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The Role of Grain Priming and its Duration on Wheat Germination and Seedling Growth

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ABSTRACT

This study was designed to investigate the effect of different grain priming methods on germination and seedling vigour of wheat cultivar (Gemmiza 9) was performed. Grains were primed for 8, 16 and 24 hours in 13 priming media (water, polyethylene glycol 6000 at 2.5, 5, 7.5 %; $CaCl_2$ at 250, 500, 1000 ppm; ascorbic acid (AsA) at 25, 50, 100 ppm; salicylic acid (SA) at 50, 100, 150 ppm) beside dry grain as a control. In general, it was observed that a priming duration of 24 hr gave the highest values of all duration studied. In addition, results indicated that all grain priming in any priming materials significantly increased seedling vigour index (SVI) represented by germination %, length of seedling and fresh and dry weights of seedling. In most cases, SA at 100 ppm was the most effective in this regard. The greatest germination % was obtained due to 7.5 % PEG; 500 ppm $CaCl_2$; 100 ppm AsA and 100 ppm SA. The highest seedling length was obtained due to 2.5 % PEG; 250 ppm $CaCl_2$; 50 ppm AsA and 100 ppm SA. In addition, seedling dry weight was highest due to 7.5 % PEG; 1000 ppm $CaCl_2$; 50 ppm AsA and 100 ppm SA. The highest SVI values were obtained due to 2.5 % PEG; 250 ppm $CaCl_2$; 50 ppm AsA and 100 ppm SA. These results indicate that priming of grains had a significant effect to promote the germination and seedlings growth of wheat.

Keywords: Ascorbic acid, calcium chloride, polyethylene glycol 6000, priming, salicylic acid, seedling vigor, wheat.

INTRODUCTION

Wheat (*Triticum aestivum* L.) is one of the majority staples food crops globally, it accounts for nearly twenty percent of the human foodstuff requirement and it cultivated on approximately 2.15 million hectares globally (CGIAR, 2014). According to the estimates of the International Food Policy Research Institute, the world's wheat demand (approximately 552 million tons in 2013) will be 40 percent higher in 2020. However, achievable resources to produce this amount of wheat demand will be much lesser at that time. Thus, it is predicted that the wheat price will achieve much higher rates than the current 150 dollars per ton and purchasing from the world markets will not be possible with the current rates (CGIAR, 2014). Inappropriate seedbed preparation, lower seed quality and inappropriate sowing (Van Oosterom *et al*., 1996), insufficient soil moisture, undesirable soil conditions (Lee *et al*., 1998); small seed-zone water potential, deep cultivation depth and soil crusting resulted from precipitation and earlier than the seedling emergence was amongst the serious problems restriction wheat stand and productivity (Giri and Schillinger, 2003). Seed priming is one of vital seed enhancement has been engaged successfully for several crops. The major purpose of this practice is to accelerate the germination and seedling growth under normal and stress conditions (Khafagy *et al*., 2017; Khan *et al*., 2017).

One of the easiest methods that can enhance seedling establishment and therefore crop performance is seed priming. Priming strategy decreased the time between sowing, enhance germination homogeny and seedling growth. Priming led to some physiological modifications are gained into target grain with utilize of natural and synthetic compounds before germination. Priming is a well-established treatment for enhancing seed quality throughout the transient activation of the pre-germinative metabolism that involved antioxidant function, DNA replication responses and greater ATP availability, induced biosynthesis of proteins and the repairing of cellular bio membranes (Chen and Arora, 2013; Hussain *et al*., 2015); induces osmotic adjustments (Bradford, 1986) and membrane re-organization throughout, restoring their original structures, decreasing electrolytes leakage (Fujikura and Karssen, 1995). It can furthermore improve antioxidant enzymes (Roqueiro *et al*., 2012). It optimizes seed performance with fast and homogeneous germination, healthy and vigorous seedling growth, accomplishment a physiological condition leading to faster and better germination and the emergence of different crops (Yücel and Heybet, 2016; Vaz Mondo *et al*., 2016; Patel *et al*., 2017; Shah *et al*., 2017; Khafagy *et al*., 2017; Khan *et al*., 2017). Generally, priming technique is a pre-soaking seed treatment in water and dried back to storage water contents pending additional use. Ghassemi-Golezani *et al*. (2008) verified that encouraging impacts of priming technique on seed invigoration associated with priming duration.

Seed priming can be carried out using different techniques viz., Hydro-priming (soaking in water), Halo-priming (soaking in salt solutions), Osmo-priming (soaking in osmotic solutions such as PEG), bio-stimulants inducers (SA and AsA) and solid-matrix priming (El-Saidy Aml *et al*., 2011; Farouk and El-Saidy Aml, 2013; Rehman *et al*., 2015; Ruttanaruangboworn *et al*., 2016; Vaz Mondo *et al*., 2016; Shah *et al*., 2017; Khafagy *et al*., 2017; Khan *et al*., 2017).

Hydro-priming is the simplest method to hydrate seeds and decrease the extra application of the chemical. Farouk and El-Saidy Aml, 2013; Vaz Mondo *et al*., 2016; Patel *et al*., 2017; Shah *et al*., 2017; Khafagy *et al*., 2017, reported that hydro-priming basically ensured quick and homogeneous germination associated with little irregular seedling %. Hydro-priming in other crops such as corn may increase the germination %, germination characteristics and decrease the mean germination time (Ahmmad *et al*., 2014).

Osmo-priming is the majority widespread kind of priming technique wherein seeds are pre-soaked in small water potential solution like PEG (Ashraf and Foolad, 2005). Osmopriming of wheat grains may enhancement grain germination and emergence and may promote proliferous and vigorous root growth (Rehman *et al*., 2015; Yücel and Heybet, 2016; Shah *et al*., 2017). Khafagy *et al*. (2017) found that priming barley grain in

PEG increased percentage of germination and growth of seedling under normal or draught. Dashtmian *et al*. (2014) and Yücel and Heybet (2016) reported that seed priming with salicylic acid enhanced seedling vigor by increased photosynthesis and biochemical processes under normal or salinity conditions. Halo-priming is soaking seed techniques which improve germination (Shah *et al*., 2017; Khan *et al*., 2017). In this concern, Yücel and Heybet (2016) proved that calcium chloride at 450 mM increased wheat grain vigor under normal or salinity stress.

A wheat farmer or producer should be capable of identifying the most excellent priming solution and duration that is consistently cost effective to meet his objective of early seedling development and increase yield income. Therefore, the aimed of the current investigation was to assess the effect of priming strategy and duration on wheat germination and growth of seedling.

MATERIALS AND METHODS

This study was carried out at the laboratories of Agric. Botany Dept, Fac. of Agric, Mansoura Univ, during the season of 2016. Wheat cultivar Gemmiza 9 was used in the experiment. The experiment comprised of 2 factors, the first factor was priming methods (a) Hydr-opriming, soaking grains in water, (b) Osmo-priming, soaking grains solution of polyethylene glycol (PEG 6000) at 2.5, 5.0 or 7.5% (c) Halo-priming , soaking grains in calcium chloride at 250, 500 and 1000 ppm (d) Vitamin priming, soaking grains in ascorbic acid (AsA) at 25, 50 or 100 ppm and (e) Hormone priming , soaking grains in salicylic acid at 50, 100 or 150 ppm. Priming durations as the second factor (8, 16, and 24 hr.). A factorial completely randomized design with 5 replicates was performed. Grains of wheat were secured from Field Crop Res. Inst., Agric. Res. Cent., Egypt. The grains were surface-sterilized for five min. with 1% of sodium hypochlorite, then ethyl alcohol at 96% for 30 sec followed by distilled water three times. The following priming method was adopted in the experiment: a single layer of wheat grain was submerged in every priming solution in the dark at laboratory temperature, the ratio of solution volume to grain weight solution volume was 5:1 (ml/g), after soaking, the grains were rinsed three times in distilled water (El-Saidy Aml *et al*., 2011). The primed grains were surface-dried under compulsory air on filter paper to their original water content.

Five replicates of 15 grains were sown in 15-cm diameter plastic plates. Each plate was supplied with 20 ml distilled water or the test solution. After 8 days from sowing (the end of the experimental) were determined germination %, length of seedling, seedling fresh and dry weight and seedlings vigour index. The seedling vigor index (SVI) was assessed according to the subsequent equation:

Seedling Vigour Index = [length of seedling (cm) × percentage of germination]

The using one-way ANOVA for analyzed data and followed by Duncan's Multiple Range Test using COSTAT software.

RESULTS AND DISCUSSION

Germination %

The data indicated that priming grain wheat and Nacl salinity caused a significant effect on percentage of germination alone or in combinations and different priming treatments and duration (Figure, 1). In general, all treatments markedly increased the germination % except AsA at 25 and 50 ppm for 16 hr that non-significantly decreased germination %. The figure also postulated that in general, priming duration for 24 hr gave the maximum germination % relative to the supplementary priming duration in all priming solution. The maximum germination % was obtained by using 2.5 and 7.5 % PEG; 50 and 100 ppm SA for 24 hr and 100 ppm AsA and 150 ppm SA for 16 hr. The potential cause for an enhancement in germination % by priming for 24 hr may result from larger hydration of colloids superior glueyness and protoplasm elasticity, increased inbound water content, lesser water shortage, and amplified activity of metabolic.

Germination and seedling development are an important stage in the plant life cycle, as they are the majority susceptible to environmental stress. Priming treatment is becoming the familiar and successful practice to a farmer for improving seed germination criteria performance on large number of crops (Harris *et al*., 2002; Rashid *et al*., 2004). These results corroborate with those of Vaz Mondo *et al.* (2016); Patel *et al*., (2017) for hydropriming; as well as Khafagy *et al.* (2017) for PEG; El-Saidy Aml *et al.* (2011) for AsA; Yücel and Heybet (2016) for SA and Yücel and Heybet (2016); Shah *et al.* (2017) and Khan *et al.* (2017) for Ca+2, that they establish that germination invogration were considerably improved by seed priming strategies. Furthermore, seedlings from primed seeds are recognized to appear extra rapidly and produce additional vigorously that those from un-primed ones (Arif *et al*., 2005)

Several metabolic changes in the lag phase of germination was come during priming of seed which was crucial for germination, i.e. dormancy breaking, hydrolysis or inhibitors of metabolism, imbibition and enzyme activities (Ajouri *et al*., 2004). The encouraging outcome of priming possibly resulted from the stimulator impacts of the mediation of cell division in the embryo on the early stage of germination processes (Hassanpouraghdam *et al*., 2009). Additionally, the earlier reports reported the positive impacts of priming-related with early restore and assimilation of nucleic acids, increase protein biosynthesis and repair cellular bio membranes (Moradi Dezfuli *et al*., 2008). Additionally, priming induces the hydrolysis of abscisic acid and cytokinin, coumarin and phenol compounds that can work as a germination inhibitor (Demir and Van DeVenter, 1999). In addition, priming induced premobilization of seed reserves (Job *et al*., 2000) and/or absorption of water (Ghassemi-Golezam *et al*., 2010), enhancement analysis of endosperm by higher hydrolase activities (Bradford *et al*., 2000). Finally, priming enhances the activities of antioxidant enzymes (Wang *et al*., 2003), as well as superior ATP accessibility and energy for quicker grain germination, repair of deteriorated seed parts and declined leakage of metabolism happen throughout the priming treatment that permits seeds to initiate the germination sequence and therefore get better emergence (Farooq *et al*., 2006).

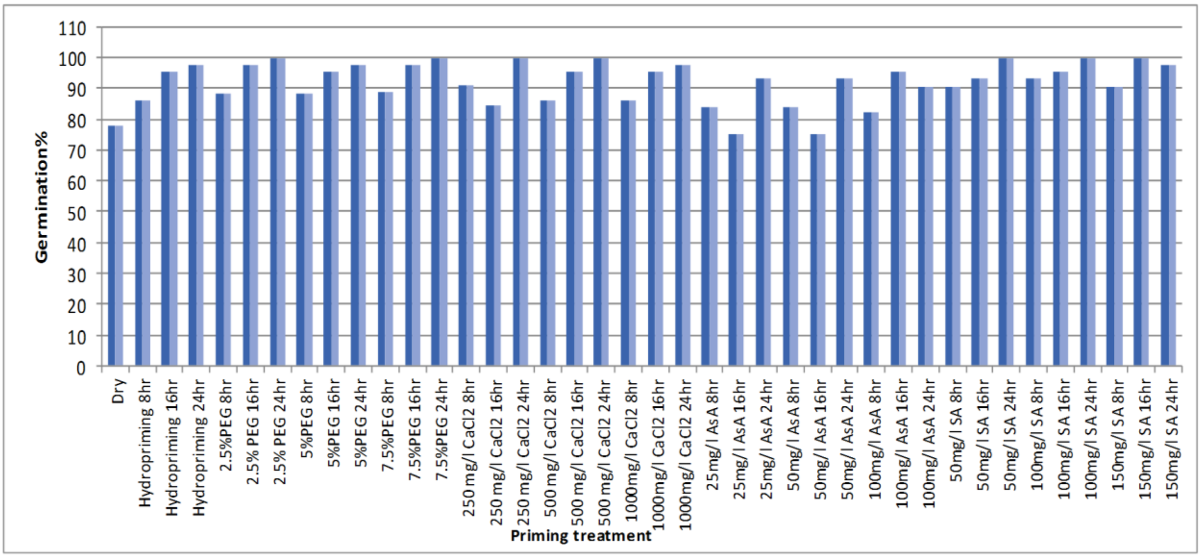

Figure 1. Effect of priming treatments and duration on wheat grain germination percentage at 8 days from sowing.
Data represent the mean± SE of n=5. Significance difference between treatment was tested by one way analysis of variance (ANOVA at P < 0.05 by Duncan's Multiple Range Test

Seed priming technique is attractive well-known to farmers in numerous parts of the globe and has currently been improved there on a variety of crops (Harris *et al*., 2002; Rashid *et al*., 2004). The responses of plants to priming duration are differing by the priming agent and crop cultivars. In this concern, Giri and Schillinger (2003) verified that wheat grain hydroprimed for 12 hr acted alike or even superior than other priming treatments. Moradi Dezfuli *et al.* (2008) observed that the greatest maize germination % obtained when grains water primed for 36 hr. Correspondingly, Basra *et al.* (2002) recognized that grains of wheat responded to diverse soaking seed with hydropriming for a 48 hr performance the highest invigoration followed by 24 hr. Additionally, Yari *et al.* (2011) indicate that all grains priming methods enhanced rice germination and the maximum germination and the most excellent speed of germination were reported on the priming of 48 hr duration. Moreover, Ghassemi-Golezani *et al.* (2008) observed that helpful impacts of seed priming on seed invigoration depended on priming duration and observed that chickpea seeds hydropriming for 8, 16, and 48 hr improved seedling emergence, but the best improvement was obtained with 16 hr priming duration.

Seedling Length

Seedling length of wheat genotype was significantly affected by different priming solutions or duration (Figure, 2). Generally, seedling length increased significantly by application of priming materials under all priming duration in special for 24 hr. In general, the result revealed that the highest seedling length was obtained by 24 hr priming durations. The maximum seedling length (24 cm) was recorded with 2.5 % PEG concentration for 24 hr, whereas the minimum shoot length (4.3 cm) was recorded with control treatment (non-primed seed), the second most effective in increasing seedling was 100 ppm SA for 24 hr.

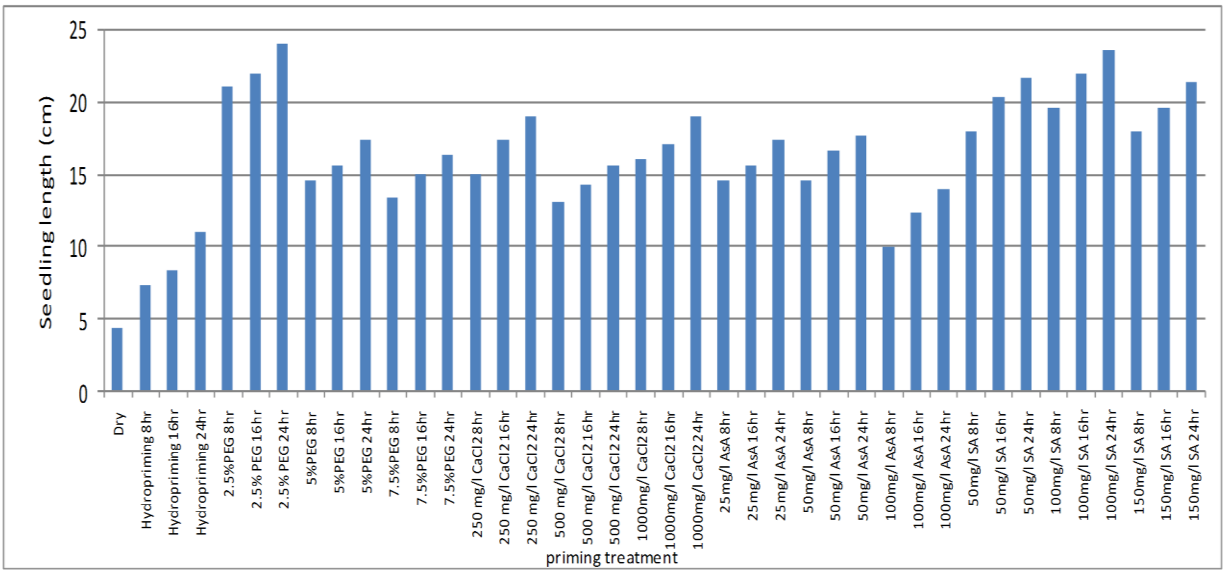

Figure 2. Effect of priming treatments and duration on 8 days wheat seedling length
Data represent the mean± SE of n=5. Significance difference between treatment was tested by one way analysis of variance (ANOVA at P < 0.05 by Duncan's Multiple Range Test.

This result is agreeing with Vaz Mondo *et al.* (2016); Patel *et al.* (2017); Yücel and Heybet (2016); Shah *et al.* (2017) and Khafagy *et al.* (2017). Findings of Rennick and Tiernan (1978) found that a rapid and more extended elongation of coleoptile occurred in treating seeds than non-treated and over pre-sowing seeds. Lee and Kim (2000) reported that priming increased the metabolic activities of seed ultimately gained the substantial shoot length than unprimed seed. Baque *et al.* (2016) revealed that primed wheat with 10 % PEG solution gave the highest shoot length. The increase in length of seedling might result from the motivation of cell division and enlargement (Golizadeh *et al.*, 2015).

Seedling fresh and Dry Weight

The data in Figure (3) revealed that the application of any priming materials for a different duration significantly increased seedling fresh and dry weights related to unprimed dry grains. In general, the result revealed that the maximum seedling fresh and dry weights (0.497 and 0.086 g, respectively) were scored with 100 ppm SA for 24 hr, whereas the minimum fresh and dry weight of shoot (0.077 and 0.011 g, respectively) was recorded with control treatment (non-primed seed). This result was also supported by Ghassemi-Golezani *et al.* (2008); Yücel and Heybet (2016); Shah *et al.* (2017); Khafagy *et al.* (2017). Also, Baque *et al.* (2016) reported that highest dry weight of shoot was recorded in the seed primed with 10 % PEG solution compared to that of osmo and hydro primed seed.

Perhaps priming was repair of damaged to membrane resulted from weakening and exerted enhanced germination model and superior vigor level than unprimed (Ruan *et al.*, 2002). The improvement in germination might be resulted from maintain transportation of reserved food substances, trigger, and resynthesis of enzymes, and biosynthesis of DNA and RNA establish throughout priming. The promotive outcome of calcium chloride on increasing seedling growth could be due to a promotive effect on cotyledon reserve mobilization (Franco *et al.*, 1999).

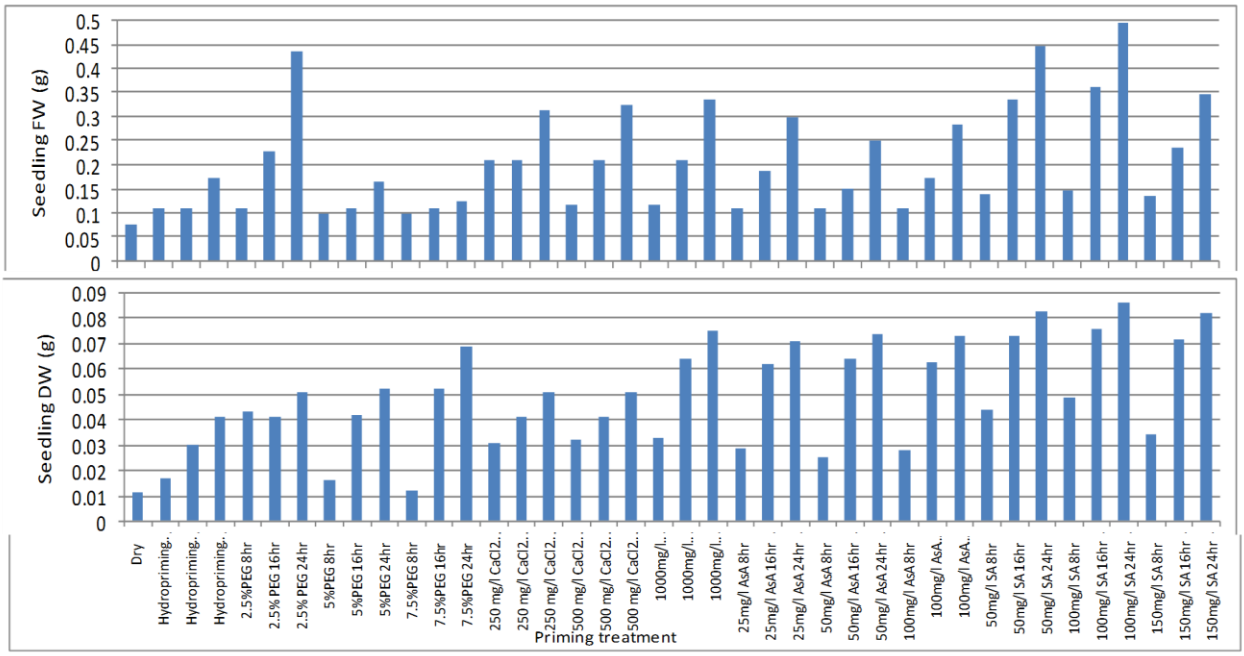

Figure 3. Effect of priming treatments and duration on 8 days wheat seedling fresh and dry weights.

Data represent the mean± SE of n=5. Significance difference between treatment was tested by one way analysis of variance (ANOVA at $P < 0.05$ by Duncan's Multiple Range Test.

The favorable influence of hydropriming has been ascribed to the enlistment activities of enzyme requisite for quick germination and compounds like proteins, soluble sugars and free amino acids from storage organs (Ashraf and Foolad, 2005). The better seedling fresh and dry weight may be associated with the most favorable accessibility of nutrients to seedling after emergence that improved the seedling growth and therefore enhanced wheat fresh and dry weights in the nutrient primed seed treatments (Yari *et al.*, 2010).

Supplementation of ascorbic acid may be influenced on a series of varied metabolic processes in plants, i.e., seed germination, and cellular membrane permeability. One of the noteworthy functions of ascorbic acid on germination of seed and cellular differentiation is activity of its antioxidant than its possible efficacy as an organic material for respiratory metabolism energy. Furthermore, ascorbic acid could be concerned directly or by maintaining the favorable cellular redox balance in the procedure accountable for depiction the reserve food substances accessible for the germinating embryos. Our investigation has reported that ascorbic acid raised the root and shoot length in primed grains. In this regard, Wahid *et al.* (2008) observed that priming sunflower seeds in ascorbic acid enhances vigor; however, priming treatment may differ seriously in their efficiency. Additionally, presoaking seeds in ascorbic acid improve seedling growth by the direct impact's metabolism of vitamins, act as co-enzymes, constituents of enzyme cofactor and/or functions as hormone precursors (Oertli, 1987), antioxidative properties and possible as well yet unidentified modes of actions (Bayer and Schmidt, 1991). Moreover, ascorbic acid implicated the regulation of cell division, cell elongation and cell wall expansion (El-Yazal, 2007).

Seedling Vigor Index (SVI)

Figure (4) postulated that priming grain in water, polyethylene glycol, calcium chloride, ascorbic acid, salicylic acid concentrations significantly increased SVI as compared with unprimed dry grains. The maximum vigor index (24) was show cause when the grain primed with 2.5 % PEG solution for 24 hr and the minimum vigor index (3.33) was achieved when the seeds were not primed.

Seed vigor is the sum of the performance and activity lots of seed of suitable germination in a broad assortment of mediums (ISTA, 2011). SVI is not an only assessable immovable, nevertheless is an impression describing numerous trials connected with the subsequent aspects of seed lot performance rate and germination and seedling growth homogeny, emergence capability of seeds under adverse environmental circumstances (ISTA, 2011). The promotive effect of priming strategy on increasing SVI was also with the resultes of Khafagy *et al*. (2017); Patel *et al*. (2017) who found that seed priming increases seedling vigor of several plants. Probably, the improvement in soybean germination and vigor may be activation and resynthesis some enzymes, mobilization of food material, synthesis of DNA and RNA begin during osmotic priming (Sadeghi *et al*. 2011). Baque *et al*. (2016) found that maximum vigor index was recorded when primed seed with 10 % PEG. Osmo-priming in 20 % PEG-8000 was increased the vigor index Italian ryegrass and sorghum grains.

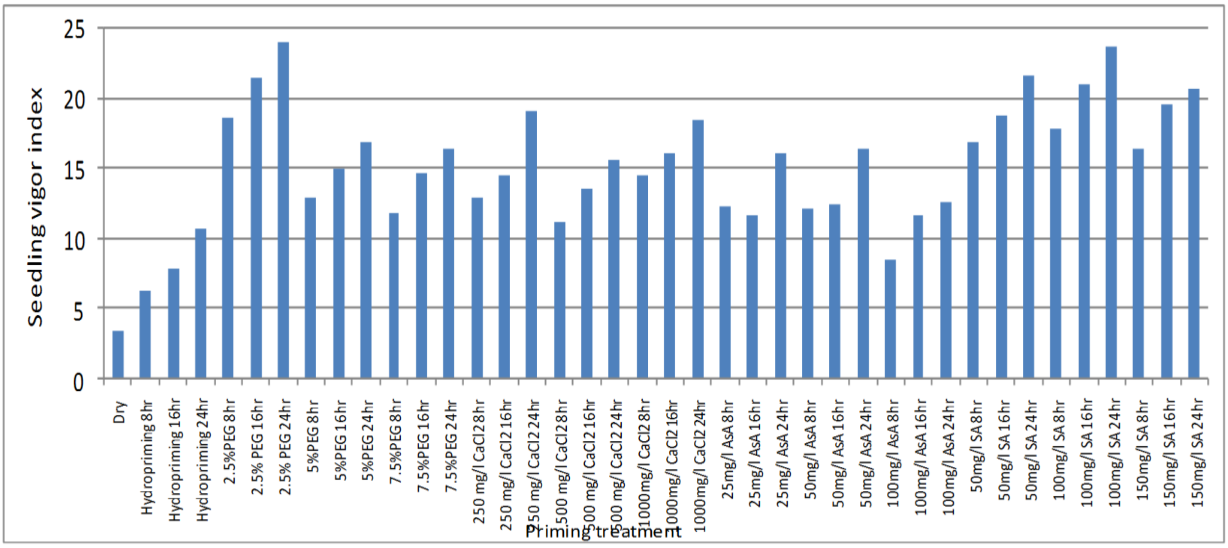

Figure 4. Effect of priming treatments and duration on 8 days wheat seedling vigor index

Data represent the mean± SE of n=5. Significance difference between treatment was tested by one way analysis of variance (ANOVA at P < 0.05 by Duncan's Multiple Range Test.

CONCLUSION

It can be concluded that the priming technique is a useful to advance wheat grains germination and stimulate growth of seedling. The current investigation has provided positive preliminary data on the impacts of priming methods with some chemicals before germination and seedling growth trails of wheat plants.

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دور معاملات تهيئة الحبوب، وفترات النقع عل إنبات ونمو بادرات القمح

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أجريت تجربة معملية بهدف تقييم أثر استخدام طرق مختلفة من معاملات تهيئة الحبوب علي إنبات وقوة بادرات القمح صنف جميزة 9. تم نقع الحبوب لفترات زمنية (8، 16، و 24 ساعة) في 13 مادة من مواد التهيئة (ماء & بولي إيثلين جليكول 6000 بتركيزات 2.5، 5 و 7.5% & كلوريد الكالسيوم بتركيزات 250، 500 و 1000 ملليجرام/لتر & حامض الأسكوربيك بتركيزات 25، 50 و 100 ملليجرام/لتر & حامض السالسيلك بتركيزات 50، 100 و 150 ملليجرام/لتر) بالإضافة للحبوب الجافة (كنترول). بصفة عامة، أعطت معاملات التهيئة لمدة 24 ساعة أعلي القيم مقارنة بفترات النقع الأخري. بالإضافة إلي ذلك، أوضحت النتائج أن جميع معاملات تهيئة الحبوب زودت معنوياً دليل قوة البادرات (SVI) متمثلاً في نسبة الإنبات، طول البادرات، الوزن الطازج والجاف للبادرات. في معظم الأحوال، كان السالسيلك بتركيز 100 ملليجرام/لتر هو الأكثر فاعلية في هذا السياق. أقصي نسبة إنبات تم الحصول عليها نتيجة المعاملة بـالبولي إيثيلين جليكول بتركيز7.5%، كلوريد الكالسيوم بتركيز 500 ملليجرام/لتر، حامض الأسكوربيك بتركيز100 ملليجرام/لتر، حامض السالسليك بتركيز100 ملليجرام/لتر. أقصي طول للبادرات تم الحصول علية بإستعمال 2.5% بولي إيثلين جليكول، 250 مليجرام/لتر كلوريد كالسيوم، 50 مليجرام/لتر حامض الأسكوربيك، 100 ملليجرام/لتر حامض السالسيلك. أقصي وزن جاف للبادرات تم الحصول علية بإستعمال 7.5% بولي إيثلين جليكول، 1000 ملليجرام /لتر كلوريد كالسيوم، 50 ملليجرام/ لتر حامض الأسكوربيك، 100 ملليجرام/لتر حامض السالسيلك. أما أقصي دليل لقوة البادرات فقد تم الحصول علية بإستعمال 2.5% بولي إيثلين جليكول، 250 ملليجرام/لتر كلوريد كالسيوم، 50 مليجرام/لتر حامض الأسكوربيك، 100 ملليجرام /لتر حامض السالسيلك. تلك النتائج تُبين أن معاملات تهيئة الحبوب أثرت معنوياً بزيادة إنبات ونمو بادرات القمح.