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Low Doses of Gibberellic Acid can Enhance Germination of Wheat Seed under Drought Stress

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Abstract

Increased drought incidence due to global warming has been affecting agriculture and wheat production in particular. Insufficient water during germination stage is one of the main constraints for crops which cause reduction in seedling emergence and establishment. At this point of view, this study was carried out to investigate the effect of priming with gibberellic acid GA_3 on wheat seed germination (Slemani-2 cultivar) under drought stress. The experimental design was completely randomized design with three GA_3 concentrations (0, 100 and 200 ppm), and four stress levels (0, -0.35, -0.70 and -1.40 MPa) of polyethylene glycol PEG 6000 with three replications. The response to drought and priming effect was measured based on some germination (germination rate, mean germination time and germination index) and growth (seedling vigor I and II, radicle and plumule; length, fresh and dry weight) parameters. The results show that increasing PEG concentrations affects most of the parameters. The low dose of GA_3 pre-treatments had significant effects on the germination and growth parameters of Slemani-2 cultivar, the best results were obtained from the 0 and 100 ppm GA_3, under 0 and -0.35 MPa PEG applications.

Keywords: Gibberellic acid GA_3; Drought stress; Polyethylene glycol PEG 6000; Seed germination

Introduction

Wheat *(Triticum aestivum L.)* is a main source of food for approximately one third of the world. It is one of the most important cereal crops worldwide with global production of 767 million tonnes of grain [1]. The demand for wheat is increasing with the increase in world population, however climate change and water scarcity are threatening agriculture especially wheat production which will affect and world food security. Wheat is mainly grown in arid or semi-arid regions of the world under rain-fed conditions facing low moisture stress at seed germination and seedling [2]. Therefore, plant breeders focused on and developed physiological, morphological and adaptive traits in the past decade to reduce the impact of drought [3].

Drought is a natural hazard, affects a significant proportion of the global population, particularly those living in semi-arid and arid regions. It is a climatic event characterized with long period of precipitation shortage for an area compare to normal average precipitation which causes water shortage. It occurs in most environmental zones of the world, but its great impact is in arid and semi-arid regions [4,5].

Seed priming is a cheap approach to cope with the negative effects of abiotic stresses at early growth and developmental stages of crops [6]. Gibberellic Acid (GA) or Gibberellins is a plant hormone stimulating plant growth and development. One of its functions is stimulating seed germination and grain development along with an interaction of different environmental factors; light, temperature and water [7]. It has two effects, increasing the growth potential of embryo and inducing hydrolytic enzymes.

The aim of this study was to investigate the effect of priming with different concentrations of gibberellic acid (GA_3) on wheat seed under drought stress during the germination and seedling establishment.

Materials and Methods

In order to evaluate the effect of seed priming on the germination and seedlings growth traits of wheat variety Slemani-2 under different drought stress, a factorial experiment was conducted in completely randomized design (CRD) with three replications, at the laboratory of Seed Science and Technology in the Department of Field Crops, Faculty of Agriculture, Van Yuzuncu Yil University, Turkey in 2019.

The factors included four levels of polyethylene glycol (PEG) 6000 (0, -0.35, -0.70 and -1.40 MPa) and three gibberellic acid (GA_3) levels (0, 100 and 200 ppm) were used in the experiments. Seeds were rinsed with distilled water, then their surface sterilized in 10% sodium hypochlorite (vol/vol) for 5 minutes, after then, were thoroughly rinsed with sterile deionized water before priming. The seed lots were imbibed within different doses of GA_3 in priming treatments. Treated seed lots with GA_3 were kept in darkness in an incubator at 25 ± 0.5°C for 8h, and unprimed seeds were used as control. For each treatment, 20 seeds were placed in 9 cm diameter sterile petri dishes with filter paper and 2 ml of PEG solution was added to all petri except for control treatments only (2 ml) distilled water were given. The petri dishes were placed in a totally dark incubator; temperature and relative humidity were 20°C and 20% respectively during the experimentation period.

Data computing and analysis

Germinated seeds were counted daily and observed at the specified time for 10 consecutive days then all parameters were measured and recorded. Seeds with at least 2 mm radicle length were considered germinated.

Germination rate, germination power, germination index and mean germination were calculated with the following formulas.

Germination rate (GR) was calculated using Equation 1 [8].

GR=Total seeds germinated after day 14/Total number of planted seeds

Germination index (GI) was calculated using Equation 2 [9].

$$GI = \Sigma\left(\frac{Gi}{Tt}\right)$$

GI: Germination index; Gi: Days germinated seed rate; Tt: count day

Mean germination time (MGT) was calculated using the Equation 3 [10].

$$MGT = \Sigma\frac{(fx)}{\Sigma f}$$

f: Number of seeds germinated; x: germination day

The average seedlings plumule and radicle lengths (cm) were measured for three normal seedlings from each replicate after the final count in standard germination test. The plumule length was measured from the point of the attachment of the cotyledon to the tip of the seedling. Similarly, the radicle length was measured from the point of attachment to the tip of the root.

The plumule and radicle and radicle fresh weight (gr) were calculated by weighing all seedlings using electric balance. Then, placed in a hot air oven (70°C for 24 hours) to dry and weighed to calculate plumule and radicle and radicle dry weight (gr) [11].

Seedling Vigor Index I (SVI-I) was computed by multiplying the standard germination with the average sum of plumule length (cm) and radicle length (cm) on the 10th day of germination [12].

SVI-I=Germination (%) × Seedling length (plumule and radicle length)

Seedling Vigor Index II (SVI-II) was computed by multiplying the standard germination with mean seedling dry weight [13].

SVI-II=Germination (%) × Seedling dry weight (gr)

All data were subjected to Analysis of Variance and significance of mean values was tested by least significant difference test (LSD) using COSTAT (version 6.3) software.

Results and Discussion

Osmotic stress during the seed germination has negative effects on seed germination traits. Current findings reveal the effect of PEG (except for germination rate) and GA_3 (except for plumule and radicle dry weight) different concentrations on most of the parameters of wheat seedlings. With the increase of PEG concentration, mean germination time increased, while there was no significant effect on seed germination rate. The interactions of GA_3 and PEG applications were determined as significant statistically on the all parameters except for plumule and radicle dry weight. Low concentrations of growth regulator hormones have much positive effect on wheat seed germination under drought stress [14-16].

The effect of gibberellic acid on the germination rate was significant. The highest germination rate was 96.7% obtained from 0 (control) applications of PEG and GA_3, it is the same LSD group with 0 and100 ppm GA_3 dose applications of other PEG applications. Also, the lowest germination rate value was 58.3% obtained from 200 ppm GA_3 dose in the -0.35 MPa osmotic pressure applications. The effect of different PEG concentrations on germination rate was non-significant. Treatments, different PEG and GA_3 concentration had significantly affected mean germination time. At PEG concentration of 1.40 MPa osmotic pressure, the obtained mean values (2.95 day) were the highest, and the lowest value of mean germination time was 2.36 day at -0.35 MPa osmotic pressure. In terms of gibberellic acid and PEG applications interaction the highest value is 3.53 day. It is the same group with 200 ppm GA_3 application, of different PEG applications (control and 0.70 MPa osmotic pressure). The seeds required longer to germinate with the increase of PEG concentration. According to GA_3 applications, the germination index had the highest value at 0 ppm (control) and the lowest value at 200 ppm GA_3 priming, 8.87% and 2.87% respectively. It is the same LSD group with control and 100 ppm GA_3 applications. For PEG doses, the highest value (6.82%) was obtained from 0.35 MPa osmotic pressure applications, while the lowest value (5.65%) was obtained from -1.40 MPa osmotic pressure applications. But, all doses except for -1.40 MPa are statistically at the same group. The highest mean germination time value was 2.95 days at PEG concentration of 35 mm, while it was 2.36 days at PEG concentration of 15 mm. In terms of interaction of both applications, the highest germination index was obtained as 9.17% and it is the same group with control and 100 ppm applications of GA_3 at the -0.35 MPa, control application of GA_3 at the -0.70 MPa osmotic stress. In this study, it was observed that traits such germination index and mean germination time were affected with the increase of drought stress. These findings are also in agreement with Aydin et al. in their experiment on studying the effect of hormonal priming on seed germination of wheat under drought condition in the lab, they documented that mean germination time increased whereas germination rate and index significantly decreased [17]. Under drought stress, cultivars varied in their performance, and parameters such as germination rate and index might be used as an indicator for drought tolerance at germination stage [18].

Seedling vigor index I and II parameters were affected in the significant level statistically by GA_3, PEG and GA_3 × PEG interaction. The highest values for gibberellic acid applications were (1485.1 and 22.7) obtained from control of GA_3. But for both parameters they are not different in the between with 100 ppm GA_3 applications. Also for PEG means the highest seedling vigor index I value was obtained as 1342.0 from -0.35 MPa PEG dose, it is at the same LSD group with control. The minimum value was recorded as 657.8 from the highest dose of PEG (Table 1). For interaction, the best value (1860.0) was found from control of both applications. However, there are no significant differences between with 0 and 100 ppm GA_3 applications at the -0.35 MPa PEG dose. In related to PEG dose mean values, seedling vigor index II had the higher value (18.8) at -0.35 MPa PEG dose, it is at the same LSD group with control. The lowest value (12.1) was obtained from -1.40 MPa PEG application. For GA_3 × PEG applications interaction, the maximum value was found as 26.8 from control of gibberellic acid at the -0.35 MPa PEG application. It is at the same LSD group with 100 ppm GA_3 at the same PEG application. Seedling vigor I and II decreased with increasing of drought stress. This was also proposed by Thomas et al. when they tested chickpea under salinity stress [19].

The different concentrations of PEG and GA_3 had various effects on the investigated characters, and most of the parameters recorded values reduced with the increase of PEG concentration (Table 2). It was found significant GA_3, PEG applications and GA_3 × PEG interaction on the radicle length. For GA_3, the higher value of the radicle length (8.06 cm) was obtained from 0 ppm (control) GA_3. However, it is the same group with 100 ppm GA_3 application. Also, the lowest value was found (2.98 cm) from 200 ppm GA_3 application. In related to PEG applications the lowest value (4.54 cm) was obtained from 200 ppm GA_3, but there are not different statistically in the between with -1.40 MPa osmotic pressure. In the GA_3 × PEG interaction the highest value was found as 10.5 cm from 0 ppm GA_3 application at the -0.35 MPa osmotic stress while the lowest radicle length value was determined as 1.42 cm from 200 ppm GA_3, at the control applications of PEG. According to gibberellic acid applications, it is presented that the average radicle length was (7.71 cm) under medium stress level, -0.35 MPa was higher than control this might be due to roots searching for water. Similar results were suggested by Movaghatian and Khorsandi for lower concentrations of priming under drought stress conditions [20]. Furthermore, higher radicle length was also reported for sunflower seedling examined under PEG and NaCl stress [21]. Plumule length value was the higher (7.60 cm) at the control application of GA_3, but it is at the same group with 100 ppm GA_3 application. The lowest value (3.35 cm) was obtained from 200 ppm GA_3 application. For PEG, the highest plumule length (9.29 cm) was recorded from control (PEG) and the lowest value (2.77 cm) was obtained from -1.40 MPa (PEG) osmotic stress application. Also, for GA_3 × PEG interaction the highest value (12.1 cm) was obtained from 100 ppm GA_3 at the control application of PEG, while the lowest value (2.0 cm) was determined from 200 ppm GA_3 at the -1.40 MPa osmotic stress application. As in Kizilgeci et al. research, the data analysis reveals reduction in plumule length, fresh and dry weight of seedlings with the increase of PEG concentration [22]. The higher value of radicle fresh weight at the GA_3 applications was (0.39 gr) recorded from 0 ppm GA_3 application, but there are not different between with 100 ppm GA_3. The lowest value (0.06 gr) was determined from 200 ppm GA_3. At PEG concentrations, the highest radicle fresh weight (0.325 gr) was obtained from -0.70 MPa PEG and also, the lowest value (0.170 gr) were determined from control of PEG application. In term of interaction the highest (0.477 gr) was obtained from control of gibberellic acid application at the -0.70 MPa of PEG doses. Plumule fresh weights were significantly affected from both applications. The maximum values were recorded as 0.57 and 0.56 from control and 100 ppm of GA_3 applications. The minimum value was obtained from 200 ppm GA_3. However, the higher PEG mean value was (0.688 gr) obtained from control of PEG, it is at the same group with -.0.35 MPa. The lowest value (0.250 gr) was determined from the most PEG doses. In this study, at the GA_3 × PEG interaction the most plumule fresh weights were obtained as 0.848 and 0.782 gr from 0 and 100 ppm GA_3, at the control of PEG applications, respectively.

There were not any significant differences in the radicle and plumule dry weight of the GA_3 and GA_3 × PEG interaction. However, it was found significant effect of PEG doses on the radicle and plumule dry weight. The higher value for radicle dry weight was (0.093 gr) obtained from -0.35 MPa PEG application. Nevertheless, it is at the same group with other applications except for the control. The lowest value (0.062 gr) was determined from control of PEG. Analysis of variance in the present study showed that average radicle dry weight increased at PEG -0.35 MPa (0.093 gr), -0.70 MPa (0.089 gr), and -1.40 MPa (0.081 gr), as compare to control (0.062 gr). This is the agreement with a previous research conducted by Gahtyari et al., They studied the effect of osmotic stress on wheat germination, and they reported that dry weight increases with the increase of stress level [23-25]. The higher value for plumule dry weight was (0.112 gr) obtained from -0.70 MPa PEG application. However, it is at the same group with -0.35 MPa PEG applications. The lowest value (0.058 gr) was determined from -1.40 MPa applications of PEG.

Applications		Germination rate (%)	Mean germination time (day)	Germination index (%)	Seedling vigor index I	Seedling vigor index II
PEG Doses	GA_3					
Control	GA0 (control)	96.7a	2.05bc	9.34a	1860.9a	22.9ab
	GA100	93.3a	2.47bc	7.74ab	1531.0ab	20.8bc
	GA200	61.7bc	3.18ab	2.64d	349.7ef	4.17e
Mean	-	83.9	2.57ab	6.57a	1247.2a	15.9ab
-0.35 MPa	GA0 (control)	95.0a	1.98c	9.22a	1852.4a	26.8a
	GA100	96.6a	2.35bc	8.53a	1816.4a	25.5a
	GA200	58.3c	2.75b	2.72d	357.1ef	4.18e
Mean	-	83.3	2.36b	6.82a	1342.0a	18.8a
-0.70 MPa	GA0 (control)	95.0a	2.02c	9.17a	1372.2b	23.2ab
	GA100	90.0a	2.48bc	6.98b	1152.0c	18.3c
	GA200	63.3bc	3.03ab	2.97d	551.8e	5.12e
Mean	-	82.8	2.51ab	6.37ab	1025.3b	15.6b
-1.40 MPa	GA0 (control)	90.0a	2.25bc	7.76ab	854.7d	17.8c
	GA100	90.0a	3.07ab	6.05c	782.4d	13.8d

	GA200	73.3ab	3.53a	3.15d	336.27f	4.73e
Mean	-	84.4	2.95a	5.65b	657.8c	12.1c
GA_3 Doses	GA0 (control)	94.2a	2.08	8.87a	1485.1a	22.7a
Mean	GA100	92.5a	2.59	7.33a	1320.5a	19.6a
	GA200	64.2b	3.12	2.87b	398.7b	4.55b
CV (%)	-	10.5	15.7	9.6	13.5	13.9
*There is no significant difference between the means indicated with same letter (5%)						

Table 1: Effect of gibberellic acid application on wheat seed germination drought stress.

Applications		**Radicle length (cm)**	**Plumule length (cm)**	**Radicle fresh weight (gm)**	**Plumule fresh weight (gm)**	**Radicle dry weight (gm)**	**Plumule dry weight (gm)**
PEG Doses	**GA_3**						
Control	GA0 (control)	7.87bc	11.3ab	0.294bcd	0.782a	0.101	0.136
	GA100	4.32de	12.1a	0.202de	0.848a	0.078	0.144
	GA200	1.42f	4.44de	0.014f	0.432b	0.006	0.061
Mean	-	4.54c	9.29a	0.170c	0.688a	0.062b	0.114a
-0.35 MPa	GA0 (control)	10.5a	9.01b	0.376b	0.759a	0.146	0.136
	GA100	9.71ab	9.08b	0.301bc	0.756a	0.121	0.143
	GA200	2.92ef	3.23ef	0.026f	0.277cd	0.012	0.056
Mean	-	7.71a	7.11b	0.234b	0.597a	0.093a	0.112a
-0.70 MPa	GA0 (control)	7.66c	6.79c	0.477a	0.446b	0.124	0.12
	GA100	6.84c	5.96cd	0.361b	0.383bc	0.115	0.088
	GA200	5.02d	3.72e	0.137e	0.230d	0.028	0.053
Mean	-	6.51b	5.49c	0.325a	0.353b	0.089a	0.087b
-1.40 MPa	GA0 (control)	6.22c	3.24e	0.401b	0.268d	0.124	0.074
	GA100	5.60cd	3.06ef	0.281cd	0.281cd	0.096	0.056
	GA200	2.57f	2.00f	0.061f	0.200d	0.021	0.043
Mean	-	4.79c	2.77d	0.247b	0.250c	0.081a	0.058c
GA_3 Doses	GA0 (control)	8.06a	7.60a	0.39a	0.56a	0.12	0.12
Mean	GA100	6.62a	7.55a	0.29a	0.57a	0.1	0.11
	GA200	2.98b	3.35b	0.06b	0.28b	0.02	0.05
CV (%)	-	13.5	14.5	16	15	15.9	14.5
*There is no significant difference between the means indicated with same letter (5%)							

Table 2: Effect of gibberellic acid application on wheat seedlings under drought stress.

Conclusion

Seed priming with optimal dosage of plant growth regulators have been showed enhancement in crop seed germination and growth characters under abiotic stress conditions. The conducted experiment was to evaluate a number of germination and growth characters of drought stressed wheat *(Triticum aestivum L.)* Slemani-2 cultivar. It was concluded that high PEG concentrations have negative effects on most of the studied parameters. Seed pre-treatment with low GA_3 concentration can improve the pre and post emergence water stress tolerance of Slemani-2 wheat cultivar. The best results were obtained from the 0 and 100 ppm GA_3, under 0 and -0.35 MPa PEG applications.

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