

SEED PRIMING WITH THIOUREA IMPROVES THE PERFORMANCE OF LATE SOWN WHEAT

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ABSTRACT

A study was conducted at Agronomic Research Farm, Department of Agronomy, University of Agriculture, Faisalabad, Pakistan during 2012-13 to explore the influence of seed priming treatments especially thiourea on the emergence, growth and yield of late sown wheat. Six seed priming treatments (T₁: control, T₂: hydropriming, T₃: 200 ppm, Thiorea T₄: 400 ppm thiourea, T₅: 600 ppm thiourea, and T₆: 800 ppm thiourea were trial on two wheat (*Triticum aestivum* L.) cultivars viz; Millat-2011 and Punjab-2011. Leaf area index and crop growth rate were recorded at 15 interval from 45 to 105 days after sowing while data on yield and yield related traits were recorded at crop maturing. The results showed that seed primed with thiourea at 800 ppm took minimum time to reach at 50 percent emergence. In both cultivars, maximum leaf area index and crop growth rate were observed in thiourea primed seed at 400 ppm concentration at each interval while minimum dry weight was observed in control at each interval. It further improved yield and yield related attributes (productive tillers, 1000 grain weight, number of grain per spikes, grain and biological yield as well as harvest index). Improved yield in thiourea primed seeds mainly is attributed to uniform germination, good stand establishment and increase in productive tillers. Hence, thiourea seed priming can be used to improve the performance of late sown wheat.

KEYWORDS: *Triticum aestivum* L; seed treatment; liovrea; growth; yield; Pakistan.

INTRODUCTION

Wheat (*Triticum aestivum* L.) is the most important cereal and staple food in Pakistan. Area under wheat cultivation during previous year was about 9,039 thousand hectares with a production 25.3 million tons. It contributes about 12.5 percent in value added in agriculture and shares 2.6 percent in GDP (2).

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Rice-wheat and cotton-wheat are two major cropping systems in Pakistan. In Indo-Pak, farmers follow rice-wheat cropping system who mostly prefer basmati rice cultivars rather than coarse or hybrid rice. These cultivars that mature late than coarse rice, result in delay in wheat sowing. Further, late rice harvest combined with deformed soil structure, rice residue management and occurrence of rainfall (during seed bed preparation) often lead to delay in wheat sowing (24). Farmer community often sacrifices for wheat crop to get more cotton pickings which ultimately results in delaying in sowing of wheat crop (17). Except Pakistan, late sowing of wheat is a major limitation in many South Asian countries (1). Previous studies showed that after mid-November, one day delay in wheat sowing may reduce one percent yield per hectare (4). Due to delay in sowing till December 5th, a yield loss of 42 percent has been recorded in rice-wheat and cotton-wheat cropping systems (30). Overall impact of late sowing in wheat reflected in form of shortening of plant height, reduction in number of internodes, less number of days to heading, maturity and grain filling period and finally in the reduction of yield and yield components (28). Farmers often use broadcast method for wheat sowing which further results in poor and sparse stand establishment. Thus good and uniform stand establishment is a major challenge for wheat growers and its importance is well recognized by the farmers and plant scientists (7).

Different strategies like seed priming (12), development of early maturing varieties of rice and cotton (26, 29), relay cropping and zero tillage (19) can be adopted to overcome the problem of late sowing of wheat. Among these, seed priming is one of the most economical and easy method which can be easily practiced by the farmers (5). Seed primed with ideal concentration of plant growth hormones showed higher germination and also improved the growth and yield of many crops under normal and stress conditions (3). Over night seed soaking of wheat has resulted in vigorous early growth, better stand establishment, higher tillering capacity and grain yield (13). Seed priming improved seed emergence, seed vigor, and increased yields of vegetables and field crops (20). Seed priming is doable and being used commercially in many crops to get uniform germination and better stand establishment under environmental stress condition (16). Primed seeds took less time to accomplish tillering, jointing, heading and flowering stages than unprimed seeds (21). Harris *et al.* (18) reported an increase in wheat mean yield upto 36% (due to seed priming) in on-farm trials at various locations. Hence seed priming can be used in seed invigoration and to compensate the losses due to delayed sowing of wheat (32).

Seed priming may be done in tap water (hydropriming), aerated low water potential solutions of polyethylene glycol or by using many priming agents like KNO_3 , KCl , K_2PO_4 , KH_2PO_4 , MgSO_4 , CaCl_2 , NaCl (osmopriming), plant growth regulators, polyamines (hormonal priming) and plant growth promoting bacteria (biopriming) (6).

Thiourea is one of the important bio-regulator used to induce tolerance against abiotic stresses due to its water solubility and quick absorption in living tissues. It is reported that thiourea at low concentrations breaks dormancy in salinity stressed seeds (15). It was reported previously that seed priming with dimethyl thiourea inhibits ROS production and favors root and shoot growth (9). Improvement in net photosynthesis and chlorophyll contents after thiourea application was also observed in clusterbean (14). However, little information is available about seed priming with thiourea under field conditions. Present study was conducted to explore the role of thiourea in emergence, growth and yield of late sown wheat under field conditions.

MATERIALS AND METHODS

This study was carried out in the Department of Agronomy, Agronomic Research Farm, University of Agriculture, Faisalabad, Pakistan during 2012-13. The experimental land was ploughed via disc plough and then fine seedbed was prepared using rotavator. Experiment was laid out in randomized complete block design under factorial arrangement with three replications. Six seed priming treatments (control, hydropriming, 200 ppm thiourea, 400 ppm thiourea, 600 ppm thiourea, 800 ppm thiourea) were trial on two local wheat cultivars Millat-2011 and Punjab-2011 obtained from Ayub Agricultural Research Institute, Faisalabad

For hydropriming treatment, 378 g seed of each cultivar was soaked in distilled water (1875 mL) in continuously aerated solution for 12 hours (23) and dried in shade before sowing. During soaking period continuous fresh air was supplied through plastic pipes connected with an aeration pump. The seeds were taken out and dried to their original weight at room temperature for two days under shade with forced air (23). In osmopriming 378 g seed of each cultivars was soaked in continuously aerated solution of thiourea according to treatment, An electro-magnetic (Magnetic stirrer / Hot plate, Model CJJ-1) stirrer was used to dissolve the solution completely. The seeds were taken out and dried near to their original weight at room temperature for two days under shade with forced air.

After treatments, seeds were given three surface washings with distilled water and re-dried near to their original weight with forced air under shade. These seeds were sealed in polythene bags and stored in a refrigerator at 7 °C for further studies.

The sowing was done with a hand drill keeping row to row distance of 22.5 cm, using 100 kg per hectare seed rate. Sowing was done on 15th December 2013. Nitrogen and phosphorus was applied @ 150-100 kg per hectare to all treatments. Whole of phosphorus and half of nitrogen was applied at sowing while remaining nitrogen was applied in two splits with two irrigations; one at 25 and second at 75 days after sowing. Weeds (grasses) were controlled by hand weeding at 15 days interval.

Time to obtain 50 percent germination (E_{50}) was calculated according to the formula of Coolbear et al. (8) modified by Farooq et al. (10):

$$E_{50} = t_i + \frac{(N/2 - n_i)(t_j - t_i)}{n_j - n_i}$$

Where N is the final count of emerged seeds, n_i and n_j are the cumulative number of seeds emerged by adjacent counts at times t_i and t_j when $n_j > N/2 > n_i$

Leaf area index was estimated using leaf area meter. At maturity, 30 cm long row area was harvested at 15 days interval for determination of fresh and dry weight. Crop samples were harvested at ground level from a randomly selected area of 1 m² at two different places of each treatment. The biomass of each harvested sample was recorded for fresh weight (g) and then sun dried for 8-10 days and oven-dried at 70 °C for 24 hours till constant weight to obtain dry weight (g). The height of three plants from each replication was measured from soil surface to the final growing point with the help of a meter rod and then averaged,

At maturity, 1 m² area from each treatment was harvested and manually threshed. Number of productive and unproductive tillers were counted from each replication of each treatment at final harvest. To record spike length, three randomly selected spikes were measured separately and then averaged. Five spikes were removed from randomly selected plants in each replication of each treatment to record number of spikelets, grains per spike and 1000-grains weight.

Sun dried wheat from each plot was threshed manually and grain yield was recorded and converted on hectare basis. Total dry matter biological yield was calculated by weighing the total sample of 1 m² of each treatment and converted to tons per hectare. Harvest index was calculated as the ratio of grain yield to total biomass at final harvest.

$$\text{Harvest index (\%)} = (\text{Grain yield} / \text{Biological yield}) \times 100$$

The collected data were analyzed statistically using Fisher's analysis of variance technique software Statistix 8.1 and least significant difference test was applied at 0.05 probability level to compare treatment means (30).

RESULTS AND DISCUSSION

Time taken to complete 50% emergence:

The data revealed (Table 1) that cultivars showed non-significant effect on T50. Different priming treatments showed significant impact on this trait. Maximum time taken to 50 percent emergence (10.30 days) was recorded in control (untreated seed). Seed primed with 800 ppm thiourea took minimum time to complete 50 percent emergence (8.24 days) that was comparable to seed primed at 200, 400, 600 ppm thiourea for both cultivars (Table 1). Seed priming enhances germination speed thus enables the plants to complete its developmental and other phenological stages timely (22). Zheng (33) also recorded improved seed germination in primed seeds of rice. Seed priming also better enables the seeds to complete pre-germination processes in time thus required less E50 (11).

Crop/growth:

Leaf area index (LAI) is considered an important parameter and factor for the estimation of evapo-transpiration, photosynthetic activity and ultimately the crop growth. And crop growth rate (CGR) were determined at 45, 60, 75, 90 and 105 days after sowing. Seed primed with 400 ppm thiourea produced maximum leaf area in both cultivars while minimum was observed in control at each interval (Fig. 1 a and b). CGR expressed the dry matter accumulation by crop plants. CGR at each interval was significantly improved except 90-105 days interval in which it decreased. In both cultivars (Millat-2011, Punjab-2011) maximum CGR was observed where seed was primed with thiourea 400 ppm at each interval (Fig. 2a and b). Minimum CGR was observed in

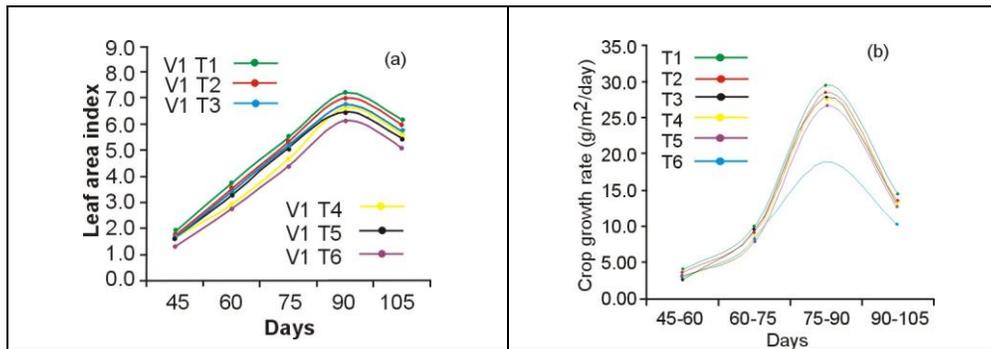


Fig.1. Effect of thiourea seed priming on a) leaf area index and b) crop growth rate in Millat-2011. (V₁ = Millat-2011; T₁ = Control; T₂ = Hydropriming; T₃ = Thiourea 200 ppm; T₄ = Thiourea 400 ppm; T₅ = Thiourea 600 ppm; T₆ = Thiourea 800 ppm).

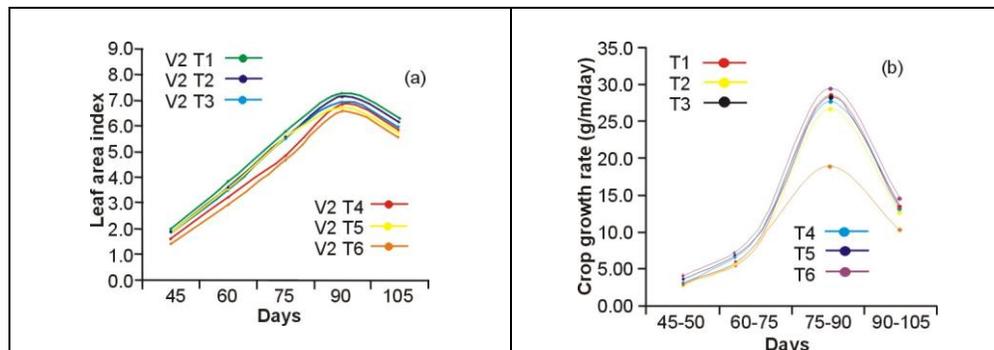


Fig. 2. Effect of thiourea seed priming on a) leaf area index and b) crop growth rate in Punjab-2011. (V₁ = Millat-2011; T₁ = Control; T₂ = Hydropriming; T₃ = Thiourea 200 ppm; T₄ = Thiourea 400 ppm; T₅ = Thiourea 600 ppm; T₆ = Thiourea 800 ppm).

control at each interval. Seed priming with thiourea also showed significant effect on spike length. Interestingly, both cultivars showed statistically similar trend regarding spike length where longest spikes (16.15 cm) were measured in case of seed priming with 400 ppm thiourea followed by hydropriming (15.16 cm) (Table 1). Minimum spike length (14.17 cm) was observed in control where seed was sown without any priming treatment that was statistically similar to seed priming with 200 ppm and 600 ppm thiourea. Improved growth and phenology was also reported by Farooq *et al.* (12) when primed seeds were used in late sown of wheat. They further reported higher post-anthesis CGR than pre-anthesis stage in late sown wheat. The increased spike length with primed seed might be due to the reason that thiourea enhanced enzymatic activity and produced more resistance in plant against abiotic stresses. Subhan *et al.* (31) also reported larger spike length due to priming in wheat.

Table 1. Effect of different concentrations of thiourea priming on time to 50% emergence, plant height, productive and non-productive tillers and spike length of two wheat cultivars.

Treatments	Time to 50% emergence (Days)	Plant height (cm)	Productive tillers/m ² (m ⁻²)	Non-productive tillers/m ² (m ⁻²)	Spike length (cm)
Millet-2011	8.56	98.18	284.67	14.66	15.17
Punjab-2011	8.68	101.47	277.22	14.38	15.23
LSD (p≤0.05)	NS	NS	NS	NS	NS
Control	10.30 A	97.05 B	258.83 C	19.16 A	14.17 C
Hydropriming	8.63 B	98.55 AB	266.50 BC	17.33 AB	15.46 AB
Thiourea (200 ppm)	8.24 BC	100.38 AB	282.33 B	14.33 ABC	14.89 BC
Thiourea (400 ppm)	8.08 BC	101.66 A	310.00 A	10.50 BC	16.15 A
Thiourea (600 ppm)	8.25 BC	100.23 AB	284.17 B	13.00 BC	15.43 AB
Thiourea (800 ppm)	8.24 C	101.08 A	283.83 B	12.83 BC	15.10 BC
LSD (p≤0.05)	0.5998	3.94	23.453	5.35	0.94

Table values are means of three replicates (n=3). Values shares a letter in common within a column do not differ significantly at p≤0.05. NS: non-significant.

Table 2. Effect of different concentrations of thiourea priming on spikelet per spike, grains per spike, 1000-grain weight, grain and biological yield and harvest index of wheat cultivars.

Treatments	Spikelet / spike	Grain/ per spike	1000-grain weight (g)	Grain Yield (t/ ha)	Biological Yield (t ha ⁻¹)	Harvest index (%)
Millet-2011	16.5	43.34	35.00	4.83	14.56	16.5
Punjab-2011	16.5	43.2	35.27	5.03	14.89	16.5
LSD (p≤0.05)	NS	NS	NS	NS	NS	NS
Control	14.00 C	39.39 C	31.16 D	3.97 D	10.59 C	32.00 B
Hydropriming	15.77 BC	42.83 B	33.91 C	4.42 C	13.40 BC	33.25 AB
Thiourea (200 ppm)	17.00 AB	43.70 B	35.71 B	5.11 B	16.33 AB	31.28 B
Thiourea (400 ppm)	18.11 A	47.77 A	39.06 A	5.77 A	17.25 A	33.75 AB
Thiourea (600 ppm)	16.83 AB	43.38 B	35.51 B	5.01 B	15.81 AB	37.18 A
Thiourea (800 ppm)	16.77 AB	42.55 BC	35.43 B	5.18 B	13.97 AB	36.87 A
LSD (p≤0.05)	1.84	3.24	1.36	0.30	1.01	2.07

Table values are means of three replicates (n=3). Values shares a letter in common within a column do not differ significantly at p≤0.05. NS: non-significant.

Yield and related attributes

Significant effects of seed priming were also noted on spikelets per spike, grains per spike, 1000-grain weight, grain yield, biological yield and harvest index. Thiourea (400 ppm) produced markedly higher yield and related attributes than other seed priming treatments (Table 2). However, maximum harvest index (37.18%) was recorded in 600 ppm thiourea primed seed followed by 800 and 400 ppm thiourea treatments. Further, hydropriming also improved wheat yield (33.25%) compared to control (32.50%) but not proved as effective as thiourea. Too concentrated or too diluted thiourea solutions for seed priming must be cared.

The increased number of grains might be due to early germination with priming and good stand establishment. These results are in line with Farooq *et al.* (13) who also recorded improved yield in primed than non-primed seeds. Moreover, thiourea has been reported as significantly improving growth yield and water use efficiency of wheat under arid and semi-arid conditions (27). Moreover, thiourea not only increased K^+ mobilization but also helps in greater accumulation of malate due to the carboxylation of phosphoenol pyruvic acid indicating stimulated dark fixation of CO_2 in embryonic axes. In this way growth and yield increase significantly (25).

In crux, seed primed with thiourea not only reduced the time taken to complete 50 percent emergence but also enhance yield attributes and grain yield. Therefore it can be employed to improve the performance of late sown wheat.

REFERENCE

1. Ahmed, S.M. and C. Meisner. 1996. Wheat research and development in Bangladesh. Bangladesh Australia Wheat Improvement Project and CIMMYT-Bangladesh.
2. Anon. Pakistan Economic Survey. 2013-14. Ministry of Finance, Islamabad, Pakistan.
3. Anosheh, H.P., Y. Emam and M. Ashraf. 2014. Impact of cycocel on seed germination and growth in some commercial crops under osmotic stress conditions. Arch. Agron. Soil Sci. 60:1277-1289.
4. Aslam, M., A. Majid and M.A. Gill 1999. Zero tillage wheat production technology: prospects and threats. J. Sci. Tech. Dev. 18:17-23.
5. Bell, R.W. and B. Dell. 2008. Micronutrients for sustainable food, feed, fibre and bioenergy production. Int. Fert. Indus. Assoc. Paris, France. PMID: 14506888.
6. Chiu, K.Y., C.L. Chen and J.M. Sung. 2002. Effect of priming temperature on storability of primed sweet corn seed. Crop Sci. 42: 1996-2003.
7. Chivasa, W., D. Harris, C. Chiduzza, P. Nyamudeza and A.B. Mashingaidze. 1998. Agronomic practices, major crops and farmers' perceptions of the importance of good stand establishment in Musikavanhu Communal Area, Zimbabwe. J. Appl. Sci. South Afr. 4:9-25.
8. Coolbear, P., A. Francis and D. Grierson. 1984. The effect of low temperature pre-sowing treatment under the germination performance and membrane integrity of artificially aged tomato seeds. J. Exp. Bot. 35: 1609-1617.

9. De Zacchini, M. and M. De Agazio. 2001. Dimethyl thiourea, a hydrogen peroxide trap, partially prevents stress effects and ascorbate peroxidase increase in spermidine treated maize roots. *Plant, Cell Environ.* 24:237-244.
10. Farooq, M., S.M.A. Basra, N. Ahmad and K. Hafeez. 2005. Thermal hardening: a new seed vigor enhancement tool in rice. *J. Integrative Plant Biology.* 47:187-193.
11. Farooq, M., S.M.A. Basra and A. Wahid. 2006: Priming of field-sown rice seed enhances germination, seedling establishment, allometry and yield. *Plant Growth Regul.* 49:285-294.
12. Farooq, M., S.M.A. Basra, H. Rehman and B.A. Saleem. 2008. Seed priming enhances the performance of late sown wheat (*Triticum aestivum* L.) by improving chilling tolerance. *J. Agron. Crop Sci.* 194:55-60.
13. Farooq, M., S.M.A. Basra, A. Wahid, A. Khaliq and N. Kobayashi. 2009. Rice seed invigoration. In: *Sustainable Agriculture Reviews*. Lichtfouse, E. (ed.). Book Series, Springer.
14. Garg, B.K., U. Burman and S. Kathju. 2006. Influence of thiourea on photosynthesis, nitrogen metabolism and yield of clusterbean (*Cyamopsis tetragonoloba* (L.) Taub.) under rainfed conditions of Indian arid zone. *Plant Growth Regul.* 48:237-245.
15. Gul, B. and D.J. Weber. 1998. Effect of dormancy compounds on the seed germination of non-dormant *Allenrolfea occidentalis* under salinity stress. *Ann. Bot.* 82:555-560.
16. Rowse, H.R. 1996. Drum priming-an environmentally friendly way of improving seed performance. *J. R. Agric. Soc. England.* 157:77-83.
17. Hameed, E., W.A. Shah, A.A. Shad, J. Bakhat and T. Muhammad. 2003. Effect of different planting dates, seed rates and nitrogen levels on wheat. *Asian J. Pl. Sci.* 2:467-474.
18. Harris, D., B.S. Raghuvanshi, J.S. Gangwar, S.C. Singh, K.D. Joshi, A. Rashid and P.A. Hollington. 2001. Participatory evaluation by farmers of on-farm seed priming in wheat in India, Nepal, and Pakistan. *Exp. Agric.* 37:403-415.
19. Iqbal, M., M.A. Khan, M.Z. Anwar and A.Q. Mohsin. 2002. Zero-tillage Technology and Farm Profits: A case study of wheat growers in the rice zone of Punjab. *The Pakistan Development Review.* 41:665-682.
20. Johnson, S.E., J.G. Lauren, R.M. Welch and J.M. Duxbury. 2005. A comparison of the effects of micronutrient seed priming and soil fertilization on the mineral nutrition of chickpea (*Cicer arietinum*), lentil

- (*Lens culinaris*), rice (*Oryza sativa*) and wheat (*Triticum aestivum* L.) in Nepal. Exp. Agric. 41:427-448.
21. Kant, S., P. Verma and S.S. Pahuja. 2003. Growth and yield maintenance in bread wheat by seed priming under late-sown condition. Acta Agronomica Hungarica. 51:445-453.
 22. Kant, S., S.S. Pahuja and R.K. Pannu. 2006. Effect of seed priming on growth and phenology of wheat under late-sown conditions. Trop. Sci. 44: 9-15.
 23. Lee, S.S. and K.J. Hyeun. 1998. Total sugars, α -amylase activity and germination after priming of normal and aged rice seeds. Korean J. Crop Sci. 45:108-111.
 24. Mann, R.A., M. Ramzan and A. Munir. 2008. Improving the sustainability of wheat production in irrigated areas of Punjab, Pakistan through conservation tillage technology. Int. J. Agri. Biol. 10: 249-54.
 25. Pandey M., Srivastava A.K., S.F. D'Souza and S. Penna. 2013. Thiourea, a ROS Scavenger, Regulates Source-to-Sink Relationship to Enhance Crop Yield and Oil Content in *Brassica juncea* (L.). PLoS ONE 8: e73921.
 26. Rashid, M., A.A. Cheema and M. Ashraf. 2007. Clustering of Basmati rice mutants through metro glyph analysis. Pak. J. Bot. 39:2043-2049.
 27. Sahu, M.P. and D. Singh. 1995. Role of thiourea in improving productivity of wheat (*Triticum aestivum* L.). J. Plant Growth Regul. 14: 169-173.
 28. Sial, M.A., M.A. Arain, S.K. Mazhar, H. Naqvi, M. Umar and A.N. Nizamani. 2007. Yield and quality parameters of wheat genotypes as affected by sowing dates and high temperature stress. Pak. J. Bot. 3:575-584.
 29. Singh, P. 2004. Cotton breeding. Kalyani Pub., New Delhi, India.
 30. Steel, R.G.D., J.H. Torrie and D.A. Dickey. 1997. Principles and Procedures of Statistics. A biometrical approach, 3rd Ed. McGraw Hill Book Co. Inc., New York. Pp: 172-177.
 31. Subhan, F., A. Nazir, M. Anwar, N.H. Shah, M. Siddiq, I. Ali, J. Rahman and T. Sajjad. 2004. Response of newly developed wheat cultivars/advance lines to planting dates in the central Agro-ecological zones of NWFP. Asian J. Plant Sci. 3:87-90.
 32. Welbaum, G.E. and K.J. Bradford. 1991. Water relations of seed development and germination in muskmelon. VI. Influence of priming on germination responses to temperature and water potential during seed development J. Exp. Bot. 42:393-399.

33. Zheng, H.C., H.U. Jin, Z. Zhi, S.L. Ruan and W.J. Song. 2002. Effect of seed priming with mixed-salt solution on germination and physiological characteristics of seedling in rice (*Oryza sativa* L.) under stress conditions. *J. Zhejiang Univ. Agric. Life Sci.* 28:175-178.

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Usman Bilal : Conducted the research