Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

E-ISSN: 2278-4136
P-ISSN: 2349-8234
www.phytojournal.com
JPP 2020; 9(4): 804-808
Received: 02-05-2020
Accepted: 04-06-2020

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Effects of magnesium nitrate and boric acid on germination and seedling growth parameters of wheat (*Triticum aestivum* L.) var. HUW-468

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Abstract
Plant requires both macro and micronutrients for regulating their metabolic process throughout different phases of growth and development. Magnesium and Boron as a part of macro and micronutrient respectively perform specific function inside plant body. Here, to find out the effect of these two nutrients as seed priming agent, an experiment was conducted. In the present piece of experiment wheat seeds of variety HUW -468 were soaked for 12 hours in solutions of $Mg(NO_3)_2$ (macro nitropriming, C_1), H_3BO_3 (micro nutripriming, C_2), combination of $Mg(NO_3)_2$ + H_3BO_3 (combined nutripriming C_3), distilled water (hydropriming, C_4) for priming purpose. Thereafter all the four sets are dehydrated under forced air and used for experimentation. Various parameters like germination percentage, germination parameters includes: FGP, MGT, GI, GRI, CVG, MGR, plumule, radicle and seedling lengths, fresh and dry weights of radicle & plumule, alpha amylase activity, soluble sugar contents and seedling vigour (index I and II) were determined at 3, 5, 7 and 9th days of seed germination and seedling growth and were compared with non- primed control sets (C_0). Result concluded that magnesium nitrate primed seeds (C_1) showed better result for all the parameters mentioned except alpha amylase activity & soluble sugar content, later two parameters were best in boric acid (C_2) primed one. However, seeds primed with combination of magnesium nitrate and boric acid (C_3) showed best result in radicle length and fresh & dry weights of it. In all the observed parameters it was noted that macro and micro primed sets performed better alone or in combination than hydro primed and non-primed control sets.

Keywords: Magnesium Nitrate, Boric Acid, Seedling Growth, *Triticum aestivum* L. and HUW-468

Introduction
Many scientists are working diligently to increase the production & productivity of wheat. Crop need macronutrient & micronutrient both for their growth and development. Magnesium, the macronutrient is a constituent of chlorophyll and also required for the pigment formation which is important for photosynthesis. Magnesium also regulates the activity of enzyme rubisco, hence directly or indirectly effects the yield. It is also require to join two units of ribosomes hence involve in translation of protein. The micronutrient boron also involved in reproduction via influencing germination of pollen as well as translocation of sugars. Hence magnesium and boron being one of the most important macro and micronutrient respectively also play an important role in wheat crop production. $Mg(NO_3)_2$ primed wheat seeds of var. HUW-206, HUW-234 & HP-1102 performed better than the control one (Bose *et al*. 1992) [6]. Several researches has been carried out in which it was found that various field crops like wheat, rice, maize & mustard primed seeds showed improvement in germination physiology, vegetative growth and yield (Bose & Sharma 2006; Bose 2007; Bose *et al*. 2007; Mondal *et al*. 2011; Anaytullah 2012) [13, 7, 7, 11, 3]. Boron enhances the mean germination, germination percentage, germination energy & germination time in rice (Farooq *et al*. 2011) [9] and seedling emergence, growth & yield in wheat (Iqbal *et al*. 2017) [10]. Every mineral nutrient has its specific role to perform in plant metabolism and in present investigation it was noticed that magnesium and boron perform their role in a justified manner when their salts are used as priming agent in seed priming technique.

Materials and methods
The present piece of experiment was conducted in seed priming research laboratory of department of plant physiology of Institute of Agricultural Sciences, Varanasi. Wheat seeds of variety HUW-468 used was obtained from the department of Agronomy of the same institute. Healthy, disease free and vigorous seeds were first sterilized by using 0.1% $HgCl_2$ solution for 2-3 minutes and washed thoroughly with distilled water. The seeds were then soaked for 12 hours in 7.5mM Mg $(NO_3)_2$ and in 8mM H_3BO_3 alone, or in combination of both and in

distilled water (hydro primed). After 12 hours of soaking, seeds were dried naturally so that there was no moisture present on the surface of the seeds. After proper drying, seeds was taken for germination studies in which seeds were placed in Petri-plates containing germination paper and 10 ml of distilled water was poured in each Petri-plate. Another set (without any treatment) which is depicted as control will also placed in Petri-plates for germination studies. All the Petri-plates containing 4 treatments and control one along with their four replications are kept in laboratory at normal room temperature about 20±2 °C. The Soaked seeds were then dried up by keeping them under fan until to come back to its original weight. Germination and seedling growth studies were made upto 9 days in petri-plates under laboratory condition. The treatments were C_0, C_1, C_2, C_3, C_4 representing non primed seeds, seeds primed with 7.5mM $Mg(NO_3)_2$, 8mM H_3BO_3, the combination of 7.5mM $Mg(NO_3)_2$ and 8mM H_3BO_3 and distilled water (hydro primed).

Seed germination and measurement of growth

Seed germination percentage and other seed germination parameters were calculated at 24, 48, 72, 96 & 120 hours during germination by using the following formula:

i) Germination percentage = Number of seeds germinated/ Total number of seeds sown × 100
ii) Final germination percentage (FGP in %)= Total no. of seeds germinated on end day/No. of seeds present in Petri dish × 100
iii) Mean time germination (MTG in days)= ΣF*x/ΣF
Where F is the number of seeds germinated on day x; by Al-Mudaris (1998) [1]
iv) Germination index (GI)= GI= (3×n1) + (2×n2) + · · · + (1×n4)
n1= one day germinated seed, n2= second day germinated seed, n3= third day germinated seed, n4= fourth day germinated seed; by Ranal *et al*. (2009) [12]
v) Germination rate index (GRI%/day)= G1/1 + G2/2 +· · ·+ Gx/x
G1 = Germination percentage ×100 on the first day after sowing, G2 = Germination percentage ×100 on the second day after sowing; by Al-Mudaris (1998) [1]
vi) Coefficient of velocity of germination (CVG)= N1 + N2 + · · · + Nx/100 × N1T1 + · · · + NxTx ; N=No. of seeds germinated each day, T=No. of days from seeding corresponding to N; by Al-Mudaris (1998) [1]
vii) Mean germination rate (MGR)= CVG/100; by Ranal *et al*. (2009) [12]

The radicle & plumule lengths of growing wheat seedlings were measured by using cm scale. Fresh and dry weights of plumule and radicle were measured by conventional method. For estimation of dry weights, the samples of plumule and radicle were kept in oven for one hour pre-set at 100-110 °C for killing purpose. Thereafter, the temperature was set at 60±2 °C till to get constant weight of the samples. Seedling vigour I & II was calculated by using the following formula:
Seedling vigour I: germination %× (shoot length + root length).
Seedling vigour II: germination % × seedling dry weight.
Biochemical parameters i.e., α-Amylase and soluble sugar were estimated using the methods of Bernfield (1955) [5] and Dubois (1956) [8] respectively to validate the effect of macronutrient and micronutrient in the form of magnesium & boron respectively on growing wheat seedlings. All the physico chemical & biochemical parameters were studied at 3, 5, 7, and 9th days during germination under laboratory condition using statistical analysis.

Result and Discussion

Table 1. Represented percentage germination in presence of H_3BO_3, $Mg(NO_3)_2$, H_3BO_3 + $Mg(NO_3)_2$, hydro primed and non-primed control. In 24 hours during seed germination it was noted that C_1 showed better germination percent than C_2, C_3, C_4 and C_0 respectively. The same pattern of germination percentage was observed at 48 and 72 hours of germination. At 96 hours of germination C_3, C_2, C_1 all these three sets showed 99 percent germination while C_0 & C_4 showed 94 and 95 percent germination respectively. And at 120 hours of germination C_3, C_2, C_1 all the three remained the same showing 99 percent germination while C_0 and C_4 showed 95 and 96 percent germination respectively. It was observed that the seed set which was primed with magnesium nitrate showed better germination percentage than the other four treatments. Hydropriming and osmopriming of wheat seeds may improve seed germination and emergence which ultimately leads to vigorous root growth and development (Ashraf *et al*., 2005) [4]. As magnesium nitrate salt tends to increase germination percentage in wheat seed. Application of $Mg(NO_3)_2$ as priming agent improves the process of germination via activating various enzymes (Hexokinase, Phosphofructokinase, Enolase) of respiratory cycle & (Decarboxylase, Nitrate reductase) nitrogen metabolism and it also improves plant height, fresh & dry weight, total nitrogen content & nitrate reductase activity in roots & shoots etc. (Anaytullah & Bose *et al*., 2007) [2].

Table 2. Showed that the plumule length is found more in C_1 which is followed by C_2, C_3, C_4 and C_0 respectively at 3, 5, 7, 9 days during germination. In case of radicle length it is found that C_3 shows highest radicle length followed by C_1, C_2, C_4 and C_0 at 3, 5, 7 and 9th days during germination. The total seedling length at 3, 5, 7 and 9th days during germination was found more in C_1 followed by C_3, C_2, C_4 and C_0. Seeds primed with Mg $(NO_3)_2$ (C_1) provided better length of plumule and total seedling length at each observation date whereas seeds primed with H_3BO_3 + Mg $(NO_3)_2$ (C_3) only provided better radicle length. In case of plumule length after C_1 better result was observed in C_2 whereas in case of radicle length after C_3, C_1 gave good result but in case of total seedling length after C_1, second best result was obtained with C_3. Magnesium nitrate hardened seeds showed maximum plumule and seedling length (Sharma *et al*. 2006) [13], (Anaytullah & Bose 2007) [2].

Table 3 and 4 represented the fresh and dry weights of plumule and radicle. Observation was taken at 3, 5, 7 and 9th days during germination. It was found that seeds treated with magnesium nitrate (C_1) showed best result in respect to fresh plumule followed by C_2, C_3, C_4 and C_0 respectively. In case of fresh weight of radicle best result was obtained in C_3, followed by C_1, C_2, C_4 and C_0 respectively. Different treatments have represented different result for various parameters. C_1 set has given best result in respect to fresh plumule weight where C_3 set provided better result in case of fresh radicle weight. Fresh weight of plumule was more in magnesium nitrate hardened seeds (Sharma & Bose 2006) [13], whereas fresh weight of radicle were more in seeds treated with combination of both magnesium nitrate and boric acid as compared to other nutri-treatments. Data was taken and analyzed at 3, 5, 7 and 9th days during germination in respect to calculate the dry weights of plumule and radicle. It was found that C_1 showed best result in respect to dry plumule

weight, which was followed by C_2, C_3, C_4 and C_0 respectively. In case of dry weight of radicle, best result was obtained in C_3, C_1, C_2, C_4 and C_0 respectively. Different pre-soaking nutrient solution yields different results in different parameters like C_1 provided best result in observations like dry weight of plumule, C_3 provide better result in case of dry weight of radicle. Dry weight of plumule was more in magnesium nitrate hardened seeds as compared to other nutri treatments (Bose & Mishra 1992) [6], whereas dry weight of radicle were more in seeds treated with combination of both magnesium nitrate and boric acid as compared to other nutri-treatments.

In these observed parameters as regard to seedling vigour index I and II, C_1 showed better result than other treatments, this may be due to seeds primed with magnesium nitrate which has already shown highest germination percentage and highest total seedling length. However, both these parameters has shown that seeds treated with magnesium nitrate (C_1) gave best result followed by seeds treated with combination of magnesium nitrate and boric acid (C_3), boric acid (C_2), distilled water (C_4) & non primed seeds (C_0) respectively based on their respective germination percentage and total seedling length (Table 5 and 6).

α-amylase activity: This has been represented in Figure 1, where boric acid primed sets (C_2) showed maximum α-amylase activity, it was observed that boric acid enhanced the activity of the enzyme α-amylase followed by combination of magnesium nitrate & boric acid (C_3), magnesium nitrate (C_1), hydro (C_4) and control (C_0) sets respectively. Result showed that the effect of boron on activity of α-amylase was more prominent and visible in respect to magnesium and nitrate. α-amylase activity was noted to increase steadily from 3rd day to 7th day and then declined at 9th day, where the seeds primed with boric acid (C_2), showed highest activity of enzyme in comparison to other four treatments. Therefore, it may be suggested that Boron is better for solubilization and mobilization which might be regulated via α-amylase activity.

Soluble sugar: Figure 2 represented the soluble sugar concentration in endosperm. In this case, seeds primed with boric acid (C_2) showed highest content of soluble sugar, followed by combination of magnesium nitrate & boric acid primed (C_3), magnesium nitrate primed (C_1), hydro primed (C_4) & non primed control (C_0) respectively at 3, 5, 7, 9th days during germination. Soluble sugar content increases rapidly from 3rd day to 7th day then after 7th days onwards it started to decrease upto 9th day. Plants can easily uptake the sugar in soluble form which acts as substrate to provide energy for further biochemical & metabolic processes and this solubilization & mobilization process are better in boric acid primed seeds.

Table 1: Effect of magnesium nitrate and boric acid priming (used either alone or in combination), hydropriming and non priming control on wheat seed variety HUW-468 on germination percentage at different hours

Treatments	Hours of germination (Hours)				
	24	48	72	96	120
C_0	81.50	85.00	91.00	94.00	95.00
C_1	93.25	96.50	98.75	99.00	99.00
C_2	93.00	96.25	98.25	99.00	99.00
C_3	92.00	96.00	98.00	99.00	99.00
C_4	89.50	92.50	96.50	95.00	96.00
SEd	1.22	0.93	0.59	0.58	0.58
CD	2.60	1.98	1.26	1.23	1.23

Table 2: Effect of magnesium nitrate and boric acid priming (used either alone or in combination), hydropriming and non-priming control on wheat seed variety HUW-468 on plumule, radicle and seedling length at different days of seed germination/seedling growth

Treatments	Plumule length (CMS) (Days)				Radicle length (CMS) (Days)				Seedling length (CMS) (Days)			
	3	5	7	9	3	5	7	9	3	5	7	9
C_0	0.43	2.45	4.65	7.45	0.26	2.57	5.07	9.58	0.69	4.95	9.72	17.03
C_1	0.64	3.93	7.84	9.68	0.50	4.75	8.50	12.83	1.13	8.35	16.33	22.52
C_2	0.62	3.60	6.32	9.43	0.47	4.39	7.32	12.28	1.08	7.79	13.64	21.70
C_3	0.59	3.38	6.11	9.23	0.54	4.83	9.44	13.28	1.12	8.03	15.54	22.50
C_4	0.52	2.85	6.05	8.80	0.33	3.62	6.19	11.45	0.84	6.37	12.24	20.25
SEd	0.02	0.10	0.09	0.11	0.01	0.05	0.04	0.12	0.03	0.13	0.09	0.15
CD	0.04	0.20	0.18	0.23	0.02	0.10	0.09	0.25	0.06	0.27	0.20	0.31

C_0-control non primed seeds, C_1-seeds primed with 7.5mM $Mg(NO_3)_2$, C_2-seeds primed with 8mM H_3BO_3 boric acid, C_3-seeds primed with the combination of 7.5mM $Mg(NO_3)_2$ and 8mM H_3BO_3, C_4-seeds primed with distilled water (hydro primed) respectively.

Table 3: Effect of magnesium nitrate and boric acid priming (used either alone or in combination), hydropriming and non-priming control on wheat seed variety HUW-468 on fresh weight of plumule, and radicle at different days of seed germination/seedling growth

Treatments	Plumule (Days)				Radicle (Days)			
	3	5	7	9	3	5	7	9
C_0	11.44	17.53	24.40	38.25	11.43	17.97	33.55	34.34
C_1	21.85	29.40	40.45	48.61	19.60	28.48	42.67	47.82
C_2	18.77	27.84	38.63	44.57	16.45	25.70	39.49	44.69
C_3	16.47	26.14	36.06	41.44	21.54	31.45	44.87	49.58
C_4	12.34	21.13	31.45	37.61	13.73	21.44	36.43	39.44
SEd	0.02	0.03	0.20	0.15	0.11	0.12	0.12	0.08
CD	0.04	0.06	0.42	0.30	0.22	0.25	0.25	0.16

fresh weight of plumule, and radicle at different days of seed germination /seedling growth.

C_0-control non primed seeds, C_1-seeds primed with 7.5mM $Mg(NO_3)_2$, C_2-seeds primed with 8mM H_3BO_3 boric acid, C_3-seeds primed with the combination of 7.5mM $Mg(NO_3)_2$ and 8mM H_3BO_3, C_4-seeds primed with distilled water (hydro primed) respectively.

Table 4: Effect of magnesium nitrate and boric acid priming (used either alone or in combination), hydropriming and non-priming control on wheat seed variety HUW-468 on dry weight of plumule, radicle and seedling at different days of seed germination/seedling growth

Treatments	Plumule (mg) (Days)				Radicle (mg) (Days)			
	3	5	7	9	3	5	7	9
C_0	0.68	1.28	2.32	5.38	0.55	1.26	2.17	2.94
C_1	1.95	2.37	3.15	6.95	1.43	2.18	3.14	3.70
C_2	1.67	2.15	2.95	6.75	1.39	1.97	2.86	3.34
C_3	1.33	1.92	2.71	6.44	1.88	2.43	3.36	3.39
C_4	1.06	1.61	2.43	5.09	1.16	1.40	2.47	3.16
SEd	0.01	0.03	0.02	0.02	0.07	0.02	0.04	0.03
CD	0.02	0.05	0.03	0.04	0.14	0.04	0.08	0.06

C_0- control non primed seeds, C_1- seeds primed with 7.5mM $Mg(NO_3)_2$, C_2- seeds primed with 8mM H_3BO_3 boric acid, C_3- seeds primed with the combination of 7.5mM $Mg(NO_3)_2$ and 8mM H_3BO_3, C_4- seeds primed with distilled water (hydro primed) respectively.

Table 5: Effect of magnesium nitrate and boric acid priming (used either alone or in combination), hydropriming and non-priming control on wheat seed variety HUW-468 on Seedling vigour index l at different days of seed germination/seedling growth

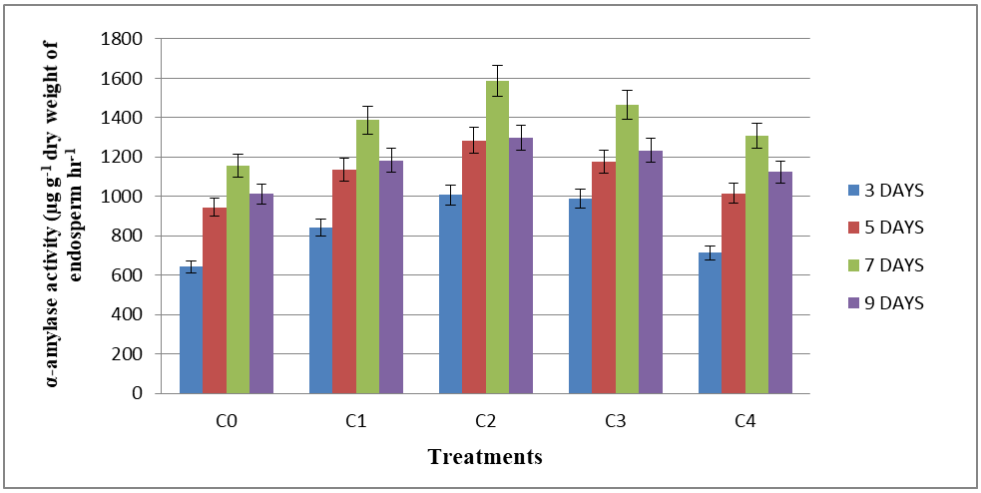
Treatments	Seedling vigour index l (Days)			
	3	5	7	9
C_0	58.28	450.31	913.97	1617.38
C_1	108.50	817.81	1616.92	2327.50
C_2	104.23	768.73	1350.11	2148.30
C_3	107.74	788.73	1538.71	2227.50
C_4	77.94	614.38	1163.06	1944.00
SEd	3.07	12.74	12.00	18.69
CD	6.55	27.15	25.58	39.82

C_0- control non primed seeds, C_1- seeds primed with 7.5mM $Mg(NO_3)_2$, C_2- seeds primed with 8mM H_3BO_3 boric acid, C_3- seeds primed with the combination of 7.5mM $Mg(NO_3)_2$ and 8mM H_3BO_3, C_4- seeds primed with distilled water (hydro primed) respectively.

Table 6: Effect of magnesium nitrate and boric acid priming (used either alone or in combination), hydropriming and non priming control on wheat seed variety HUW-468 on Seedling vigour index ll at different days of seed germination /seedling growth.

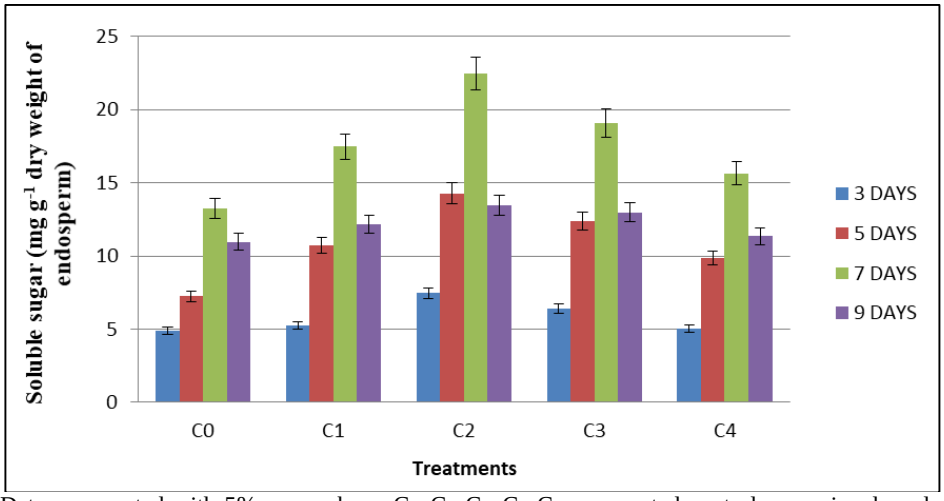
Treatments	Seedling vigour index ll (Days)			
	3 (Days)	5 (Days)	7 (Days)	9 (Days)
C_0	104.54	230.68	421.11	789.45
C_1	320.23	445.66	622.22	1054.10
C_2	299.39	406.62	575.19	997.92
C_3	307.92	426.91	600.68	1027.13
C_4	204.89	289.97	465.27	791.52
SEd	7.49	3.62	5.08	5.96
CD	15.96	7.72	10.83	12.71

C_0- control non primed seeds, C_1- seeds primed with 7.5mM $Mg(NO_3)_2$, C_2- seeds primed with 8mM H_3BO_3 boric acid, C_3- seeds primed with the combination of 7.5mM $Mg(NO_3)_2$ and 8mM H_3BO_3, C_4- seeds primed with distilled water (hydro primed) respectively.



Data represented with 5% error where, C_0, C_1, C_2, C_3, C_4 represented control non primed seeds, seeds primed with 7.5mM $Mg(NO_3)_2$, 8mM H_3BO_3, the combination of 7.5mM $Mg(NO_3)_2$ and 8mM H_3BO_3 and distilled water (hydro primed) respectively.1

Fig 1: Effect of magnesium nitrate and boric acid priming (used either alone or in combination), hydropriming and non-priming control on wheat seed variety HUW-468 on α-amylase activity during different days of seed germination



Data represented with 5% error where, C_0, C_1, C_2, C_3, C_4 represented control non primed seeds, seeds primed with 7.5mM $Mg(NO_3)_2$, 8mM H_3BO_3, the combination of 7.5mM $Mg(NO_3)_2$ and 8mM H_3BO_3 and distilled water (hydro primed) respectively.

Fig 2: Effect of magnesium nitrate and boric acid priming (used either alone or in combination), hydropriming and non-priming control on wheat seed variety HUW-468 on soluble sugar content during different days of seed germination

Data represented with 5% error where, C_0, C_1, C_2, C_3, C_4 represented control non primed seeds, seeds primed with 7.5mM $Mg(NO_3)_2$, 8mM H_3BO_3, the combination of 7.5mM $Mg(NO_3)_2$ and 8mM H_3BO_3 and distilled water (hydro primed) respectively.

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