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Pre-Storage Fungicidal Treatment Effects on Prevalence of Seed-Borne Fungi and Quality of Onion (*Allium cepa* L.) Seed

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Abstract

Effect of pre-storage fungicidal treatment on prevalence of seed-borne fungi and quality of onion (*Allium cepa* L.) seeds was evaluated under laboratory condition. Seeds were treated separately with five fungicides namely Bavistin 50 WP (Carbendazim) and Dithane M-45 (Mancozeb) @ 2.2 g/kg, Homai (Thiophanate) and Rovral (Iprodione) @ 2.0 g/kg and Provax-200 (Carboxin) @ 3.0 g/kg of seeds before storage. Prevalence of seed-borne fungi, percent seeds germination and vigor index were evaluated just before storage, 75 and 150 days after storage. Fungicides produced significant negative impact on seed-borne fungi and positive impact in maintaining quality during storage. Provax-200, Rovral, Dithane M-45 and Bavistin effectively reduced seed-borne fungi and performed better for germination and vigor of seeds. Seed treatment with Provax-200, Rovral, Bavistin or Dithane M-45 would therefore be suggested as effective fungicides to control seed-borne fungal infection and maintain better germination and vigor of onion seed in storage.

Keywords: Pre-storage, seed treatment, seed health, germination, vigor index.

1. Introduction

Onion (Allium cepa L.) is one of the most demanding spice crops in Bangladesh. Onion has manifold uses such as spice, vegetable, salad and condiments for foods and medicines. As demand of onion is increasing, production of onion needs to increase in order to meet the requirement in the years to come. There exist enormous scope to increase the productivity of onion of which the most important one is use of good quality seed having high germination capacity and health status (Kant *et al.*, 1999). There are so many factors like loss of seed vigor and viability, low germination and fungal prevalence which may impair onion seed health status during storage (Justice and Bass, 1979; Singh and Bhonde,

2003). Fungal pathogens like Aspergillus niger, A. flavus, Fusarium oxysporum, Alternaria porri and Penicillium spp. are responsible for onion seed infection. These pathogens are frequently transmitted through onion seeds (Hayden and Mude, 1992; Beliard et al., 1998; Fakir, 1998). Aspergillus spp. Penicillium spp. and Fusarium spp. can cause losses of quality of onion seeds in storage (Gupta et al., 1984). Ahmed and Hossain (1985) stated that the fungus can affect seed germination and seedling growth. Seed-borne fungi adversely affect seeds in storage. They bring down germinability, discolor the seeds, enhance biochemical changes, and accumulate toxins, and results loss in weight (Neergaard, 1979). Thus control of fungal infection of seeds in storage is one of the important factors to maintain proper germination, vigor and health of seeds. Among recommended practices, seed treatment probably the cheapest and easiest method of plant disease control. Treatment of seeds with chemicals has been proved to be effective in reducing seed-borne infection (Dubey and Singh, 2005; Barua et al., 2007). Application of fungicides effectively inhibits F. oxysporum and A. niger and contributes to seed germination and vigor index (Behrani et al., 2015, Patra, 2017, Saranya et al., 2017). Prestorage seed treatment may reduce the prevalence of seed-borne fungi and protect seeds from the invasion of fungi in storage condition. The present research work has therefore been undertaken to study the effect of pre-storage seed treatment of fungicides on health and quality of onion seed in storage.

2. Materials and Methods

A seed sample of onion variety Taherpury was used in the experiment. Onion seeds were treated with five fungicides namely Bavistin 50 WP (Carbendazim) and Dithane M-45 (Mancozeb) @ 2.2 g/kg, Homai (Thiophanate) and Rovral (Iprodione) @ 2.0 g/kg and Provax-200 (Carboxin) @ 3.0 g/kg of seeds before storage. Fifty gram of seeds was treated separately for each fungicide. Required amount of fungicide was mixed properly with seeds in a conical flask by hand shaking. Treated seeds were then stored in plastic containers at ambient temperature. An untreated control was maintained. Seed health, germination and vigor index were tested following standard methods (ISTA, 1996) just before storage, 75 and 150 days after storage (DAS).

2.1 Seed health test

Seed health test of onion seeds was carried out by standard blotter method (ISTA, 1996). In this method, three layers of blotter papers (Whatman No. 1) were soaked in sterilized distilled water and placed on the bottom of the 9 cm diameter Pyrex glass Petridish. Four hundred seeds from each treatment were taken randomly and then placed on the moist blotter paper at the rate of 25 seeds per plate. The experiment was laid out following Completely Randomized Design (CRD) with four replications. The petridishes with seeds were then incubated at $25\pm2^{\circ}$ C temperature in 12/12 hours alternating cycles of light and darkness for seven days. After incubation, the seeds were examined under stereo-binocular microscope for the presence of seed-borne fungal pathogens and identified by observing their growth characters. In case of confusion, temporary slide was prepared and examined under a compound microscope and identified using appropriate keys of Mathur and Kongsdal (2003). Results were expressed in percentage of seeds infected by the pathogens.

2.2 Germination test

Four hundred seeds were randomly taken from each treatment for germination test (ISTA, 1996). Three layered moistened blotter paper was taken on petridishes. Hundred seeds were placed on each Petridish and kept at 25° C temperature for seven days for germination. Seedlings were counted every day up to the completion of germination at 7th day. Only the normal seedling was counted and germination percentage was calculated.

2.3 Seed vigor index

Seed vigor index (VI) was assessed through speed of germination of seeds. Speed of germination was derived from the germinated seedlings at an interval of 24 hours for seven days according to Copeland (1976) using the following formula:

No. of germinated seeds at first count

VI = ______ Days of first count No. of germinated seeds at final count

Days of final count

3. Results and Discussion

3.1 Effect of pre-storage seed treatment on the prevalence of seed- borne fungi

Two genera of fungi Aspergillus and Fusarium were recorded from seed. The fungal prevalence

in seeds increased with the increase in storage period. Fungal prevalence was minimal just after treatment (0 DAS) with fungicides. The highest population of Aspergillus spp. (A. niger and A. flavus) was found in control (8.5%). The lowest prevalence on seeds was found at immediately after storage with Provax-200 and Bavistin (4.0%). At 75 days after storage (DAS), prevalence of the fungi was statistically similar in all the fungicides treated seeds but significantly lower than control. At 150 DAS, prevalence of Aspergillus spp. was the lowest in Rovral treated seeds which was statistically similar with Bavistin, Provax-200 and Dithane M-45. Prevalence of Fusarium sp. in seeds was very low (0 to 3.00%) both in treated and untreated control seeds during the storage period of 150 days. No infection of the fungus was recorded at 75 and 150 days of storage due to treatment with Provax-200 and Dithane M-45 but in both time 3.0% infection was recorded in

control. In consideration of total fungal infection, Rovral was the most effective fungicide followed by Provax-200, Dithane M-45 and Bavistin. These four fungicides were statistically similar in controlling total seed-borne fungi and prevalence of fungi was lower than control (Table 1).

3.2 Effect of pre-storage seed treatment on germination

At the time of storage, germination of seeds ranged from 89.00 to 91.00% due to different treatments. The differences in germination under various treatments including control were insignificant. At 75 and 150 days after storage (DAS), the germination reduced to 77.50-84.25 and 69.25-78.75%, respectively due to different treatment. At 75 DAS, the highest germination of 84.25% was recorded from seed treated with Provax-200 which was statistically similar with other fungicides but significantly higher compared to control.

 Table 1. Effect of pre-storage seed treatment on prevalence of seed-borne fungi of onion seeds at 0, 75 and 150 days after storage

Treatm-	Aspergillus spp*. (%)			Fusarium sp. (%)			Total fungi (%)		
ent	0	75	150	0	75	150		75	150
ent	DAS	DAS	DAS	DAS	DAS	DAS	0 DAS	DAS	DAS
Bavistin	4.00 c	6.50 b	8.75 bc	1.25	1.00 b	1.00 b	5.25 b	7.50 b	9.75bc
	(11.54)	(14.77)	(17.21)	(6.419)	(5.739)	(5.739)	(13.246)	(15.894)	(18.195)
Dithane	5.00 bc	7.00 b	9.50 bc	2.00	0.00 c	0.00 c	7.00 b	7.00 b	9.50bc
M-45	(12.92)	(15.34)	(17.95)	(8.130)	(0.286)	(0.286)	(15.342)	(15.342)	(17.952)
Homai	6.00 b	7.50 b	10.75 ab	1.50	1.00 b	1.00 b	7.50 b	7.50 b	10.75 b
	(14.18)	(15.89)	(19.14)	(7.035)	(5.739)	(5.739)	(15.894)	(15.894)	(19.140)
Rovral	5.00 bc	6.25 b	7.50 c	1.75	1.00 b	1.00 b	6.75 b	7.25 b	8.50 c
	(12.92)	(14.48)	(15.89)	(7.602)	(5.739)	(5.739)	(15.059)	(15.620)	(16.951)
Provax-	4.00 c	6.25 b	9.00 bc	2.00	0.00 c	0.00 c	6.00 b	6.25 b	9.00bc
200	(11.54)	(14.48)	(17.46)	(8.130)	(0.286)	(0.286)	(14.179)	(14.478)	(17.458)
Control	8.50 a	11.00 a	12.25 a	2.00	3.00 a	3.00 a	10.50 a	14.00 a	15.25 a
	(16.95)	(19.37)	(20.49)	(8.130)	(9.974)	(9.974)	(18.907)	(21.973)	(22.986)
CV%	19.94	20.47	13.47	27.62	20.00	20.00	24.50	20.10	12.40
LSD	1.605	2.256	1.092	NS	0.495	0.495	2.608	2.464	1.926
0.05									

DAS: Days after storage; Values within the same column with common letter(s) did not differ significantly ($P \le 0.05$) by LSD, NS: non- significant at 5% level of probability, Values within the parenthesis are the arcsine transformed value. **Aspergillus* spp. include *A. niger* and *A. flavus*

The lowest germination of 77.50% was recorded from the control. At 150 DAS, germination of seeds treated with all five fungicides was significantly higher compared to control. The highest germination was recorded from seeds treated with Provax-200 (78.75%) which was statistically similar with Rovral (76.50%), Bavistin (76.25%) and Dithane M-45 (75.50%). The lowest germination percentage was 69.25% under control (Table 2).

3.3 Effect of pre-storage seed treatment on vigor index

At the time of storage, vigor index of fungicide treated seeds and untreated seeds (control) ranged from 34.56 to 35.60. The differences in vigor index under all treatments including control were statistically similar. Vigor of seeds decreased with the increase of storage period. At 75 DAS vigor index ranged from 28.97 to 32.78. The highest vigor index was recorded from the

seed treated with Provax-200 (32.78) which was statistically similar with Rovral (32.72) and Bavistin (31.00) and significantly higher compared to control. The lowest vigor index was recorded in control (28.97). At 150 DAS, the vigor index varied from 24.33 to 29.26 under different treatments including control. The highest vigor index was recorded from seeds treated with Provax-200 which was statistically similar with Rovral, Dithane M-45 and Bavistin. The lowest vigor index was recorded in control which was statistically significant (Table 3).

Results of the present experiment revealed that prevalence of seed-borne fungi increased and germination percentage and vigor index decreased gradually with the increase of storage period. However, seed treatment with fungicides minimizes the prevalence of seed-borne fungi and provides better germination and vigor index over untreated control.

T		Germination (%)	
Treatment	0 DAS	75 DAS	150 DAS
Bavistin 50 WP	89.00	81.00 ab	76.25 ab
	(70.63)	(64.16)	(60.83)
Dithane M-45	91.00	82.75 a	75.50 ab
	(72.54)	(65.46)	(60.33)
Homai	90.75	81.25 ab	75.00 b
	(72.29)	(64.34)	(60.00)
Rovral 50 WP	89.50	81.50 ab	76.50 ab
	(71.09)	(64.53)	(61.00)
Provax-200	90.75	84.25 a	78.75 a
	(72.29)	(66.62)	(62.55)
Control	90.00	77.50 b	69.25 c
	(71.57)	(61.68)	(56.32)
CV%	2.70	3.10	3.61
LSD (0.05)	NS	3.743	3.175

 Table 2. Effect of pre-storage seed treatment on germination of onion seeds at 0, 75 and 150 days after storage

DAS: Days after storage; Values within the same column with common letter(s) did not differ significantly ($P \le 0.05$) by LSD, NS: non- significant at 5% level of probability, Values within the parenthesis are the arcsine transformed value.

The stars and	Vigor index				
Treatment	0 DAS	75 DAS	150 DAS		
Bavistin	34.61	31.00 ab	27.21 ab		
Dithane M-45	34.56	30.17 b	28.15 ab		
Homai	35.25	29.73 b	26.30 bc		
Rovral	35.60	32.72 a	28.89 a		
Provax-200	35.41	32.78 a	29.26 a		
Control (No treatment)	34.97	28.97 b	24.33 c		
CV%	1.84	4.50	5.46		
LSD (0.05)	NS	2.065	2.218		

 Table 3. Effect of pre-storage seed treatment on vigor index of onion seeds at 0, 75 and 150 days after storage

DAS: Days after storage; Values within the same column with common letter(s) do not differ significantly ($P \le 0.05$) by LSD, NS: non- significant at 5% level of probability

Provax-200, Rovral, Dithane M-45 and Bavistin were found effective in reducing seed-borne infection of Aspergillus spp. (A. niger and A. flavus) and Fusarium sp. of onion seeds, and improved germination and vigor index were also recorded for these four fungicides. Various authors also reported that fungicide dressing significantly reduced seed-borne infection and increased the percent germination and vigor index (Akgul et al., 2011; Rohtas, 2014; Saranya et al., 2017). Efficacy of Bavistin 50 WP, Rovral 50 WP and Provax-200 was reported against F. oxysporum and Aspergillus spp. Bavistin 50 WP was found best to control seed-borne fungi followed by Provax-200 and Rovral 50 WP. Fungicidal seed treatment also increased the germination of soybean seed (Chaity et al., 2011). Fungicides like Carbendazim, Thiophanate-methyl, Benomyl, Prochloraz and Tebuconazole showed inhibiting effects against F. oxysporum and their application increased the emergence of seedlings (Song et al. 2004; Ozer and Koycu, 2004; Rajput et al., 2006; Chandel and Deepika, 2010). Gupta et al. (2012) reported that fungicides such as Carbendazim (Bavistin) and Thiram as seed dresser were effective against seed-borne fungus A. niger and concluded that these fungicides significantly increased the percent germination and vigor index of onion seeds. Verma et al. (2014) found

that after 8th month of soybean seed storage, germination and vigor index were significantly higher in Vitavax 200 @ 2 g kg⁻¹ and Carboxyn @ 2 g kg⁻¹ coated seed samples over control. Patra (2017) reported that the onion seed treated with Mancozeb as pre-storage treatment provide improved germination and field performance even after 120 days of storage. The results of the present experiment agreed well with the previous findings.

4. Conclusions

Pre-storage seed treatment with fungicides greatly contributed to health and quality of onion seeds during storage. Provax-200, Rovral, Bavistin and Dithane M-45 were the most effective fungicides which reduced seed-borne infection of *Aspergillus* spp. and *Fusarium* sp. and improved germination and vigor index over control. Therefore, seed treatments with Provax-200, Rovral, Bavistin or Dithane M-45 are suggested as effective fungicides to control seed-borne fungi and for better germination and vigor of onion seeds.

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