

Full Length Research Paper

Effect of osmotic potential of activator solution and temperature on viability and vigour of wheat seed

Michael P. Fuller^{1*} and Jalal H. Hamza^{1,2}

¹School of Biomedical and Biological Sciences, University of Plymouth, PL4 8AA, UK.

²Department of Agronomy, College of Agriculture, University of Baghdad, Iraq.

Accepted 24 May, 2013

Experiment was conducted to investigate if seed priming with polyethylene glycol (PEG) activator solutions affects the viability and vigour of deteriorating wheat (*Triticum aestivum* L. cv. Caxton) seed lot. Seeds were subjected to PEG priming solutions with varying osmotic potentials (-1, -2, -3 MPa) at temperatures of 15 or 20°C for 6, 12, 18 or 24 h and compared to the non-primed control. Highest germination percentage at first and final count, length of plumule and dry weight of seedling were all associated with Treatment 1 MPa/20°C/6 h (92%, 94%, 9.2 cm, 0.0133 mg, respectively) compared to the control (82.5%, 86%, 7.8 cm, 0.0112 mg, respectively). The best values of coefficient of velocity of germination (CVG), mean germination time (MGT) and germination rate index (GRI) were associated with Treatment 2 MPa/15°C/24 h. There were significant interactions between the factors under study and whilst most positive effects decreased with incubation time the opposite was true at 15°C Treatment 2 MPa where an initial decrease in germination after 6 h was restored with longer incubation times. Significant correlations were found between most of the characteristics under study although these did not always account for a high percentage of variation but CVG and MGT were very highly correlated. It was concluded that, 6 h in Treatment 1 MPa PEG at a temperature of 20°C resulted in significantly improved germination percentage whilst 24 h Treatment 2 MPa at 15°C was optimal for the highest CVG and MGT. The highest speed of germination was not associated with the highest germination percentage.

Key words: Osmotic potential, priming, soaking, temperature, *Triticum aestivum* L., wheat.

INTRODUCTION

Seed priming or activation, can be applied to seed lots either with weak germination potential or seed lots which demonstrate irregular germination patterns. Seed wheat typically has good germination capacity but under some agricultural production systems, such as in the Middle East, the lack of good storage facilities and the need to use 2 year old or more seed lots frequently means that wheat shows variable field germination and establishment. The principle in all priming is the treatment of seed in a controlled manner using water with or without amendments followed by drying back. Primed seed often

demonstrates greater subsequent field germination. Seed priming has been found to be a useful technology to enhance rapid and uniform emergence, and to achieve high vigour and better yields in vegetables and floriculture (Bruggink et al., 1999) and some field crops (Chiu et al., 2002; Giri and Schillinger, 2003; Basra et al., 2005; Farooq et al., 2006). Heydecker et al. (1973) defined the activation of seed as seed treatment with osmotic solutions, allowing the seeds to imbibe and proceed to the first stage of germination without allowing the radical to break the seed cover prior to drying back.

*Corresponding author. E-mail: mfuller@plymouth.ac.uk. Tel: +44 (0)1752 587635. Fax: +44 (0)1752 587634.

Tilden (1984) commented that, the activation process leads to stiffness and restoration of the plasma membrane thereby diminishing electrolyte release, which leads to the enhancement of germination and seedling vigour. The mechanism of activation appears to accelerate the production of ATP and increase enzyme activity and repair of DNA and RNA (Fu et al., 1988). Subsequent germination rate increase and improved the field emergence appears to be due to restored integrity of cellular membranes and stimulation protein metabolism and increased DNA activity leading to improved effectiveness of antioxidants (Sung and Chang, 1993; Chiu et al., 1995; Hsu and Sung, 1997). Through the process of priming increased the level of enzymes and metabolic processes result in the formation of some simple sugars which can be absorbed immediately by the embryo during early germination (Ismail, 1997).

Suitable periods of priming vary according to the kind of osmoticant chosen, the water potential of the osmotic solution, temperature during priming and species of plant (Sadeghi et al., 2009). The priming duration is important because if it becomes too long, the radicle appears and the priming benefits disappear as seeds die on dry back. Leila et al. (2010) primed seed of bread wheat for 12, 24 and 36 h at three temperatures (20, 23 and 28°C) and the highest germination percent and speed of germination were observed with 12 h priming. Also overall 20°C had better effects than other temperatures on germination attributes and seedling parameters. Sharifzadeh et al. (2006) also reported that, vigour, radicle and stem length in wheat seedlings were significantly higher at 20°C than 10°C. The factors that are subject to the control in the activation process are the quantity and rate of absorption of water, temperature and duration of the entire process. The work reported here investigated whether activation of the seed affect viability and vigour in a wheat.

MATERIALS AND METHODS

The experiments were conducted on a deteriorating 3 year old sample of winter wheat (cv. Caxton) with 85% germination potential. Seeds (250 g) for each treatment were immersed in 300 ml of polyethylene glycol (PEG) amended to distilled water having varying osmotic potential (-1, -2, and -3 MPa) in plastic containers covered with caps to prevent evaporation loss. The treatment solutions were prepared by dissolving 21.9, 28.2, and 33.4 g of PEG6000 in 100 ml distilled water to give the desired osmotic potentials. A digital OSMOMET device was used to confirm the osmotic potential of the solutions. Seeds were fully immersed in priming solutions and incubated at 15 or 20°C ±1 for 6, 12, 18, and 24 h under dark conditions. There were therefore 25 treatments (3 osmoticums × 2 temperatures × 4 durations) and non primed control.

After priming seeds were rinsed 3 times in the distilled water, it was then dried to their original moisture level (circa 12%) by spreading the seeds out on a tray in an incubator at 20 ±1°C and 50% RH for 24 h. A digital Protimeter was used to measure and monitor the moisture content during the drying period.

50 seeds from each priming treatment together with a control (unprimed) were then germinated in 140 mm Petri dishes on two

layers of Whatman Number 1 filter paper moistened with 40 ml of distilled water. The Petri dishes were placed in an incubator at a temperature of 20 ± 1°C in the dark (AOSA, 1988; ISTA, 1993). Each treatment was replicated 4 times and Petri dishes were arranged in a completely randomized design.

Seeds were considered germinated when they exhibited radicle extension > 2 mm. The germination percent was recorded every 24 h up to 8 days. A number of attributes were analyzed

Germination percentage at first count: measured after 4 days from germination. Germination percentage at final count: measured after 8 days from germination. Coefficient of Velocity of germination (CVG): gives an indication of the rapidity of germination increasing when the number of germinated seeds increases and the time required for germination decreases. Theoretically, the highest CVG possible is 100 and would occur if all seeds germinated on the first day. It was calculated using Equation (1) (Kader and Jutzi, 2004). Mean Germination Time (MGT): the lower MGT, the faster a population of seeds has germinated. It was calculated using Equation (2) (Kader, 2005). Germination Rate Index (GRI): reflects the percentage of germination on each day of the germination period. Higher GRI values indicate higher and faster germination. It was calculated using Equation (3) (Kader, 2005).

$$\text{Equation 1: CVG (\% d}^{-1}\text{)} = 100 * \sum Ni / \sum (NiTi)$$

$$\text{Equation 2: MGT (d)} = \sum (NiTi) / \sum Ni$$

$$\text{Equation 3: GRI (\% d}^{-1}\text{)} = \sum (Ni / i)$$

N is the percentage of germinated seed in day i, and Ti is the sequence of day from sowing seed.

At the end of the germination test (8 days) the lengths of plumule and radicles were measured and shoots were cut from the cotyledons and dried in an oven at 75 ± 2°C for 24 h. The dried seedlings weighted to the nearest milligram and the mean seedling dry weight determined.

Data were statistically analyzed as one way analyses of variance using SPSS. Probabilities of significance were used to indicate significance among treatments. Least significant difference (LSD) was used to make comparisons between means of treatments. Calculated coefficients of simple correlation between attributes (Quinn and Keough, 2005) were also studied.

RESULTS

Some of the priming treatments significantly improved the germination capacity over the control at both first count and final count whilst other treatments significantly reduced germination capacity and these were particularly associated with the high osmoticum used (-3 MPa) (Figure 1). CVG, MGT and GRI also showed that some treatments outperformed the control (Figure 2). Length of plumule and seedling dry weight showed that some activator treatments were capable of inducing improvements over the control (Figure 3) indicating that the improved germination also improved seedling performance.

All the attributes studied showed significant responses to the effect of treatments. Highest values of germination percentage at first and final count, length of plumule and dry weight of seedling were associated with Treatment 1Mpa/20°C/6 h (92%, 94%, 9.2 cm, 0.0133 mg), compared with the control which gave 82.5%, 86%, 7.8 cm,

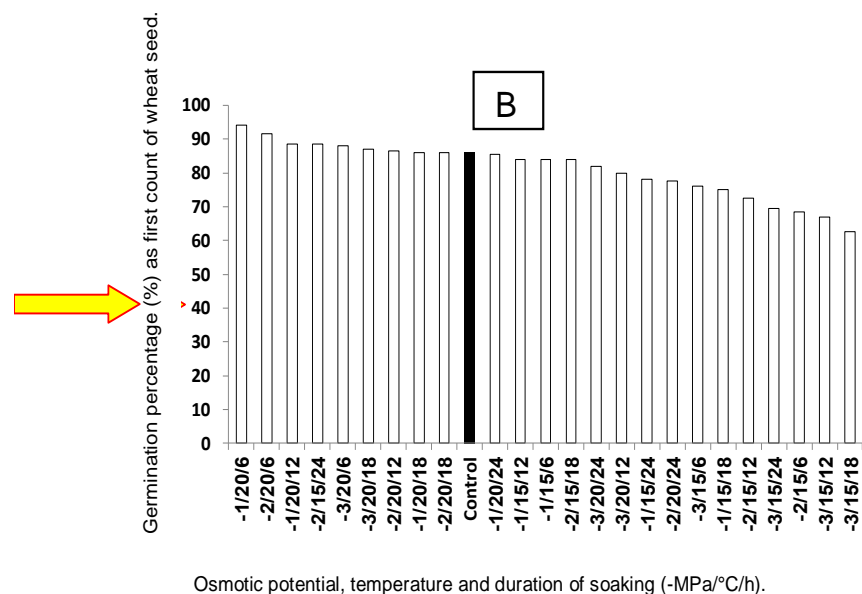
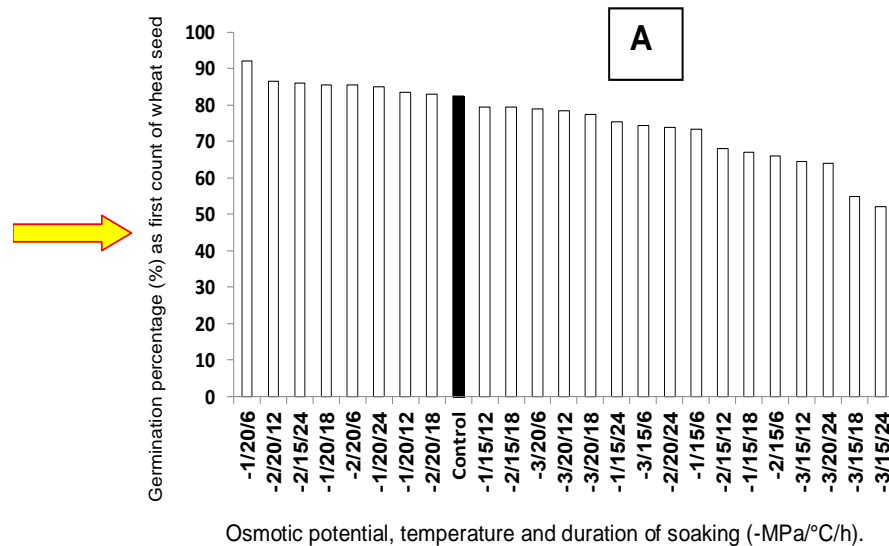


Figure 1. Effect of the osmotic potential of activator solutions and temperature on germination percentage (%) as first count (A) (LSD_{0.05} 7.2) and final count (B) (LSD_{0.05} 8.0).

0.0112 mg, respectively.

Best values of coefficient of velocity of germination, MDT and GRT were associated with Treatments 2MPa/15°C/24 h compared with the control.

There was significant interaction between the concentration of the osmoticum and both the duration and temperature of incubation (Figure 4) where the response for first germination is presented. This complex response pattern was evident for all measured and calculated parameters. Significant correlations were found between most of the characteristics under study and the highest correlation was between (CVG) and MGT indicating that these essentially measure the same parameters (Table 1).

DISCUSSION

Enhancement of viability and vigour of wheat by priming with a PEG osmoticum has been clearly demonstrated in this study. The findings are in agreement with others who also demonstrated the success of priming in wheat (Salehzade et al., 2009; Sowmya et al., 2012). The best response was found to be associated with Treatment 1 MPa incubated at 20°C for a short period of time (6 h). This agrees with Sharifzadeh et al. (2006) who also reported that vigour, radicle and stem length in wheat seedling were significantly higher at 20°C than 10°C.

Priming duration is important because if it is too extended then the radicle begins to emerge and priming

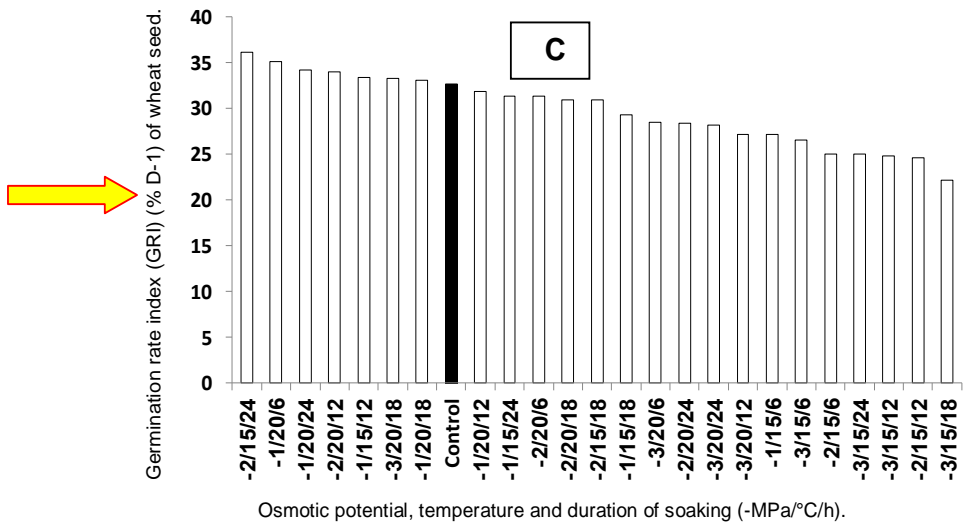
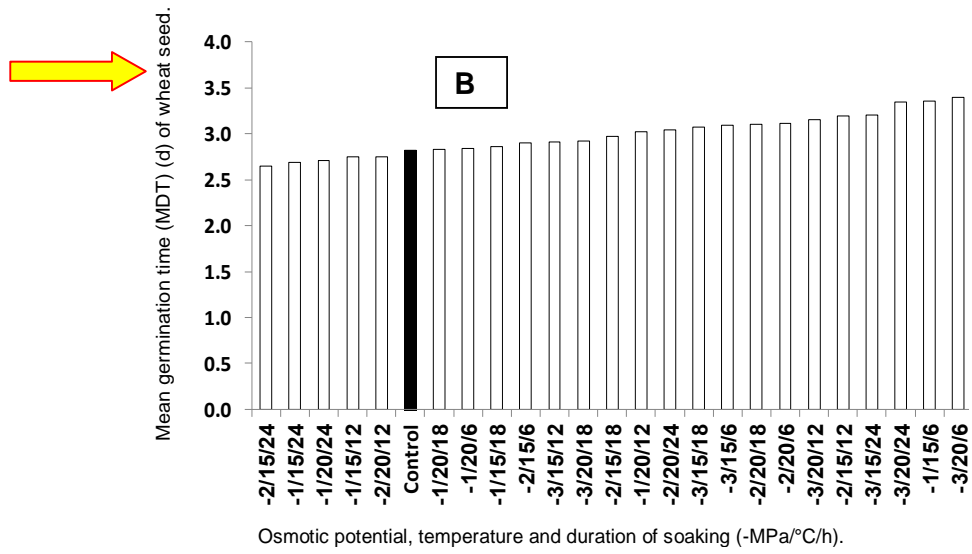
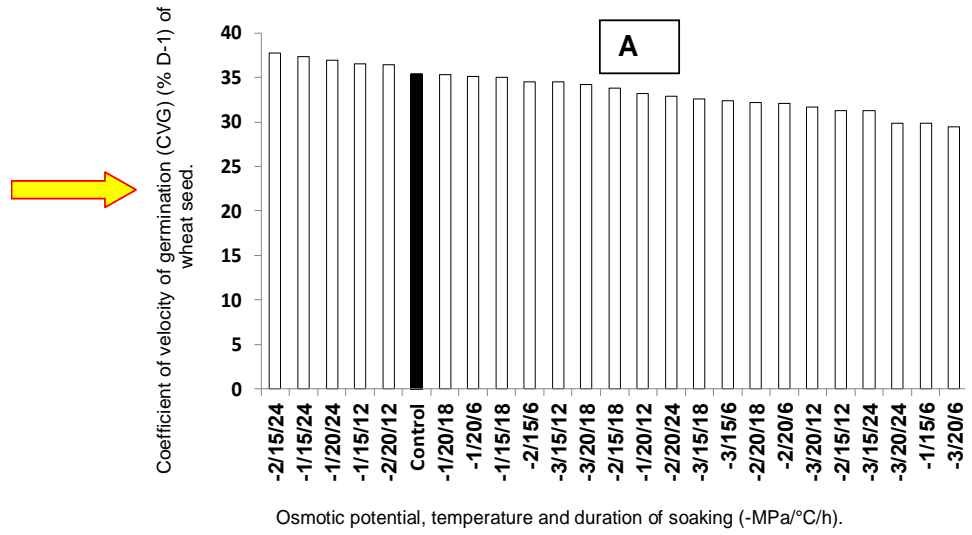


Figure 2. Effect of the osmotic potential of activator solutions and temperature on A: coefficient of velocity of germination CVG (% d⁻¹), (LSD_{0.05}2.2), B: mean germination time (MGT) (d), (LSD_{0.05}0.2) and C: germination rate index (GRI) (% d⁻¹), (LSD_{0.05} 3.1).

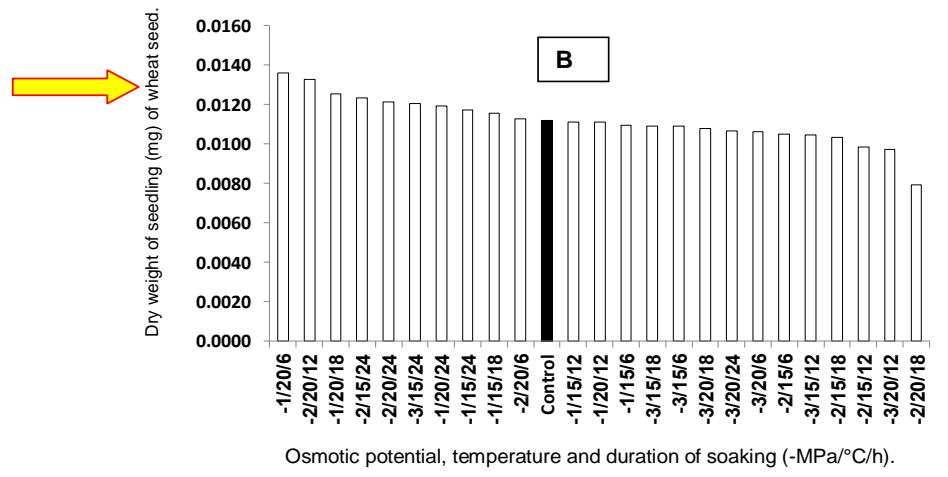
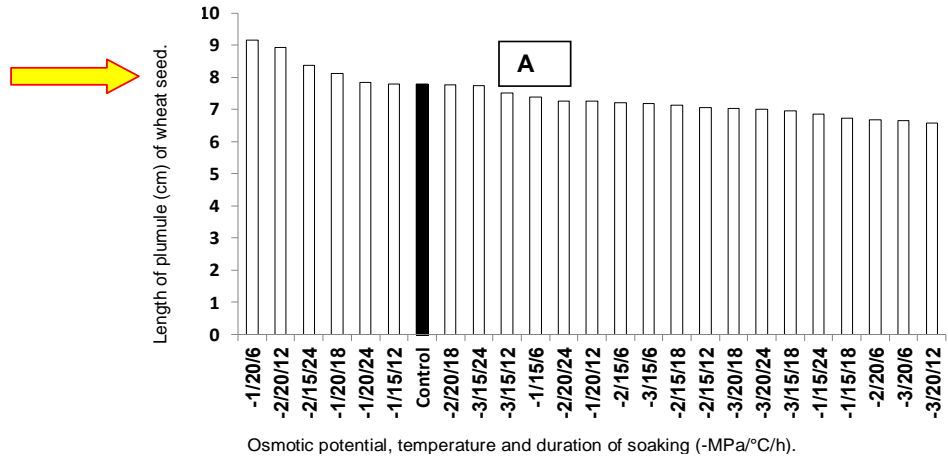


Figure 3. Effect of the osmotic potential of activator solutions and temperature A: on length of plumule (cm), (LSD_{0.05} 0.9) and B: dry weight of seedling (mg), (LSD_{0.05} 0.0016).

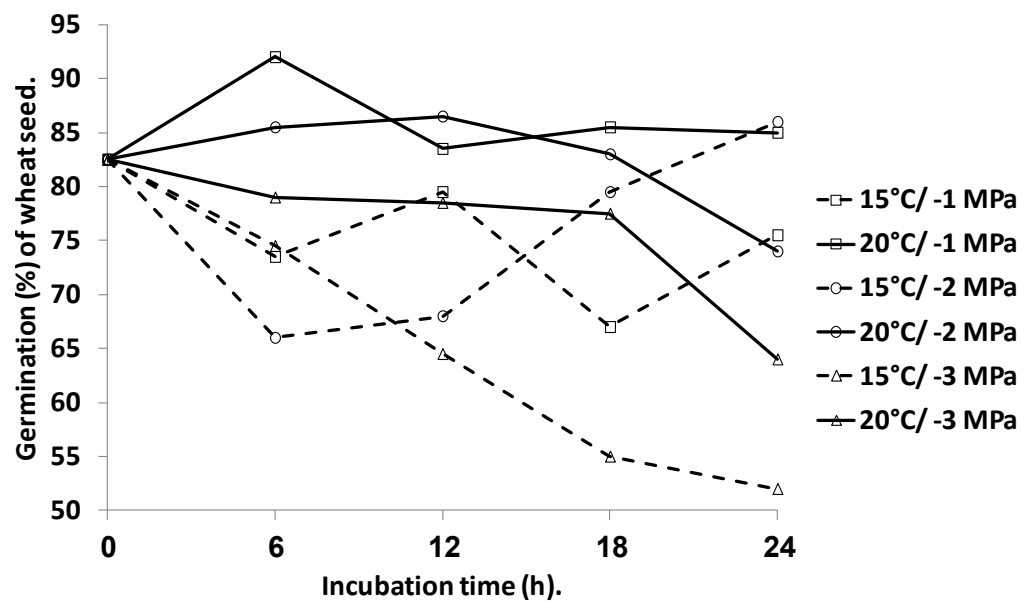


Figure 4. Interaction of temperature and osmotic primer solution over time for first germination count.

Table 1. Correlation between attributes studied of wheat seed.

Attribute	First count	Final count	CVG	MGT	GRI	Length of plumule
Seedling dry weight	0.256*	0.263**	0.401***	-0.389***	0.432***	0.549***
Length of plumule	0.369***	0.312**	0.387***	-0.386***	0.477***	
GRI	0.821***	0.847***	0.548***	-0.536***		
MGT	-0.362***	-0.042	-0.996***			
CVG	0.357***	0.054				
Final count	0.836***					

*, **and *** correlation is significant at the 0.05, 0.01 and 0.001 levels, respectively.

benefits disappear. This was clear in this study at 20°C and for Treatments 1 and 3 MPa at 15°C, but curiously for Treatment 2 MPa at 15°C germination capacity was regained with increasing duration of incubation. This finding is unexplainable and potentially suggests that the PEG is having more than a simple osmotic effect on the imbibing seeds.

The results showed that, activation of wheat seed at a temperature of 20°C improved germination parameters compared to 15°C. It is possible that the temperature of 15°C or increasing in period of soaking may have led to an increase in metabolism in the seed before emergence of radicle and that subsequently led to the increase in CVG and GRI. On other hand, this seems to have led to a loss of viability or death of some seeds during subsequent drying of the seed after soaking. It is possible that in deteriorating seed lots there is a mixed response to priming with some seeds responding in a positive manner whilst others have accentuated loss of viability. Since all tests of this sort are carried out on populations of seeds such variable response is very difficult to separate from the mean response.

The interaction of the 3 factors on the studied traits is intriguing and cannot be explained by any manipulation of the data such as thermal time or a product of thermal time and osmotic potential. Sadeghi et al. (2009) also reported that, the suitable period of priming varies according to kind of osmotic matter, potential of osmotic solution, temperature and species of plant. The Treatment 1 MPa at 20°C for 6 h significantly surpassed the control and the rest of the treatments, including the control treatment to give the highest germination percentage in the first and final count, length of plumule and dry weight of seedling but this did not coincide with the best speed of germination where Treatment 2 MPa at 15°C for 24 h excelled with the highest speed of germination, lowest time for germination and highest germination rate index. This means that, the speed of germination may not be associated with the percentage of germination in some cases, perhaps, because of varying viability and vigour of seeds in the seed lot. This probably indicates a need for a more accurate methodology to activate the seed to avoid the deterioration of marginally viable seed.

Conclusion

Priming with PEG at 1 to Treatment 2 MPa can be used to enhance the viability and vigour of wheat seed which is showing deterioration in germination and vigour, but care must be taken to use a controlled temperature of 20°C for a short duration, around 6 h, in order to maximize the effect.

REFERENCES

- AOSA (1988). Association of Official Seed Analysts. Rules for Testing Seeds. Seed Technol. J. 12(3):109.
- Basra SMA, Farooq M, Tabassum R, Ahmad N (2005). Physiological and biochemical aspects of pre-sowing seed treatments in fine rice (*Oryza sativa* L.). Seed Sci. Technol. 33(3):623-628.
- Bruggink GT, Ooms JJJ, Vander Toorn P (1999). Introduction of longevity in primed seeds. Seed Sci. Res. 9:49-53.
- Chiu KY, Chen CL, Sung JM (2002). Effect of priming temperature on storability of primed sh-2sweet corn seed. Crop Sci. 42:1996-2003.
- Chiu KY, Wang CS, Sung JM (1995). Lipid peroxidation and peroxide-scavenging enzymes associated with accelerated aging and hydration of watermelon seeds differing in ploidy. Physiol. Plantarum. 94:441-446.
- Farooq M, Tabassum R, Afzal I (2006). Enhancing the performance of direct seeded fine rice by seed priming. Plant Prod. Sci. 9:446-456.
- Fu JR, Lu SH, Chen RZ, Zhang BZ, Liu ZS, Cai DY (1988). Osmo conditioning of peanut (*Arachis hypogaea* L.) seed with peg to improve vigour and some biochemical activities. Seed Sci. Technol. 16:197-212.
- Giri GS, Schillinger WF (2003). Seed priming winter wheat for germination, emergence and yield. Crop Sci. 43:2135-2141.
- Heydecker W, Higgins J, Gulliver RL (1973). Accelerated germination by osmotic seed treatment. Nature. 246:42-44.
- Hsu JL, Sung JM (1997). Antioxidant role of glutathione associated with accelerated aging and hydration of triploid watermelon seeds. Physiol. Plantarum. 96:6-12.
- International Seed Testing Association (ISTA) (1993). International Rules for Seed Testing. Seed Sci. Technol. 21:142-168.
- Ismail AMA (1997). The Germination of Seeds. University of Qaar, College of Sciences, Department of Plant. House of Science for Printing, Publishing and Distribution, Qatar. III(17):393-394.
- Kader MA (2005). A comparison of seed germination calculation formulae and the associated interpretation of resulting data. J. P. Royal Soc. New South Wales. 138:65-75.
- Kader MA, Jutzi SC (2004). Effects of thermal and salt treatments during imbibition on germination and seedling growth of sorghum at 42/19°C. J. Agron. Crop Sci. 190:35-38.
- Leila Y, Aghaalkhani M, Khazaei F (2010). Effect of seed priming duration and temperature on seed germination behavior of bread wheat (*Triticum aestivum* L.). ARPJ. Agric. Biol. Sci. 5(1):1-6.

- Quinn GP, Keough MJ (2005). Experimental Design and Data Analysis for Biologists. 4th edn, Chapter 8, pp. 173-207. Cambridge University press, The Edinburgh Building, Cambridge CB2 2RU, UK.
- Sadeghi S, Rahnavard A, Ashrafi ZY (2009). Study of respond seeds wheat (*Triticum aestivum* L.) to osmotic priming, temperatures and local seed masses. Bot. Res. Int. 2(2):69-73.
- Salehzade H, Shishvan MI, Ghiyasi M, Forouzin F, Siyahjani AA (2009). Effect of seed priming on germination and seedling growth of wheat (*Triticum aestivum* L.). Res. J. Biol. Sci. 4(5):629-631.
- Sharifzadeh F, Heidari ZH, Mohamadi H, Janmohamadi M (2006). Study of osmotic priming effects on wheat (*Triticum aestivum* L.) germination in different temperatures and local seed masses. J. Agron. 5(4):647-650.
- Sowmya HR, Doss DD, Gowda R (2012). Effect of osmo-priming with poly ethylene glycol (PEG) on accelerated aged wheat cultivars. Res. J. Agric. Sci. 3(2):221-228.
- Sung JM, Chang YH (1993). Biochemical activities associated with priming of sweet corn seeds to improve vigour. Seed Sci. Technol. 21:97-105.
- Tilden RL (1984). Reversal of the Effects of Deterioration in Aged Soybean Seeds. pp. 42-53, Ph.D. Dissertation, Dept. of Agronomy., Coll. of Agriculture, University of Florida, Florida, US.