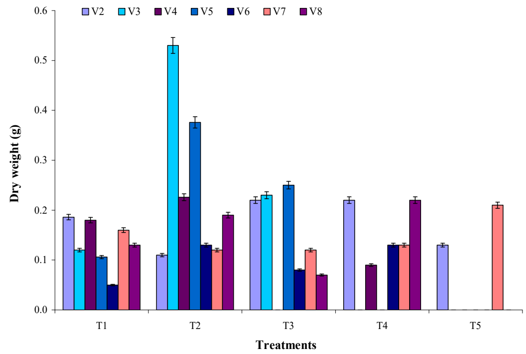

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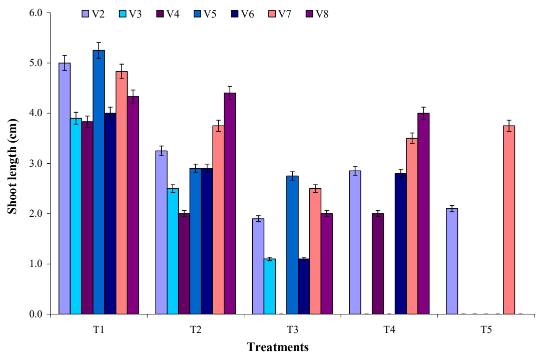

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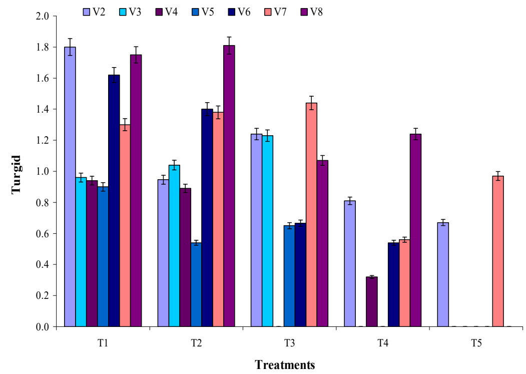


(f)

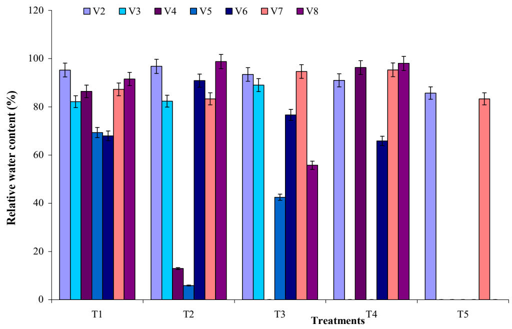
(V_2= Binachola-2, V_3 = Binachola-3, V_4 = Binachola-4, V_5 = Binachola-5, V_6 = Binachola-6, V_7 = Binachola-7, V_8=Binachola-8; T_1 = 0g/l, T_2 = 20 g/l, T_3 = 35 g/l, T_4 = 50 g/l, T_5 = 60 g/l of PEG)

Figure 1. Effect of different concentrations of polyethylene glycol (PEG) on (a) percentage of germination (b) fresh weight (c) shoot length (d) root length (e) dry weight (f) turgid weight of plantlets at different days after inoculation

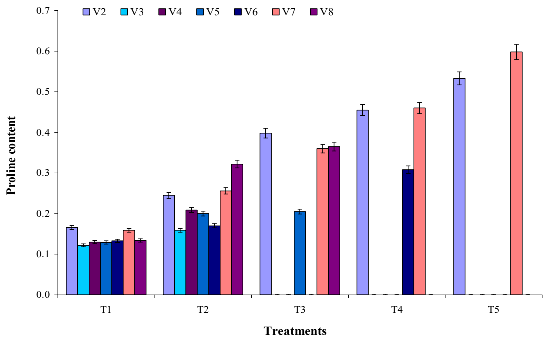
Table 1. Combined effect of varieties and different drought stress on some physiological and biochemical parameters of chickpea varieties.

Treatment combination	Germination (%)	Fresh weight (g)	Shoot length (cm)	Root length (cm)	Dry weight(g)	Turgid weight	Relative water content	Proline content
V_2T_1	100.0^{a}	1.73^{b}	5.00^{ab}	1.75^{b}	0.186^{defg}	1.80^{a}	95.27^{abcd}	0.166hi
V_2T_2	100.0^{a}	1.02^{gh}	3.25^{fg}	1.40c^{d}	0.110^{ij}	0.946^{g}	96.81^{ab}	0.245^{g}
V_2T_3	100.0^{a}	1.36^{d}	1.90^{j}	0.500^{j}	0.220^{cd}	1.24^{d}	93.44^{bcde}	0.398^{d}
V_2T_4	66.67^{b}	0.760^{ij}	2.85^{gh}	0.900^{ghi}	0.220^{cd}	0.810^{h}	91.01^{defg}	0.455^{c}
V_2T_5	66.67^{b}	0.590k^{l}	2.10^{ij}	1.15^{ef}	0.130^{ghij}	0.670^{i}	85.71^{hij}	0.533^{b}
V_3T_1	66.67^{b}	0.810^{i}	3.90^{de}	1.25^{de}	0.120^{hij}	0.960^{g}	82.14^{j}	0.122^{i}
V_3T_2	66.67^{b}	0.950^{h}	2.50^{hi}	1.75^{b}	0.530^{a}	1.04^{ef}	82.35^{j}	0.159^{hi}
V_3T_3	33.33^{c}	1.12^{fg}	1.10^{k}	1.00^{fgh}	0.230^{cd}	1.23^{d}	89.00^{efgh}	0.000^{j}
V_3T_4	0.0000^{d}	0.000^{n}	0.000^{l}	0.000^{k}	0.000^{l}	0.000^{l}	0.000^{q}	0.000^{j}
V_3T_5	0.0000^{d}	0.000^{n}	0.000^{l}	0.000^{k}	0.000^{l}	0.000^{l}	0.000^{q}	0.000^{j}
V_4T_1	66.67^{b}	0.840^{i}	3.83^{de}	0.000^{k}	0.180^{defgh}	0.940^{g}	86.40^{ghij}	0.130^{i}
V_4T_2	66.67^{b}	0.310^{m}	2.00^{j}	1.00^{fgh}	0.226^{cd}	0.890^{g}	12.93^{o}	0.209^{gh}
V_4T_3	0.0000^{d}	0.000^{n}	0.000^{l}	0.000^{k}	0.000^{l}	0.000^{l}	0.000^{q}	0.000^{j}
V_4T_4	33.33^{c}	0.350^{m}	2.00^{j}	0.400^{j}	0.0900^{jk}	0.320^{k}	96.30^{abc}	0.000^{j}
V_4T_5	0.0000^{d}	0.000^{n}	0.000^{l}	0.000^{k}	0.000^{l}	0.000^{l}	0.000^{q}	0.000^{j}
V_5T_1	66.67^{b}	0.660^{jk}	5.25^{a}	0.900^{ghi}	0.106^{ijk}	0.900^{g}	69.33^{l}	0.129^{i}
V_5T_2	66.67^{b}	0.370^{m}	2.90^{gh}	0.900^{ghi}	0.376^{b}	0.540^{j}	5.88^{p}	0.200^{gh}
V_5T_3	66.67^{b}	0.420^{m}	2.75^{h}	0.800^{hi}	0.250^{c}	0.650^{i}	42.50^{n}	0.205^{gh}
V_5T_4	0.0000^{d}	0.000^{n}	0.000^{l}	0.000^{k}	0.000^{l}	0.000^{l}	0.000^{q}	0.000^{j}
V_5T_5	0.0000^{d}	0.000^{n}	0.000^{l}	0.000^{k}	0.000^{l}	0.000^{l}	0.000^{q}	0.000^{j}
V_6T_1	66.67^{b}	1.11^{fg}	4.00^{cd}	0.750^{i}	0.050^{kl}	1.62^{b}	67.95^{l}	0.133^{i}
V_6T_2	66.67^{b}	1.29^{de}	2.90^{gh}	1.00^{fgh}	0.130^{ghij}	1.40^{c}	90.89^{defg}	0.170^{hi}
V_6T_3	66.67^{b}	0.530^{l}	1.10^{k}	1.50^{c}	0.0800^{jk}	0.666^{i}	76.68^{k}	0.000^{j}
V_6T_4	33.33^{c}	0.400^{m}	2.80^{h}	0.000^{k}	0.130^{ghij}	0.540^{j}	65.85^{l}	0.308^{f}
V_6T_5	0.0000^{d}	0.000^{n}	0.000^{l}	0.000^{k}	0.000^{l}	0.000^{l}	0.000^{q}	0.000^{j}
V_7T_1	100.0^{a}	1.16^{f}	4.83^{b}	1.10^{efg}	0.160^{efghi}	1.30^{d}	87.25^{fghi}	0.159^{hi}
V_7T_2	100.0^{a}	1.17^{f}	3.75^{de}	1.25^{de}	0.120^{ij}	1.38^{c}	83.33^{ij}	0.256^{g}
V_7T_3	100.0^{a}	1.37^{d}	2.50^{hi}	0.800^{hi}	0.120^{hij}	1.44^{c}	94.68^{abcd}	0.360d^{ef}
V_7T_4	66.67^{b}	0.540^{l}	3.50^{ef}	1.00^{fgh}	0.130^{fghij}	0.560^{j}	95.35^{abcd}	0.460^{c}
V_7T_5	66.67^{b}	0.840^{i}	3.75^{de}	1.00^{fgh}	0.210^{cde}	0.970^{fg}	83.33^{ij}	0.598^{a}
V_8T_1	100.0^{a}	1.62^{c}	4.33^{c}	0.770^{hi}	0.130^{hij}	1.75^{a}	91.58^{cdef}	0.134^{i}
V_8T_2	100.0^{a}	1.85^{a}	4.40^{c}	1.80^{b}	0.190^{def}	1.81^{a}	98.81^{a}	0.322^{ef}
V_8T_3	66.67^{b}	0.630^{kl}	2.00^{j}	0.500^{j}	0.070^{jk}	1.07^{e}	55.77^{m}	0.365^{de}
V_8T_4	33.33^{c}	1.22e^{f}	4.00^{cd}	3.00^{a}	0.220^{cd}	1.24^{d}	98.04^{ab}	0.000^{j}
V_8T_5	0.0000^{d}	0.000^{n}	0.000^{l}	0.000^{k}	0.0000	0.000^{l}	0.000^{q}	0.000^{j}
$LSD_{0.05}$	4.27	0.103	0.398	0.199	0.051	0.072	4.21	0.05^{l}
Level of significance	**	**	**	**	**	**	**	**
CV (%)	4.75	9.02	10.02	15.29	13.64	5.38	4.28	12.94

** = Significant at 1% level of probability, T_1 = 0g/l, T_2 = 20 g/l, T_3 = 35 g/l, T_4 = 50 g/l, T_5 = 60 g/l



(a)



(b)

(V_2= Binachola-2, V_3 = Binachola-3, V_4 = Binachola-4, V_5 = Binachola-5, V_6 = Binachola-6, V_7 = Binachola-7, V_8=Binachola-8; T_1 = Control, T_2 = 20 g/l, T_3 = 35 g/l, T_4 = 50 g/l, T_5 = 60 g/l of PEG, Error bars show the standard error of the mean)

Figure 2. (a) Relative water content (RWC) (b) Proline content of seven chickpea varieties under different level of drought stress induced by five different concentrations of Polyethylene glycol**.**

(a) MS medium (Control)

(b) MS medium supplemented with 20 g/l of PEG

(c) MS medium supplemented with 35 g/l of PEG

(d) MS medium supplemented with 50 g/l of PEG

(e) MS medium supplemented with 60 g/l of PEG

Plate 1. *In vitro* growth and development of seven chickpea varieties depending on different concentrations of polyethylene glycol (PEG).

Binachola-2 showed 0.533g/100g FW Proline content and 0.598g/100g FW proline were observed in Binachola-7. Proline content 0.000g was recorded in Binachola-3, Binachola-4, Binachola-5, Binachola-6, and Binachola-8 under the influence of severe osmotic stress generated by the highest dose of PEG (60 g/l) (Table 1, Figure 2-b, plate 1). Data from other four treatments (T_1, T_2, T_3 and T_4) revealed that that proline content increased with the increasing level of PEG concentration in case of the varieties that showed significant growth under high level of drought stress and the varieties having very low amount of proline showed retarded growth (Table 1, Figure 2b, plate 1). Review of the literature indicates that a stressful environment results in an overproduction of proline in plants which in turn imparts stress tolerance by maintaining cell turgor or osmotic balance; stabilizing membranes thereby preventing electrolyte leakage; and bringing concentrations of reactive oxygen species (ROS) within normal ranges, thus preventing oxidative burst in plants (Shamsul *et al.*; 2012). From this discussion, it is evident that Binachola-2 and Binachola-7 varieties are potential to survive in drought stress condition.

In this experiment, most of the parameters were negatively affected by the increase of polyethylene glycol levels. It might be due to the drought stress induced by PEG for plant which reduced the germination percentage, fresh weight, shoot length, root length, dry weight, turgid weight and relative water content (RWC). The results from the present study indicated that Binachola-2 and Binachola-7 were highly drought tolerant and these two varieties may be utilized as a selection indicator for breeding program and used as a baseline for improvement of chickpea varieties in Bangladesh. However, this is a preliminary study and needs further confirmation with other varieties of chickpea and also in the drought region of Bangladesh to confirm the results before recommending it to the growers of drought prone area.

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