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A SURVEY TO ASSESS WHEAT RUST INCIDENCE IN BANGLADESH DURING 2010-2011 WHEAT GROWING SEASON

K. Mustarin[1], P. K. Malaker[1], M. M. A. Reza[1], M. S. Hossain[1], M. A. Hakim[1], N. C. D. Barma[2], M. Mokhlesur Rahman[3], R. Islam[4] and M. Mahbubur Rahman[5]

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ABSTRACT

K. Mustarin, P. K. Malaker, M. M. A. Reza, M. S. Hossain, M. A. Hakim, N. C. D. Barma, M. Mokhlesur Rahman, R. Islam and M. Mahbubur Rahman. 2014. A survey to assess wheat rust incidence in Bangladesh during 2010-2011 wheat growing season. Bangladesh J. Plant Pathol. 30 (1&2): 1- 5.

A field survey was conducted in the major wheat growing areas of Bangladesh during 2010-2011 wheat growing season to assess the incidence of wheat rusts. Altogether 162 farmers' fields and trial sites covering 26 districts and experimental farms of Wheat Research Centre, Dinajpur, Bangladesh were visited several times during the survey. Among three rust diseases of wheat, incidence of stem or yellow rust was not found but occurrence of leaf rust was found with varying degrees of severity depending on wheat variety and locations of survey. About 37% of the visited fields were infected with leaf rust where the majority of infected fields showed low (<20%) to moderate (20-40%) levels of disease severity. The highest percentage of infected fields was observed in Rajshahi region followed by Rangpur and Mymensingh, while none of the fields was infected in Tangail district under Dhaka region. The variety Kanchan showed moderate levels of disease incidence and severity with MSS type reaction, whereas low to moderate incidence and severity with MSS type response were observed in variety Prodip. The variety Shatabdi showed low disease levels with MR type reaction in Rajshahi region but it was free from the disease in other regions. The variety BARI Gom-26 showed low disease levels with MRMS type reaction. Three advanced lines BAW-1051, BAW-1120 and BAW-1141, and varieties Bijoy and BARI Gom-25 were completely free from rust.

Key words: Rust, incidence, wheat, survey

INTRODUCTION

Wheat (*Triticum aestivum* L.) is the second most important cereal crop after rice in Bangladesh. It covers about 400,000 ha annually with an average production of one million tons (Anon. 2012). The current annual requirement of wheat is about 4 million tons. One-third of the requirement is met through local production. The shortfall of about 3 million tons per year is met through import from foreign countries. The rate of increase in wheat consumption is about 3% per year and by 2020 the annual wheat requirement of the country will be more than 5 million tons. Wheat Research Centre of BARI has so far released 28 wheat varieties of which Shatabdi, Bijoy, Prodip and BARI Gom-26 are important producing about 4.0-4.5 t/ha in the farmers' field (Uddin *et al*. 2012, 2013).

The major constraints of wheat production in Bangladesh are attributed to competition with other winter crops, inadequate crop management; late planting due to late harvest of transplanted Aman paddy and attack of various pests and diseases. Among the diseases *Bipolaris* leaf blight or spot blotch (*Bipolaris sorokiniana*) and leaf rust (*Puccinia triticina* Eriks.) are the major problems(Malaker *et al.* 2007).

(Malaker *et al.* 2007). Yield losses due to leaf rust are usually limited but can be significant if a susceptible variety is grown and infection occurs early in the crop season. The disease appears almost every year in all wheat growing areas of Bangladesh with varying degrees of severity depending on varieties grown. The disease usually appears in mid February and its severity increases between mid and late March. Wheat planted in optimum times (15-30 November), either escapes the disease to a large extent or suffers less compared to those planted in late season. Stem rust of wheat caused by *P. graminis* Pers. f. sp. *tritici* Eriks. & E. Henn. was a common disease during the early years of wheat research and development in the country. However, the disease has not been observed since mid 1980s, possibly due to introduction of several resistant varieties. Yellow rust of wheat caused by *P. striiformis* West. f. sp. *tritici* Eriks. & E. Henn. appears occasionally in north-western parts of the country, where relatively cooler climate prevails during the winter months of wheat growing season.

Rust pathogens explicitly possess two major characteristics. Firstly, they are highly mobile trans-boundary fungi capable of rapid and long distance movements even over continents, either by wind-assisted or inadvertent human-mediated dissemination. Secondly, rust pathogens have an inordinate ability to

change and evolve through mutation or sexual recombination (Waston 1981, Knott 1989, Park 2007). A virulent race of stem rust fungus called Ug99 was detected in Uganda in 1998 (Pretorius *et al*. 2000) and subsequently found in epidemic proportions in Kenya and Ethiopia. The race has knocked down many useful resistance genes including *Sr31* that were deployed in 90% of the wheat varieties grown worldwide (Singh *et al*. 2008). Seven variants of the Ug99 lineage are now known and have spread to various countries in the eastern African highlands as well as Zimbabwe, South Africa, Sudan, Yemen, and Iran (Singh *et al*. 2011). The experts of Borlaug Global Rust Initiative (BGRI) have predicted further movement of Ug99 and other virulent strains to the important wheat production areas of the Indian subcontinent and beyond (McIntosh and Pretorius 2011). Appearance of new virulence in leaf and yellow rust pathogens of wheat is also likely in future, particularly under the changing crop climate. Although none of the rusts has so far reached an epidemic level in Bangladesh, but there is no guarantee that damaging epidemics will not occur in future, particularly if a virulent race develops or is introduced. Therefore, regular survey and monitoring becomes inevitable in order to identify signs of emergence of virulent races of wheat rusts for mitigating their future threats.

In view of the above facts, a survey was conducted to assess the incidence of rust diseases in major wheat growing areas of Bangladesh.

MATERIALS AND METHODS

The survey of wheat rusts was conducted in the major wheat growing districts representing six regions of Bangladesh during 2010-2011 seasons. One hundred and sixty two farmers' fields and trial sites were selected from 26 districts under six wheat growing regions of the country. The work was carried out throughout Wheat Research Centre of Dinajpur, Joydebpur, Jessore, Jamalpur, Rajshahi and Ishurdi. Rust incidence at field level was recorded and severity of infection was estimated following the modified Cobb scale (Stubbs *et al.* 1986). The BGRI protocols for wheat rust assessment (Anon. 2008) were used.

RESULTS AND DISCUSSION

Among 162 farmers' fields and trial sites surveyed, incidence of stem and yellow rusts were not found in any location but leaf rust was observed in 60 fields that accounts for 37% of all visited fields. The highest percentage of infected field (64%) was recorded in Rajshahi region, which was followed by Rangpur, Mymensingh and Jessore region with 52%, 48% and 19% infected fields, respectively. The lowest percentage of 3% leaf rust incidence was observed in Barisal region. All of the fields visited in Tangail district under Dhaka region were free from rust incidence. However, the frequency and percentage of field infection in different regions were largely influenced by number of fields surveyed, time of planting and varieties grown. Wheat planted in November largely escaped or had less disease incidence compared to late-sown crops. Fields planted with varieties Prodip and Kanchan showed higher percentages of leaf rust infection than those planted with other varieties or advanced lines. Infection of fields planted with Prodip and Kanchan was as high as 100% in Mymensingh and Rangpur regions. When planted with BARI Gom-26 in Rangpur and Rajshahi regions, 50% of the fields showed leaf rust infection, while only 20% of the fields planted with Shatabdi was infected in Rajshahi region and found disease free in other regions. Fields planted with other varieties and advanced lines were completely free from leaf rust infection in all the regions. Severity of leaf rust infection was found higher in north-western parts compared to other wheat growing areas of the country (Table 1).

Rust resistance genes present in Bangladeshi wheat varieties observed during the present survey were reported by Malaker and Reza (2011). Genes detected in six varieties are presented in Table 2. A total of six leaf rust resistance genes viz. *Lr1*, *Lr10*, *Lr13*, *Lr23*, *Lr26* and *Lr34*; five stem rust resistance genes viz. *Sr5*, *Sr8b*, *Sr9b*, *Sr11* and *Sr31*; and two yellow rust resistance genes *Yr2KS* and *Yr9*, either singly or in combination were identified in those varieties. The gene *Lr34*, conferring adult plant slow rusting resistance to leaf rust was identified on the basis of conspicuous leaf tip necrosis developed under field condition. In addition to the named genes, some unidentified factors for resistance (+) were also inferred in most of the varieties (Table 2).

Altogether six wheat varieties and three advanced lines observed during the survey showed different levels of leaf rust incidence and severity with different types of disease reactions (Table 3). Among the varieties, Kanchan released in 1983 and having the genes *Lr13+Lr23* showed moderate levels of incidence and severity (20-40%) with MSS type reaction, while Shatabdi released in 2000 and carrying the genes *Lr1+Lr13+* displayed low levels of disease (<20%) with MR type reaction in Rajshahi region but it was disease free in other regions. The variety Prodip showed low to moderate disease levels with MSS reaction, while only low disease severity with MRMS response was recorded in variety BARI Gom-26. Advanced lines BAW-1051, BAW-1120 and BAW-1141, and variety Bijoy released in 2005 and BARI Gom-25 released in 2010 and having the gene *Lr13+* were completely free from leaf rust in all the regions surveyed.

Table 1. Leaf rust prevalence in different wheat growing regions of Bangladesh during 2010-2011

Wheat growing regions	Districts covered	Number of fields surveyed		Leaf rust infected fields	
		Variety/Line	Number of fields	Frequency	% Infected fields
Rangpur	Rangpur	Kanchan	2	2	100
	Kurigram	Shatabdi	6	0	0
	Lalmanirhat	Bijoy	4	0	0
	Dinajpur	Prodip	26	24	92
	Thakurgaon	BARI Gom-25	2	0	0
	Panchagarh	BARI Gom-26	6	3	50
		BAW-1120	6	0	0
		BAW-1141, BAW-1051	2	0	0
Region total	**6**	**9**	**56**	**29**	**52**
Rajshahi	Rajshahi	Shatabdi	5	1	20
	Natore	Bijoy	1	0	0
	Naogaon	Prodip	15	13	87
	C. Nawabganj	BARI Gom-26	4	2	50
	Pabna				
	Sirajganj				
Region total	**6**	**4**	**25**	**16**	**64**
Mymensingh	Mymensingh	Shatabdi	2	0	0
	Jamalpur	Bijoy	6	0	0
	Sherpur	Prodip	11	11	100
	Netrakona	BARI Gom-26	4	0	0
	Kishoreganj				
Region total	**5**	**4**	**23**	**11**	**48**
Jessore	Jessore	Shatabdi	5	0	0
	Magura	Bijoy	4	0	0
	Meherpur	Prodip	4	3	75
		BARI Gom-25	1	0	0
		BARI Gom-26	2	0	0
Region total	**3**	**5**	**16**	**3**	**19**
Barisal	Barisal	Shatabdi	7	0	0
	Faridpur	Bijoy	5	0	0
	Patuakhali	Prodip	8	1	13
	Barguna	BARI Gom-25	6	0	0
	Bhola	BARI Gom-26	11	0	0
		BAW-1120	1	0	0
Region total	**5**	**6**	**38**	**1**	**3**
Dhaka	Tangail	Bijoy	1	0	0
		Prodip	2	0	0
		BARI Gom-26	1	0	0
Region total	**1**	**3**	**4**	**0**	**0**
Grand Total	**26**	**9**	**162**	**60**	**37**

In the late 1980s and early to mid 1990s, the variety 'Columbus' having the genes *Lr13* and *Lr16* was highly resistant to leaf rust in Canada. As the frequency of isolates with virulence to *Lr13* and *Lr16* increased, the resistance conditioned by these genes has been eroded (Kolmer and Liu 2002). The pathotypes having matching virulence for genes *Lr1*, *Lr2a*, *Lr2c*, *Lr3*, *Lr10*, *Lr13*, *Lr14a*, *Lr15*, *Lr17* and *Lr23* were quite common in Bangladesh (Malaker *et al.* 2005). This also correlates with leaf rust infection on the old cultivar Kanchan that showed moderate levels of disease severity with MSS type reaction during the present survey. The variety Prodip released in 2005 and having genes *Lr1+Lr26+* showed low to moderate

levels of disease with MSS type reaction in all the regions except Dhaka. The alien gene *Lr26* (1BL/1RS translocation) has been the most widely exploited source of rust resistance in the world for developing high yielding wheat cultivars (Malaker and Reza 2011). Nayar *et al*. (2001) reported that at least 10 pathotypes with matching virulence for *Lr26* have been identified in India between 1984 and 2001. Leaf rust reaction and severity on Prodip in different wheat growing areas indicates the presence of this type of *Lr26*-virulent pathotypes in Bangladesh. The variety BARI Gom-26 released in 2010 and having genes *Lr10+Lr13+Lr34+* exhibited low levels of disease incidence and severity with MRMS type of reaction. The resistance conferred by *Lr34* is non-specific in nature, since isolates that are fully virulent to this gene have not yet been detected (Kolmer 1999, 2001). Adult plant gene *Lr34* along with some minor resistance genes provides durable resistance to leaf rust in wheat throughout the world (Kolmer 1996). Wheat materials of Bangladesh have a narrow genetic base for resistance to rust diseases. Cultivars with narrow genetic base are unlikely to provide durable resistance and may lead to vulnerability to new races of the rust pathogens. This underlines the needs for field monitoring on a wider scale and development of increased genetic diversity with durable type of resistance in order to mitigate the future threat of wheat rusts in Bangladesh.

Table 2. Rust resistance genes present in the wheat varieties of Bangladesh observed during 2010-2011

Variety	Postulated rust resistance genes		
	Lr gene	*Sr* gene	*Yr* gene
Kanchan	*Lr13+Lr23*	R	-
Shatabdi	*Lr1+Lr13+*	*Sr8b+Sr9b+Sr11+*	*Yr2KS*
Bijoy	-	R	-
Prodip	*Lr1+Lr26+*	*Sr5+Sr31+*	*Yr9+*
BARI Gom-25	*Lr13+*	R	-
BARI Gom-26	*Lr10+Lr13+Lr34+*	*Sr8b+Sr9b+Sr11+*	-

Genes were not postulated; [R]Resistant to all Indian stem rust pathotypes; [+]Unidentified factors for resistance.
Source: Malaker and Reza (2011).

Table 3. Wheat varieties and advanced lines of Bangladesh with their reactions to leaf rust during 2010-2011

Variety/Line	Year of release	Incidence at field level	Severity on infected plants	Leaf rust reaction
Kanchan	1983	M	M	MSS
Shatabdi	2000	0-L	L	MR
Bijoy	2005	0	0	0
Prodip	2005	0-M	L-M	MSS
BARI Gom-25	2010	0	0	0
BARI Gom-26	2010	0-L	L	MRMS
BAW-1051	2011(Advanced)	0	0	0
BAW-1120	2011(Advanced)	0	0	0
BAW-1141	2011(Advanced)	0	0	0

L (low) = less than 20%; M (moderate) = 20-40%; H (high) = more than 40%
R = Resistant; MR = Moderately Resistant; MS = Moderately Susceptible; S = Susceptible; MSS = Moderately Susceptible + Susceptible; MRMS = Moderately Resistant + Moderately Susceptible

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LITERATURE CITED

Anonymous. 2012. Statistical Yearbook of Bangladesh 2011. Bangladesh Bureau of Statistics (BBS), Ministry of Planning, Government of the People's Republic of Bangladesh. p. 206.

Anonymous. 2008. Wheat Rust Survey Protocol 2008. Borlaug Global Rust Initiative (BGRI), Cornell University, Ithaca, NY, USA. 23 pp.

Knott, D. R. 1989. The wheat rusts-breeding for resistance. *In*: Monograph on Theoretical and Applied Genetics. Springer, Verlag, London, Paris, Tokyo. pp. 14–37.

Kolmer, J. A. 1996. Genetics of resistance to wheat leaf rust. Annu. Rev. Phytopathol. 34: 435–455.

Kolmer, J. A. 1999. Physiologic specialization of *Puccinia triticina* in Canada in 1997. Plant Dis. 83: 194–197.

Kolmer, J. A. 2001. Physiologic specialization of *Puccinia triticina* in Canada in 1998. Plant Dis. 85: 155–158.

Kolmer, J. A. and Liu, J. Q. 2002. Inheritance of leaf rust resistance in the wheat cultivars AC Majestic, AC Splendor, and AC Karma. Can. J. Plant Pathol. 24: 327–331.

Malaker, P. K. and Reza, M. M. A. 2011. Resistance to rusts in Bangladeshi wheat (*Triticum aestivum* L.). Czech J. Genet. Plant Breed. 47(Special Issue): S155–S159.

Malaker, P. K., Ahmed, M. U. and Reza, M. M. A. 2007. Research on cereal disease management at Bangladesh Agricultural Research Institute. *In*: Bakr M.A., Ahmed H.U. and Wadud Mian M.A. (eds.). Advances in Plant Pathological Research in Bangladesh. Plant Pathology Division, BARI, Joydebpur, Gazipur. pp. 3–20.

Malaker, P. K., Nayar, S. K., Prasher, M., Bhardwaj, S. C. and Reza, M. M. A. 2005. Analysis of pathotypes and postulation of genes for resistance to leaf rust in wheat. Bangladesh J. Agric. 30: 33–42.

McIntosh, R.A. and Pretorius, Z. A. 2011. Borlaug Global Rust Initiative provides momentum for wheat rust research. Euphytica 179: 1-2.

Nayar, S. K., Nagarajan, S., Prasher, M., Bhardwaj, S. C., Jain, S. K. and Datta, D. 2001. Revised catalogue of genes that accord resistance to *Puccinia* species in wheat. Research Bulletin No. 3. Directorate of Wheat Research Regional Station, Flowerdale, Shimla, pp. 1–48.

Park, R. F. 2007. Stem rust of wheat in Australia. Aust. J. Agric. Res. 58: 558–566.

Pretorius, Z. A., Singh, R. P., Wagoire W. W. and Payne, T. S. 2000. Detection of virulence to wheat stem rust resistance gene *Sr31* in *Puccinia graminis* f. sp. *tritici* in Uganda. Plant Dis. 84: 203.

Singh, R. P., Hodson, D. P., Huerta-Espino, J., Jin, Y., Njau, P., Wanyera, R., Herrera-Foessel, S. A. and Ward, R. W. 2008. Will stem rust destroy the world's wheat crop? Adv. Agron. 98: 271–309.

Singh, R. P., Hodson, D. P., Huerta-Espino, J., Jin, Y., Bhavani, S., Njau, P., Herrera-Foessel, S., Singh, P. K., Singh, S. and Govindan, V. 2011. The emergence of Ug99 races of the stem rust fungus is a threat to world wheat production. Ann. Rev. Phytopathol. 49: 465–481.

Stubbs, R.W., Prescott, J. M., Saari, E. E. and Dubin, H. J. 1986. Cereal Disease Methodology Manual. CIMMYT, Mexico. 46 pp.

Uddin, J., Barma, N. C. D., Malaker, P. K., Sarker, Z. I., Sarker, A. Z., Bodruzzaman, M. and Hakim, M. A., Eds. 2012. WRC Annual Report 2011-2012. Wheat Research Centre, BARI, Nashipur, Dinajpur. 183 pp.

Uddin, J., Barma, N. C. D., Malaker, P. K., Hossain, M. I., Sarker, Z. I., Khaleque, M. A., Bodruzzaman, M., Hakim, M. A. and Hossain, A. 2013. WRC Annual Report 2012-2013. Wheat Research Centre, BARI, Nashipur, Dinajpur. 249 pp.

Watson, I. A. 1981. Wheat and its rust (*Puccinia*) parasites in Australia. *In*: Evans, L.T. and Peacock, W.J. (eds.). Wheat Science: Today and Tomorrow. Cambridge University Press, Cambridge, England. pp. 129–147.

EVALUATION OF FUNGICIDES AGAINST *RHIZOCTONIA ORYZAE-SATIVAE*, THE CAUSAL FUNGUS OF AGGREGATE SHEATH SPOT OF RICE

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ABSTRACT

S. B. Jahan, M. A. Ali, S. Alam, Z. R. Moni and M. S. Mian. 2014. Evaluation of fungicides against *Rhizoctonia oryzae-sativae*, the causal fungus of aggregate sheath spot of rice. Bangladesh J. Plant Pathol. 30(1&2): 7-12.

An *in-vitro* investigation was conducted to screen eight fungicides against *Rhizoctonia oryzae-sativae*, the causal fungus of aggregate sheath spot disease of rice. The fungicides were Amistar top 325 SC (Azoxystrobin), Carbendazim 50WC (Carbendazim), Differ 300EC (Difeconazole-propiconazole), Folicur 250EW (Tebuconazole), Monceren 250SC (Pencycuron), Mancodazim 63%+12% (Mancozeb-Carbendazim), Nativo 75WG (Trifloxystroin-Tebuconazole) and Propi 25EC (Propiconazole). Potato dextrose agar (PDA) was amended with the fungicides at 0.00, 0.25, 0.50, 1.00, 10.00 and 100.00 ppm. Amended PDA was poured into Petri dishes at 20 ml per dish and inoculated with mycelia blocks of *R. oryzae-sativae*. *Rhizoctonia oryzae-sativae* was found to be sensitive to all the fungicides tested. Carbendazim showed strong activity against the pathogen showing the lowest LD (lethal dose) 90 values (0.8 ppm) and LD 50 (0.1 ppm). Field trial was conducted to test the efficacy of all fungicides to control aggregate sheath spot of rice. Of them Carbendazim, Tebuconazole and Trifloxystroin-tebuconazole significantly reduced disease development and increased rice yield by 14.58, 11.88 and 9.79%, respectively.

Key words: *Rhizoctonia oryzae-sativae*, aggregate sheath spot, fungicides, rice

INTRODUCTION

Rice is the major food grain in Bangladesh. The requirement of rice is increasing every year in the country with the increasing population. In Bangladesh, rice yield per hectare is about 3.0 tons which is very low compared to global average (Anon. 2014). Low production of rice per unit area is attributed to various biotic and abiotic factors. Disease is one of the most important factors for low yield of rice. Among different diseases attacking rice, aggregate sheath spot caused by *Rhizoctonia oryzae-sativae* (teleomorph *Ceratobasidium oryzae-sativae*) is notable to cause considerable reduction in yield (Ou 1985). Aggregate sheath spot may cause 20% yield losses in Australia and 4 to 9% in Uruguay (Lanoiselet *et al*. 2005). Its incidence and severity began to increase with the introduction of semi-dwarf rice cultivars in different rice growing countries of the world (Gunnel and Webster 1984).

Different methods of plant disease control including chemical, physical, cultural measures are recommended to control aggregate sheath spot of rice. Globally chemical control represents the most widely used practice to reduce yield loss caused by *Rhizoctonia* spp. because no alternative satisfactory method is available. In California, Azoxystrobin has been registered for the control of rice aggregate sheath spot disease. In Australia, Pyraclostrobin tolclofos-methyl and Propiconazole were found effective fungicides for reducing mycelial growth of *R. oryzae-sativae in vitro* and disease severity under field conditions (Lanoiselet *et al*. 2005).

In Bangladesh, aggregate sheath spot was observed first time in the research farm of Bangladesh Rice Research Institute (BRRI) in both local and modern cultivars (Shahjahan *et al*. 1988). Presently, the disease is widely distributed throughout the country (Ali and Archer 2004). Sharma (2002) found that the growth of *R. oryzae-sativae* was completely inhibited at 1 ppm *in vitro* due to amendment of medium with fungicides.

The present piece of research was conducted to evaluate efficacy of eight fungicides *in-vitro* and under field condition to control *R. oryzae-sativae* isolated from aggregate sheath spot infected rice sheaths.

MATERIALS AND METHODS

In-vitro evaluation of fungicides

Eight fungicides namely Amistar top 325SC (Azoxystrobin), Carbendazim 50WC (Carbendazim), Differ 300EC (Difeconazole-propiconazole), Folicur 250EW (Tebuconazole), Monceren 250SC (Pencycuron), Mancodazim 63%+12% (Mancozeb-Carbendazim), Nativo 75WG (Trifloxystroin-

Tebuconazole) and Propi 25EC (Propiconazole) were tested in the experiment. Each of the fungicides was tested @ 0.00, 0.25, 0.50, 1.00, 10.00 and 100.00 ppm. Independent experiment was conducted to test each fungicide.

The test fungus, *R. oryzae-sativae* (Isolate MY-1) was obtained from Plant Pathology Division, BRRI, Gazipur. Poison food technique was followed in the experiment using potato dextrose agar (PDA) as the basal medium (Dhingra and Sinclair 1985, Ali and Archer 2003). Stock solution of 1000 ppm concentration was prepared in distilled water. Ingredients of PDA (Dhingra and Sinclair 1985) were mixed in 500 ml distilled water in conical flasks and appropriate quantity of stock solution was added to each flask to have required concentrations of individual fungicides. The mixture was thoroughly mixed and autoclaved at 120C under 0.80 kg/cm^2 pressure for 15 min. The amended PDA was dispensed into glass Petri dishes (90 mm dia) at 20 ml per dish. Plates received PDA without amendment served as control.

Five millimeter diameter mycelium blocks were cut from actively growing zones of 3-day-old *R. oryzae-sativae* culture which were used as inocula. The inocula were placed at the center of PDA plate keeping the up side down. The inoculated plates were arranged on the laboratory desks of Plant Pathology Division, BRRI, Gazipur. Three plates (replications) were used for each treatment. The plates were incubated at room temperature (30±2C). Diameter of colony of the fungus in plates containing amended PDA was measured when the mycelia in control plates reached the rim of the Petri dishes. Percentage of mycelium growth inhibition was computed based on colony diameter in control Petri dishes using the following formula:
Growth inhibition (%) =(C-T)/C X 100, Where, C= Growth of fungus in control plate, T = Growth of fungus in treated plates.

Field evaluation of fungicides

Based on findings of the *in vitro* test, five fungicides namely Carbendazim 50WC, Differ 300EC, Folicur 250EW, Nativo 75WG and Propiconazole 25EC were selected for their field evaluation against aggregate sheath spot during T. Aus seasons of 2012 and 2013 in the Research Farm of BRRI, Gazipur. BRRI dhan 48 was used in the experiment. Carbendazim, Propiconazole and Folicur were suspended in water at 0.90%. Suspension of Differ and Nativo was prepared in water at 0.18 and 0.036%, respectively. The experiment was laid out in randomized complete block design with three replications and 3m x 2m unit plot and plot to plot distance was 50 cm.

Seeds of variety BRRI dhan 48 were treated with hot water at 52C for 20 min and soaked in plain water for 24 hours for sprouting. Sprouted seeds were sown in tray to raise seedlings. Thirty days old seedlings were transplanted in the experimental plot at 3-4 seedlings per hill maintaining 15 cm x 20 cm spacing. Pure culture of *R. oryzae-sativae* (Isolate MY-1) was multiplied on PDA plate and incubated at ambient temperature. After 7 days of incubation the culture in a plate was divided into 8 equal sections, which were used as inocula to inoculate test plants according to Lanoiselet *et al.* (2001). For inoculation, each portion of the PDA culture was inserted at the base of each hill at maximum tillering stage. Nine hills in the middle of each unit plot were inoculated. The fungicides were suspended in water at required concentration and sprayed twice at 7 days starting from 15 days after inoculation. Standard agronomic practices were followed to grow the crop to maintain normal growth of plants.

Data on infected and healthy tillers, plant height and top most lesion height were recorded from which incidence, relative lesion height (RLH) and severity was computed according to Ahn *et al.* (1986) and Yoshimura (1954) as described below:

Relative lesion height (RLH) (%) = (Lesion height/Plant height) X 100.

Incidence (%) = Number of infected plant / Number of total plant checked X 100.
Severity (%) = $(3N_1+ 2N_2 + N_3 + 0N_4)/3N)$ X 100, where N_1= number of tillers with all four uppermost infected sheaths; N_2=number of tillers with the three uppermost infected sheaths; N_3=number of tillers with the two uppermost infected sheaths and N_4=number of tillers with the four uppermost sheaths disease free. The crops were harvested at 80% maturity. After threshing, the grains were sun dried to have 14% moisture content and grain yield per nine selected hills was recorded. The grain was expressed in kilogram per hectare. Sterility and thousand grain weight were also recorded.

Data analysis

Standard errors of means of three replications (mycelial inhibition) were calculated using computer software Stata (version 12). The LD_{50} and LD_{90} values were calculated using the software Origin 7.0. Data of field trial were analyzed using MSTAT-C computer program and mean separation was performed using least significant difference (LSD) test.

RESULTS AND DISCUSSION

In-vitro evaluation of fungicides

All fungicides at 0.10, 0.25, 0.50, 1.00, 10.00 and 100.00 ppm concentrations significantly (P=0.05) inhibited radial colony diameter of *R. oryzae-sativae* on amended PDA compared to control. The effectiveness of every fungicide to inhibit colony growth was corroborated with its dosage showing the highest inhibition at the highest concentration and the lowest at the lowest concentration. Differences in growth inhibition at different concentrations of Carbendazim, Differ, Folicur, Mancodazim, Nativo and Propiconazole were significant. The growth inhibition was 100% at 100, 10 and 1 ppm of Carbendazim, 100 and 10 ppm of Folicur and 100 ppm of Differ, Mancodazim, Nativo and Propiconazole. Gowth inhibition of 50.4-68.3, 32.9-98-4, 12.7-75.0, 13.5-88.9, 28.6-98.2 and 27.8-921% was obtained with Carbendazim, Differ, Folicur, Mancodazim, Nativo and Propiconazole, respectively. The growth inhibition ranged 12.3-54.4 and 16.7-94.1% at 0.1-100.00 ppm of Amistar Top and Monceren, respectively (Table 1).

Carbendazim effectively inhibited the fungal growth more than 50% at 0.1 ppm. It means the LD 50 (lethal dose 50) of the fungicide was 0.01 ppm. LD 50 of Nativo and Propiconazole was 0.40 ppm and that of Folicur was 0.50 ppm. Similarly, LD 90 of Carbendazim was the lowest at 0.8 ppm followed by Folicur (0.9 ppm), Nativo (5 ppm) and Propiconazole (8 ppm). LD 50 of Monceren was 14 ppm followed by Mancodazim (2.1 ppm) and Differ (1.1 ppm). LD 90 of Amistar top was 15 ppm and that of Differ was 13 ppm (Fig. 1 and 2).

Field evaluation of fungicides

Results on the efficacy of five fungicides tested under field conditions during T. Aus season of 2012 and 2013 to control aggregate leaf spot of rice are shown in Tables 2 and 3.

In both years of experimentation, significantly the highest relative lesion height (RLH), disease incidence (DI), disease severity (DS) and grain sterility, and the lowest thousand grain weight (TGW) and grain yield were recorded from diseased control. On the contrary, the lowest disease related parameters and the highest yield related parameters were recorded from healthy control.

Significant decrease in RLH, DI, DS and grain sterility, and increase in TGW over diseased control was achieved with the application of Carbendazim, Differ, Folicur and Nativo. The most effective one was Carbendazim followed by Folicur and Nativo to decrease disease related parameters and to increase grain yield significantly compared to control. Efficacy of Folicur and Nativo was statistically similar and significantly lower compared to Carbendazim. Effectiveness of Differ and Propiconazole was almost similar. Propiconazole was noted as the least effective fungicide to control aggregate leaf spot. In general, DI, RLH and DS were appreciably lower under all treatments in 2013 than 2012 (Tables 2 and 3).

In the present investigation, *R. oryzae sativae* reduced yield by 0.73 t/ha that counts to 13.44% in diseased control compared to healthy control during 2012. Application of Carbendazim, Folicur and Nativo increased rice yield by 10.63, 8.51 and 6.38%, respectively over control. In 2013, rice yield was also significantly affected by aggregate sheath spot disease. *Rhizoctonia oryzae sativae* reduced yield of BRRI dhan48 by 0.83 t/ha (14.74%) over healthy control. Yield recovery in Differ and Propiconazole treatments over disease control were not significantly different. In contrast, Carbendazim, Folicur and Nativo increased yield by 14.58, 11.88 and 9.79%, respectively over control. The disease incidence, RLH and disease severity were lower in 2013 compared to 2012.

Results of both *in vitro* and *in vivo* screening of fungicides against *R. oryzae sativae* and aggregate sheath spot disease caused by the fungus reveal that Carbendazim, Folicur and Nativo are most effective for controlling the disease. Effectiveness of fungicide tested in the present investigation to inhibit mycelial growth increased with increasing concentration from 0.1 to 100 ppm. Similar results have been reported by Lanoiselet *et al.* (2005). They found significant reduction of *R. oryzae-sativae* growth when concentration increased from 0.1 to10 µg/ml. Only Carbendazim inhibited 100% mycelial growth at 1.0 ppm and higher concentrations used. Similar result with Carbendazim was also reported by Sharma (2002) based on an *in vitro*. Amister top (Azoxystrobin), Monceren (Pencycuron) and Mancodazim were poorly effective in vitro and excluded from field test. Poor efficacy of Azoxystrobin on mycelial growth of *R. oryzae-sativae* and *R. solani* has also been reported by Lanoiselet *et al.* (2005) and Ali and Archer (2003). In the present experiment, Differ and Propiconazole showed better inhibition *in vitro* but did not perform well *in vivo* in field test. The findings are in agreement the findings of Carling *et al.* (1990) and Martin *et al.* (1984a, 1984b).

Differ and Propiconazole though decreased disease severity over control but yield was not increase accordingly. Similar result was also found by Lanoiselet *et al.* (2005). The maximum yield loss caused by the disease was 14.74% (on BRRI dhan48 in 2013) that relates with the findings of Lanoiselet *et al.* (2005) who found 20.30% yield loss in Australia. Based on findings of the present investigation it may be concluded that aggregate sheath spot disease causes considerable yield loss and Carbendazim, Folicur or Nativo may be recommended to control the disease and to save yield loss. However, before final recommendation benefit cost ration need to be determined.

Table 1. Effect of different concentrations of eight fungicides on *in vitro* mycelial growth of *R. oryzae-sativae*

Fungicide	Radial colony diameter (mm) at different concentrations (ppm)						
	0.00 (Control)	0.10	0.25	0.50	1.00	10.00	100.00
Amistar Top 325 SC	84.00	70.00[a] (16.70)[b]	69.33 (17.50)	68.67 (18.30)	25.67 (69.40)	21.67 (74.20)	5.00 (94.10)
Carbendazim 50WC	84.00	41.67 (50.40)	31.67 (62.30)	26.67 (68.30)	0.00 (100.0)	0.00 (100.00)	0.00 (100.00)
Differ 300EC	84.00	73.33 (12.70)	60.33 (28.20)	46.33 (44.90)	43.33 (48.40)	21.00 (75.00)	0.00 (100.00)
Folicur 250EW	84.00	56.33 (32.90)	52.00 (38.10)	44.67 (46.80)	1.33 (98.40)	0.00 (100.00)	0.00 (100.00)
Mancodazim 63%+12%	84.00	72.67 (13.50)	72.00 (14.30)	68.67 (18.30)	59.33 (29.40)	9.33 (88.90)	0.00 (100.00)
Monceren 250SC	84.00	73.67 (12.30)	72.67 (13.50)	72.33 (13.90)	57.00 (32.10)	41.67 (50.40)	38.33 (54.4)
Nativo 75WG	84.00	60.00 (28.60)	54.00 (35.70)	32.67 (61.10)	25.67 (69.40)	1.50 (98.20)	0.00 (100.00)
Propiconazole 25EC	84.00	60.67 (27.80)	59.00 (29.80)	33.67 (59.90)	26.00 (69.10)	6.67 (92.10)	0.00 (100.00)

[a]LSD (0.05) computed to compare among means of different treatments except control is 4.07, and LSD computed to compare all means with the means of control is 3.73.
[b]Figures within parenthesis are % inhibition in radial colony diameter.

Table 2. Evaluation of fungicides against aggregate sheath spot disease of rice during T.Aus 2012

Treatment	Relative lesion height (RLH %)	Disease index (DI%)	Disease Severity (DS%)	Thousand grain weight (g)	Sterility (%)	Yield (t/h)
Carbendazim	10.92 D	38.89E	45.40D (32.15)*	22.26 AB	21.02 D	5.2 B (10.64)**
Differ	27.39 B	62.22B	59.57B (10.97)	21.52 CD	27.31 AB	4.8 D (2.13)
Folicur	15.01 C	52.27D	51.21C (23.46)	21.92 BC	22.61 CD	5.1 BC (8.51)
Nativo	17.15 C	55.56C	53.61C (19.88)	21.85 BCD	25.58 BC	5.0 C (6.38)
Propiconazole	28.14 B	64.44B	62.24B (6.98)	21.24 DE	27.77 AB	4.7 D (0.00)
Diseased Control	31.08 A	67.78A	66.91A	20.80 E	29.76 A	4.7 D
Healthy Control	7.82 E	6.67F	18.68E	22.58 A	20.53 D	5.43 A

In a column, means followed by the same letter are not significantly different (P= 0.05).
Values within parentheses are *% reduction in disease severity and **% increase in yield over diseased control.

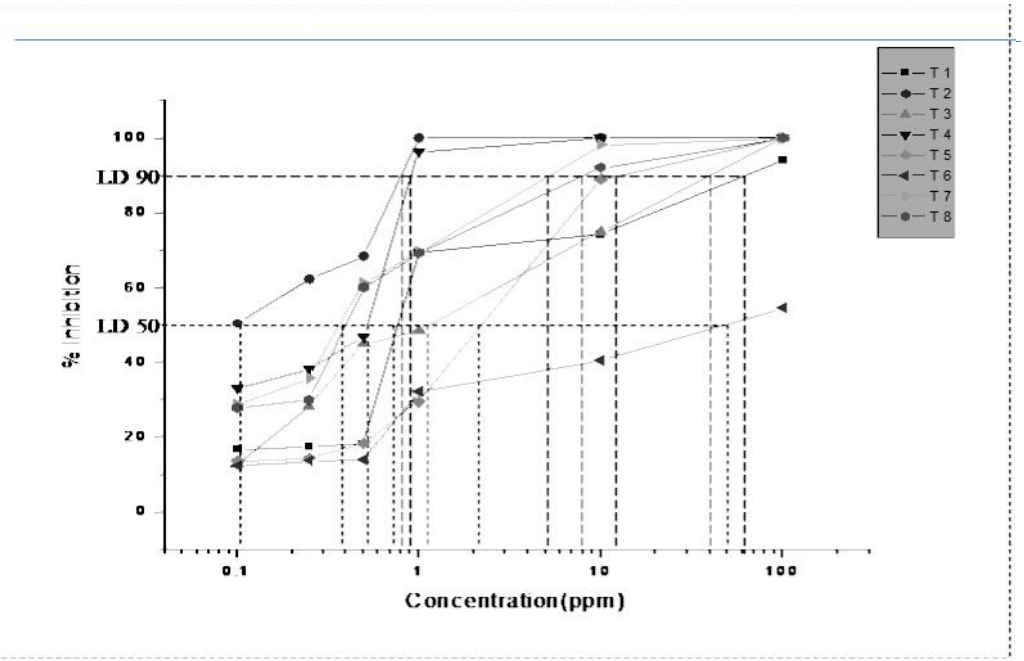

Figure 1. LD $_{50}$ and LD $_{90}$ values of different fungicides on growth inhibition of *R. oryzae-sativae* [T_1= Amister Top, T_2= Carbendazim, T_3= Differ, T_4= Folicur, T_5= Mancodazim, T_6= Monceren. T_7= Nativo and T_8= Propi]

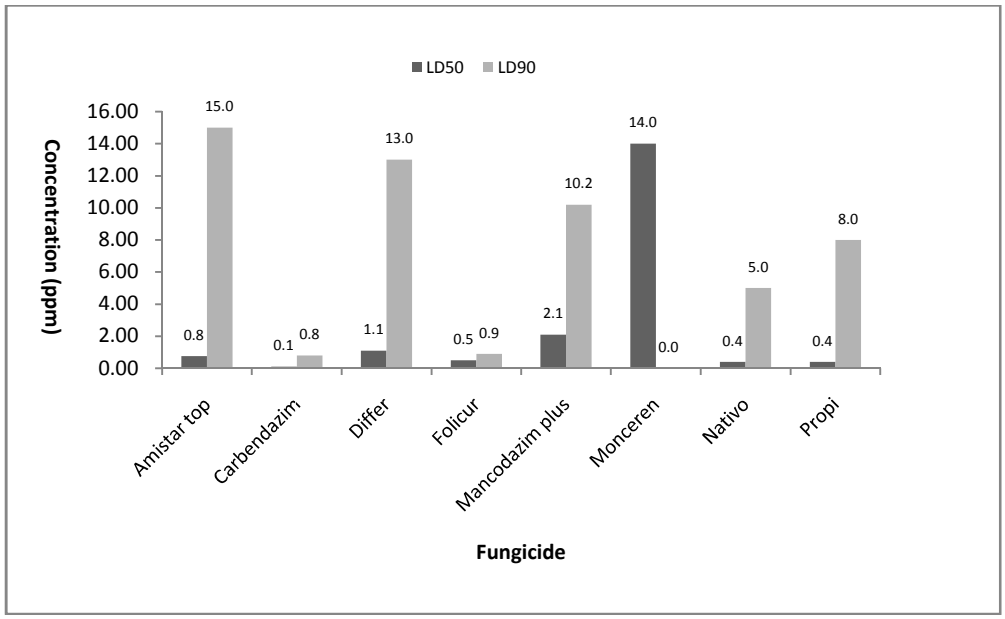

Figure 2. Values of LD 50 and LD 90 of different fungicides tested *in vitro* against mycelial growth of *R. oryzae-sativae*

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Table 3. Evaluation of fungicides against aggregate sheath spot disease of rice during T.Aus 2013

Treatment	Relative lesion height (RLH %)	Disease index (DI%)	Disease Severity (DS%)	Thousand grain weight (g)	Sterility (%)	Yield (t/h)
Carbendazim	6.93 E	12.22 D	40.79 E (38.14)	22.44 B	20.87 E	5.50 AB (14.58)
Differ	18.63 C	53.33 B	55.53 B (15.79)	21.80 D	26.01 BC	4.90 D (2.08)
Folicur	12.69 D	34.44 C	51.11 C (22.49)	22.17 C	22.40 DE	5.37 BC (11.88)
Nativo	14.12 D	37.78 C	47.29D (28.28)	22.09 C	23.56 CD	5.27 C (9.79)
Propiconazole	27.74 B	58.89 A	58.17 B (11.78)	21.37 E	26.50 AB	4.83 D (0.62)
Diseased Control	30.98 A	61.11 A	65.94 A	21.02 F	28.95 A	4.80 D
Healthy Control	5.48 F	4.44 E	14.50 F	22.80 A	18.12 F	5.63 A

In a column, means followed by the same letter(s) are not significantly different (P=0.05).
Values within parentheses are percent *% reduction in disease severity and ** Perent increase in yield over control.

LITERATURE CITED

Ahn, S. W., Delapena, R. C., Candole, B. L. and Mew, T. W. 1986. A new scale for rice sheath blight (ShB) disease assessment. Intl. Rice Res. Newsl. 11(6): 17.

Ali, M. A. and Archer, A. 2003. Evalution of some new fungicides against sheath blight disease of rice caused by *Rhizoctonia solani*. Bangladesh J. Pl. Pathol. 19 (1&2): 13-20.

Anonymous. 2014. Bangladesh rice knowledge bank. URL: http://knowledgebank-brri.org/. opened on 19-08-2014.

Carling, D. E., Helm, D. J. and Leiner, R. H. 1990. *In vitro* sensitivity of *Rhizoctonia solani* and other multinucleate and binucleate *Rhizoctonia* to selected fungicides. Plant disease 74: 860-863.

Dhingra, O. D. and Sinclair, J. B. 1985. Basic Plant Method. CRP Press, Inc. Boca Raton, Florida. 132 pp.

Gunnel, P. S. and Webster, R. K., 1984, Aggregate sheath spot of rice in California, Pl. dis. 68, 529-531.

Lanoiselet, V. M., Ash, G. J., Cother, E. J., Priest, M. J. and Watson, A. 2001. First report of *Waitea circinata* causing sheath spot and *Rhizoctonia oryzae-sativae* causing aggregate sheath spot on rice in south-eastern Australia. Plant Pathol. 30:369-370.

Lanoiselet, V.M., Cother E. J., Ash, G. J. and Harper, J. D. I. 2005. Yield loss in rice caused by *Rhizoctonia oryzae-sativae* in Australia. Australian Pl. Pathol. 34: 175-179.

Martin, S. B., Campbell, C. L., Lucas L. T. 1984a. Comparative sensitivity of *Rhizoctonia solani* and *Rhizoctonia*- like fungi to selected fungicides *in vitro*. Phytopathology 74: 778-781.

Martin, S. B., Campbell, C. L., Lucas L. T. 1984b. Response of *Rhizoctonia* blight of all tall fescus to selected fungicides in the glasshouse. Phytopathology 74: 782-785.

Ou, S. H. 1985. Rice Diseases. Commonwealth Mycological Institute, Kew, Surrey, England, pp. 272-286.

Shahjahan, A. K. M., Akanda, S. I., Ahmed, H. U. and Miah, S. A. 1988. Aggregate sheath spot caused by *Rhizoctonia oryzae-sativae* in BRRI farm. Bangladesh J. Plant Pathol. 4(1&2):146.

Sharma, N. R. 2002. Studies on the *Rhizoctonia* sheath disease complex of rice on Bangladesh. Ph. D. Dissertation, Department of Plant Pathology, Bangabandhu Sheikh Mujibur Rahman Agricultural University, Gazipur-1703. 79 pp.

Yoshimura, S. 1954. On the scale for estimating degree of severity of sheath blight by *Hypochus sasakii* Shirai in rice plants. Ann. Phytopathol. Soc. Japan. 9: 58-60.

POSTHARVEST DISEASES OF SELECTED FRUITS IN THE WHOLESALE MARKET OF DHAKA

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ABSTRACT

M. S. M. Chowdhury, N. Sultana, G. Mostofa, B. **K**undu and M. Rashid. 2014. Postharvest diseases of selected fruits in the wholesale market of Dhaka. Bangladesh J. Plant Pathol. 30 (1&2): 13-16.

A survey was conducted in three wholesale fruit market in Dhaka city during October 2012 to September 2013 to record postharvest diseases of mango, guava, papaya and jujube fruits collected from different parts of the country. The highest incidence and severity of postharvest diseases of the fruits were recorded in September, March, August and March, whereas the lowest incidence and severity were found in May, November, June and January, respectively. The post harvest diseases recorded during the survey from four fruits were anthracnose (*Colletotrichum gloeosporioides*), *Fusarium* rot (*Fusarium* sp.), *Rhizopus* rot (*Rhizopus stolonifer*), *Aspergillus* rot (*Aspergillus flavus*), stem end rot (*Botryodiplodia theobromae*), *Stemphylium* blight (*Stemphylium* sp.), scab (*Pestalotiopsis psidii*), *Alternaria* fruit rot (*Alternaria alternata*) and *Phomopsis* rot (*Phomopsis* sp.).

Key words: Post harvest diseases, fruits and wholesale market.

INTRODUCTION

In Bangladesh, different types fruits of diverse origins are cultivated in the country. Bangladesh produces 4.22 million tones of fruits annually from 0.15 million hectares of land (Anon. 2010). Mango, banana, jackfruit, pineapple, papaya, litchi, jujube and guava are the major fruits of the country, which can contribute in the economy and nutrition sector significantly (Rahim 2009). Approximately one third of all fresh fruits and vegetables are lost before it reaches consumers (Kader 2005). Estimated post-harvest losses in fresh fruits and vegetables are 5 to 35% in developed countries and 20 to 50% in developing countries (Kader 2002). Postharvest decay is the main reason for limiting the extension of storage and shelf life of fruits in Bangladesh. Main factors responsible for postharvest loss of fresh fruits are mechanical damage, spoilage by fungi, bacteria, insects, other microorganisms and physiological deterioration (Choudhury 2006). Mango, papaya (Ploetz *et al.* 1994), guava (Hossain and Meah 1992, Rahman *et al.* 2003) and jujube (Rai and Mamatha 2005) are attacked by a number of pathogens from bloom to harvest and in storage which cause considerably deteriorate the fruit quality. The post-harvest pathogens like *Colletotrichum gloeo-sporioides*, *Botryodiplodia theo-bromae*, *Alternaria*, *Phomopsis*, *Fusarium*, *Aspergillus*, *Stemphylium*, *Pestalotiopsis* attack the fruits from fruit set till harvest and cause considerable damage to fruit production and quality. The viable technologies need to be developed to reduce such postharvest losses caused by pre-harvest diseases. Before development of any management strategy, it is necessary to identify diseases.

In view of the facts, a survey was conducted to identify postharvest diseases of selected fruits occur in wholesale market.

MATERIALS AND METHODS

Four majors fruits of Bangladesh namely mango, papaya, guava and jujube were selected for the study. The survey was conducted in wholesale markets of Kawran Bazar, Zatrabari and Mirpur in Dhaka city during October, 2012 to September, 2013. Fruits were collected by the wholesaler from various parts of the country. One thousand infected fruit samples of each fruit were obtained from the wholesaler and brought to the Disease Diagnostic Laboratory of Plant Pathology Department, Sher-e-Bangla Agricultural University, Dhaka. The specimens were checked carefully and associated symptoms were recorded. The diseases were identified based on associated symptoms. Percentage of infected fruits was computed based on total number of fruits collected from the market according. Percent disease incidence (Rai and Mamatha 2005) and disease severity (Johnston 2000) were calculated using standard formulae. The formulae are shown below:

$$\text{Fruit infection \%} = \frac{\text{Number of diseased fruit}}{\text{Total number of fruits checked}} \times 100$$

$$\text{Disease incidence\%} = \frac{\text{Number of diseased fruit in a consignment}}{\text{Total number of fruits in same consignment}} \times 100$$

$$\text{Disease severity \%} = \frac{\text{Diseased area of fruit surface}}{\text{Total area of fruits surface}} \times 100$$

The collected data were analylized statistically for analysis of variance and means were compared using MSTAT-C computer program.

Causal fungi of the diseases were isolated and identified. For isolation of causal organism(s), 10 specimens of each fruits having similar symptoms were selected from collected samples from each location and put in sterilized polythene bags. The specimens were washed thoroughly under running tap water, surface sterilized with 5.00% NaOCl solution and rinsed in sterile water. The diseased parts were cut into small pieces and placed on three layers of moist blotting paper in a Petri dish and another set was placed in Petri dishes containing potato dextrose (PDA) medium. Both sets were incubated in an incubator at 22±2C for 7 days under 12 hour /12 hour alternate cycles of near ultra violet (NUV) light and darkness. After 8 days of incubation, the fungi grown from the specimens were transferred to PDA in Petri dishes and purified following hypal tip culture method. The pure culture was grown on PDA and identified based on the colony character and morphological characters of fruiting bodies, spores or conidia under a compound microscope. The pathogens thus recorded were identified following the keys of Mathur and Kongsdal (2003).

RESULTS AND DISCUSSION

During the survey, different postharvest diseases of fruit specimens collected from wholesale market of Dhaka were identified. The frequently occurring diseases were anthracnose (*Colletotrichum gloeosporioides*), stem end rot (*Botryodiplodia theobromae*) and *Fusarium* rot (*Fusarium* sp.) of mango; anthracnose, *Rhizopus* rot (*Rhizopus stolonifer*), *Fusarium* rot (*Fusarium* sp.), stem end rot and *Aspergillus* rot (*Rhizopus stolonifer*) of papaya; anthracnose and scab of guava; *Phomopsis* rot, *Stemphylium* blight, *Rhizopus* rot, anthracnose and *Alternaria* fruit rot of jujube. Nine pathogens viz. *Colletotrichum gloeosporioides, Fusarium* sp., *Rhizopus stolonifer, Aspergillus flavus, Botryodiplodia theobromae, Stemphylium* sp., *Pestalotiopsis psidii, Alternaria alternata* and *Phomopsis* sp. were identified from collected fruits of mango, papaya, guava and jujube (Table 1).

Incidence and severity of postharvest diseases of mango: Incidence and severity of anthracnose stem end rot and *Fusarium* rot of mango varied appreciably from one month to another. In case of anthracnose, the highest incidence and severity were recorded in the month of September 2013 whereas the lowest incidence and severity were recorded in the month of May 2013. Incidence and severity were the maximal in the month of September, 2013 whereas the lowest incidence and severity were found in the month of May 2013. In case of *Fusarium* rot the highest incidence and severity were observed in September 2013 and the lowest incidence was recorded in the month of June 2013 and the lowest severity was recorded in the month of May 2013 which was statistically similar to of July 2013 (Table 2).

Incidence and severity of postharvest diseases of papaya: Incidence and severity of anthracnose, *Rhizopus* rot, *Fusarium* rot, stem end rot and *Aspergillus* rot of papaya varied considerably from one month to of March followed by February 2013. The incidence and severity recorded in the month of February were significantly higher compared to other months except March 2013. Their lowest incidence as well as was found in the month of November followed by December 2012 (Table 3).

Table 1. Postharvest diseases of mango, papaya, guava and jujube collected from wholesale markets of Dhaka city

Fruits	Name of diseases	Causal fungi
Mango	Anthracnose	*Colletotrichum gloeosporioides*
	Stem end rot	*Botryodiplodia theobromae*
	Fusarium rot	*Fusarium* sp.
Papaya	Anthracnose	*Colletotrichum gloeosporioides*
	Rhizopus rot	*Rhizopus stolonifer*
	Fusarium rot	*Fusarium* sp.
	Stem end rot	*Botryodiplodia theobromae*
	Aspergillus rot	*Aspergillus* sp.
Guava	Anthracnose	*Colletotrichum gloeosporioides*
	Scab	*Pestalotiopsis psidii*
Jujube	Phomopsis rot	*Phomopsis* sp.
	Stemphylium Blight	*Stemphylium* sp.
	Rhizopus rot	*Rhizopus stolonifer*
	Anthracnose	*Colletotrichum gloeosporioides*
	Alternaria fruit rot	*Alternaria alternate*

Incidence and severity of postharvest diseases of guava: The highest incidence and severity of anthracnose and scab of guava were the maximal in the month of August followed by July and June 2013. Incidence anthracnose recorded in the month of June and July was statistically similar but significantly higher compared to August. Severity of anthracnose and incidence as well as severity of scab varied significantly from one month to another (Table 4).

Incidence and severity of postharvest diseases of jujube: The highest incidence and severity of *Phomopsis* rot, *Stemphylium* blight, *Rhizopus* rot, anthracnose and *Alternaria* fruit rot were recorded in the month of March followed by February and January 2013. Their differences were significant from one month to another except incidence of anthracnose which was statistically similar in the month of January and February (Table 5).

Fruit species under study had considerable amount of infection by pathogens viz. *Colletotrichum gloeosporioides, Fusarium* sp., *Rhizopus stolonifer, Aspergillus flavus, Botryodiplodia theobromae, Stemphylium* sp., *Pestalotiopsis psidii, Phomopsis* sp. and *Alternaria alternata*. This is no doubt an alarming situation for the economy of the country. Although chemicals are effective to reduce the incidence of postharvest diseases, this method is discouraged or even discarded in recent years due to economic, environment and health concerns. Interest in heat treatments waned with the development of chemical fumigants, which could be applied cheaply and easily. Similar diseases have also been recorded by other investigators. Dey *et al.* (2007) reported that anthracnose, stem end rot and fruit rot are common and destructive diseases of fruits in Bangladesh. Anthracnose was recognized as the most important postharvest fruit disease in the country (Rahman *et al.* 2003, Awasthi *et al.* 2005).

Table 2. Month wise incidence and severity of postharvest diseases of mango in wholesale market of Dhaka during May to September 2013

Month	Anthracnose		Stem end rot		*Fusarium* rot	
	Incidence (%)	Severity (%)	Incidence (%)	Severity (%)	Incidence (%)	Severity (%)
May	21.0c	20.0c	11.0c	14.0c	13.0b	13.0d
Jun	21.0c	22.0c	12.0c	18.0b	11.0c	15.0c
July	24.0c	22.0c	13.0c	18.0b	14.0b	13.0d
August	28.0b	25.0b	16.0b	19.0b	14.0b	18.0b
September	33.0a	32.0a	21.0a	31.0a	19.0a	26.0a

Values within the same column having a common letter(s) do not differ significantly (P=0.05).

Table 3. Month wise incidence and severity of postharvest diseases of papaya recorded from wholesale market of Dhaka during November 2012 to March 2013.

Month	Anthracnose		*Rhizopus* rot		*Fusarium* rot		Stem end rot		*Aspergillus* rot	
	Incidence (%)	Severity (%)	Incidence (%)	Severity (%)	Incidence (%)	Severity (%)	Incidence (%)	Severity (%)	Incidence (%)	Severity (%)
November	10.0d	15.0c	15.0d	20.0d	12.0d	14.0c	8.0e	11.0d	5.0d	6.0e
December	12.0c	17.0c	18.0c	22.0c	12.0d	14.0c	9.0d	12.0c	5.0d	7.0d
January	12.0c	17.0c	18.0c	22.0c	13.0c	14.0c	10.0 c	12.0c	6.0c	8.0c
February	15.0b	20.0b	20.0b	25.0b	15.0b	17.0b	12.0 b	15.0b	8.0b	10.0b
March	22.0a	30.0a	25.0a	32.0a	20.0a	23.0	15.0 a	18.0a	10.0a	15.0a

Values within the same column with a common letter(s) do not differ significantly (P0.05).

Table 4. Month wise incidence and severity of postharvest diseases of guava recorded from wholesale market of Dhaka during June to August 2013

Month	Anthracnose		Scab	
	Incidence (%)	Severity (%)	Incidence (%)	Severity (%)
June	3.0b	4.0c	11.0c	8.0c
July	4.0b	6.0b	16.0b	19.0b
August	11.0a	9.0a	21.0a	31.0a

Values within the same column with a common letter(s) do not differ significantly (P=0.05).

Table 5. Incidence and severity of diseases of jujube in wholesale market of during January to March 2013.

Month	*Phomopsis* rot		*Stemphylium* blight		*Rhizopus* rot		Anthracnose		*Alternaria* fruit rot	
	Incidence (%)	Severity (%)	Incidence (%)	Severity (%)	Incidence (%)	Severity (%)	Incidence (%)	Severity (%)	Incidence (%)	Severity (%)
January	8.0c	5.0c	6.0c	5.0c	5.0c	6.0c	2.0b	3.0c	10.0c	7.0c
February	10.0b	7.0b	8.0b	8.0b	8.0b	8.0b	3.0b	5.0b	15.0b	20.0b
March	15.0a	20.0a	10.0a	12.0a	12.0a	15.0a	10.0a	8.0a	20.0a	30.0a

Values within the same column with a common letter(s) do not differ significantly (P=0.05).

LITERATURE CITED

Awasthi, D. P., Sarkar, S., Mishra, N. K. and Kaisar, S. A. K. M. 2005. Disease situation of some major fruit crops in new alluvial plains of west Bengal. Environ. Ecol. 235(Special-3): 497-499.

Anonymous. 2010. BBS. Statistical Yearbook of Bangladesh, Bureau of Statistics, Ministry of Planning, Government of the People's Republic of Bangladesh, Dhaka.

Choudhury, M. L. 2006. Recent developments in reducing postharvest losses in the Asia-Pacific region. *In*: R. S. Rolle (ed) Reports of the APO Seminar on Reduction of Postharvest Losses of Fruit and Vegetables. Tokyo, Japan. pp. 15-22.

Dey, T., Ayub, K., Mortuza, G. and Sultana, N. 2007. Research on vegetables, spices and fruits disease management at Bangladesh Agricultural Research Institute. BARI, Joydebpur, Gazipur. pp. 72-85.

Hossain, M. S. and Meah, M. B. 1992. Prevalence and control of guava fruit anthracnose. Trop. Pest Manag. 38: 181-185.

Johnson, G. I., Coates, L. M., Cooke, A. W. and Wells, I. A. 1994. Post harvest control of mango diseases. Aust. Centre Intl. Agril. Res. pp. 70-71.

Johnson, G. I., Coates, L. M., Cooke, A. W. and Wells, I. A. 1994. Post harvest control of mango diseases. Aust. Centre Intl. Agril. Res. pp. 70-71.

Kader, A. A. 2002. Postharvest Biology and Technology: An Overview. *In*: Postharvest technology of horticultural crops. University of California, Division of Agric. and Nat. Resources. pp. 39-47.

Kader, A. A. 2005. Postharvest handling. *In*: The biology of horticulture, 2nd ed. John Wiley & Sons, Inc, Hoboken, NJ. pp. 379-407.

Mathur, S. B. and Kongsdal, O. 2003. Common Laboratory Seed Health Testing Methods for Detecting Fungi. Zürich: International Seed Testing Association (ISTA). 427 pp.

Ploetz, R. C., Zentmyer, G. A., Nishizima, W. T., Rohrbach, K. G. and Ohr, H. D. 1994. Compendium of Tropical Fruit Diseases, APS press, USA. pp. 34-41.

Rahim, M. A. 2009. Message from the president, Fruit Science Society of Bangladesh. *In*: International Conference on quality seed and food security, 17-19 February 2009. BAU, Mymensingh, Bangladesh.

Rahman, M. A., Ansari, T. H., Meah, M. B. and Yoshida, T. 2003. Prevalence and pathogenicity of guava anthracnose with special emphasis on varietal reaction. Pak. J. Biol. Sci. 6(3): 234-241.

Rai, V. R. and Mamatha, T. 2005. Seedling diseases of some important forest tree species and their management. *In*: Working papers of the Finish Forest Research Institute. 11 pp.

Ploetz, R. C., Zentmyer, G. A., Nishizima, W. T., Rohrbach, K. G. and Ohr, H. D. 1994. Compendium of Tropical Fruit Diseases, APS press, USA. pp. 34-41.

Rahim, M. A. 2009. Message from the president,

Fruit Science Society of Bangladesh. *In*: International conference on quality seed and food security, 17-19 February 2009. BAU, Mymensingh, Bangladesh.

Rahman, M. A., Ansari, T. H., Meah, M. B. and Yoshida, T. 2003. Prevalence and pathogenicity of guava anthracnose with special emphasis on varietal reaction. Pak. J. Biol. Sci. 6(3): 234-241.

Rai, V. R. and Mamatha, T. 2005. Seedling diseases of some important forest tree species and their management. *In*: Working papers of the Finish Forest Research Institute. 11 pp.

HEALTH AND QUALITY OF VEGETABLE SEEDS GROWN IN NORTHERN BANGLADESH

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(This is a part of MS thesis of the first author)

ABSTRACT

N. Khatun, I. Hossain and P. Dey. 2014. Health and quality of vegetable seeds grown in northern Bangladesh. Bangladesh J. Plant Pathol. 30(1&2):17-22.

Seed health and quality status of laffa (*Malva verticillata*), mustard (*Brassica* spp.), Indian spinach (*Basella alba*), jute (*Corchorus capsularis*), red amaranth (*Amaranthus tricolor*), swamp cabbage (*Ipomoea aquatica*), spinach (*Beta vulgaris* var. *Bengalensis*), cabbage (*Brassica oleracea* var. *capitata*), amaranth (*Amaranthus gangetica*), snake gourd (*Trichosanthes anguina*), cucumber var. khira (*Cucumis sativus*), cucumber var. shosa (*Cucumis sativus*), bottle gourd (*Lagenaria siceraria*), wax gourd (*Benincasa hispida*), sweet gourd (*Cucurbita moschata*), bitter gourd (*Momordica charantia*), ridge gourd (*Luffa acutangula*), radish (*Raphanus sativus*), carrot (*Daucus carrota* var. *sativa*), yard long bean (*Vigna unguiculata*), tomato (*Lycopersicon esculentum L.*), brinjal (*Solanum melongena*), cauliflower (*Brassica oleracea* var. *Botrytis*) and bean (*Phaseolus vulgaris*) grown in northern part of Bangladesh were tested. A total of ten seed-borne fungi were found to be associated with their seeds. The seed-borne fungi detected in the test were *Alternaria* spp., *Aspergillus flavus*, *Aspergillus niger*, *Curvularia* spp., *Fusarium oxysporum*, *Fusarium moniliforme*, *Penicillium* spp., *Phoma* spp., *Chaetomium* spp. and *Rhizopus* spp. Total seed-borne fungal infection was the highest in bean seed (159%) and the lowest in brinjal (3.5%). Germination of seeds ranged from 11 to 92% showing the highest germination in cucumber var. khira and the lowest in cabbage. Among the seed samples of 24 vegetables, the highest seedling vigor was found in snake gourd (3030.3) followed by bean (2433.4) and the lowest in cabbage (78.43).

Key words: Vegetable seed, seed health, germination, seedling vigour.

INTRODUCTION

Vegetables constitute an important group of crops in Bangladesh. Area under vegetables during 2010-11 was 298,380 hectares and total production was 105.68 million metric tons (Anon. 2012). Vegetables are important sources of vitamins like A, C, niacin, riboflavin and thiamine, and minerals such as calcium and iron. They contribute to intake of essential nutrients from other foods by making them more palatable. They provide dietary fiber necessary for digestion, and they are essential to maintain health and cure nutritional disorders (Mandol 2013).

In Bangladesh, production and availability of vegetables are not uniform round the year. Availability is plenty in winter but less available in summer. Bangladesh mainly produces two kinds of vegetables, namely, summer vegetables (brinjal, cucumber, pointed gourd, teasel gourd, Indian spinach, bitter gourd, etc.) and winter vegetables (cabbage, cauliflower, tomato, etc.) (Anon. 2012).

Among the agricultural inputs, seed is the most vital one. Healthy seeds are considered as the vital factor for desired plant population and good harvest. Seeds of vegetables are more vulnerable to the attack by pathogens and quickly deteriorate in storage. About 200 different seed-borne pathogens including more than 100 fungi have been reported to cause diseases in different vegetable crops in the world (Richardson 1990). Sultana (2009) estimated eight seed-borne fungi associated with the seeds of bottle gourd, sweet gourd, snake gourd, ridge gourd, cucumber, wax gourd and sponge gourd collected from Bangladesh Agricultural Development Corporation, a public seed producing organization, and private seed company. She found that *Aspergillus* spp. was highly prevalent in all the vegetable seeds ranging from 1.6-14%. A total of 18 seed-borne fungal pathogens have been reported from the seeds of those crops (Richardson 1990, Fakir *et al.* 1991, Islam 2005). Sowing of high quality healthy seeds is necessary to improve crop yields thus increasing food production. So, it is important to test the seeds for

disease organism before sowing. A substantial quantity of vegetable is grown in the northern part of Bangladesh, and therefore, information about health of vegetable seeds used by farmers of this region is crucial.

The present research project was undertaken to assess the health and quality of vegetable seeds collected from the northern part of Bangladesh.

MATERIALS AND METHODS

Collection of seed samples: Seeds of laffa (*Malva verticillata*), mustard (*Brassica* spp.), Indian spinach (*Basella alba*), jute (*Corchorus capsularis*), red amaranth (*Amaranthus tricolor*), swamp cabbage (*Ipomoea aquatica*), spinach (*Beta vulgaris* var. *Bengalensis*), cabbage (*Brassica oleracea* var. *Capitata*), amaranth (*Amaranthus gangetica*), snake gourd (*Trichosanthes anguina*), cucumber var. khira (*Cucumis sativus*), cucumber var. shosa (*Cucumis sativus*), bottle gourd (*Lagenaria siceraria*), wax gourd (*Benincasa hispida*), sweet gourd (*Cucurbita moschata*), bitter gourd (*Momordica charantia*), ridge gourd (*Luffa acutangula*), radish (*Raphanus sativus*), carrot (*Daucus carrota* var. *Sativa*), yard long bean (*Vigna unguiculata*), tomato (*Lycopersicon esculentum*), brinjal (*Solanum melongena*), cauliflower (*Brassica oleracea* var. *Botrytis*) and bean (*Phaseolus vulgaris*) were collected from northern regions of Bangladesh. The vegetables were grown in those areas and seeds were stored under farmers' conditions. Seed samples were obtained from the farmers just at sowing time. Collected seeds were poured in polyethylene bags and stored in a refrigerator at 4C.

Detection of seed-borne fungal pathogen: Working samples of 200 seeds of each crop were taken from the refrigerator and used for detection of seed-borne fungi. Standard blotter method as suggested by International Seed Testing Association (ISTA) was followed to detect seed-borne plant pathogenic fungi associated with the collected seeds (Anon. 1996). Working seed samples were surface sterilized in 1% chlorox for 5 minutes, rinsed in sterilized water for three times and plated on blotter in 90 mm glass Petri dishes. Each Petri dish received 10 to 25 seeds depending on seed size. After 10 days of incubation, fungi grown on the seeds were isolated and identified based on morphological characters observed under binocular compound microscope (Mathur *et al.* 1975).

Germination test: Standard procedure suggested by ISTA was followed for testing seed germination. Substrates, temperature and duration of test were the same as recommended by ISTA (Anon. 1976). Working samples of 400 seeds were used for germination test.

Seedling vigor test: The vigor of the seedlings was determined following the formula suggested by Baki and Anderson (1972) as shown below:

Vigor Index= (mean root length + mean shoot length) × percent seed germination

Experimental design and data analysis: All experiments were conducted in the laboratory of Seed Pathology Centre, Bangladesh Agricultural University, Mymensingh during the period from 2010 to 2011. The experiments were laid out following a completely randomized design with sufficient number of replicated plates.

Data were analyzed using MSTAT-C computer program. Analysis of variance and level of significance were done following Gomez and Gomez (1984). Mean differences were judged using Duncan's multiple range test.

RESULTS AND DISCUSSION

Prevalence of seed-borne fungi in leafy vegetable seeds: Prevalence of *Alternaria* spp., *Aspergillus flavus*, *A. niger*, *Curvularia* spp., *F. oxysporum*, *F. moniliforme*, *Penicillium* spp., *Phoma* spp., and *Rhizopus* spp. were 0.0-26.5, 0.0-12.0, 0.0-11.0, 0.0-10.0, 2.0-16.0, 1.0-26.5, 0.0-18.0, 0.0-2.0 and 0.0-20.0, respectively in seeds of different leafy vegetables. *Alternaria* spp. was found only in seeds of Indian spinach, red amaranth and laffa with the prevalence of 5.5, 1.0 and 0.5%, respectively. *Aspergillus flavus* was found in seeds of swamp cabbage, red amaranth, Indian spinach and laffa showing 12.0, 3.5, 1.0 and 1.0% prevalence, respectively. The maximum prevalence of *A. niger* was found in Indian spinach followed by swamp cabbage, amaranth and red amaranth. Seeds of other leafy vegetables were free from this fungus. *Curvularia* spp. was found in seeds of spinach, Indian spinach and amaranth at the prevalence of 10.0, 1.0 and 1.0%, respectively. Occurrences of two species of *Fusarium* were recorded from seeds of all leafy vegetables. In case of *F. oxysporum*, the maximum prevalence of 16.0% was observed in laffa followed by Indian spinach and spinach. The lowest prevalence of 2.0% was observed in red amaranth and amaranth. The maximum prevalence of *F. moniliforme* was recorded from seeds of Indian spinach and minimum was found in cabbage and amaranth seeds. The highest prevalence of *Penicillium* spp. was recorded in swamp cabbage but no prevalence of this fungus was recorded in laffa and jute seeds. The prevalence of *Phoma* spp. was recorded only in spinach. In case of *Rhizopus* spp., the prevalence was the highest in Indian spinach but the seeds of mustard, jute, red amaranth, swamp cabbage, spinach, cabbage and amaranth were free from this fungus (Table 1).

Table 1. Prevalence of seed-borne fungi on seeds of nine vegetables collected from northern region of Bangladesh

Leafy vegetables	Seed-borne infection (%)								
	Alternaria spp.	*Aspergillus flavus*	*Aspergillus niger*	*Curvularia* spp.	*Fusarium oxysporum*	*F. moniliforme*	*Penicillium* spp.	*Phoma* spp.	*Rhizopus* spp.
Laffa	0.5b	1.0c	0.0c	0.0b	16.0a	1.5b	0.0c	0.0b	1.0b
Mustard	0.0b	0.0c	0. 0c	0.0b	3.0bc	21.5a	4.0bc	0.0b	0.0b
Indian spinach	5.5a	1.0c	11.0a	1.0b	8.5b	26.5a	0.5 c	0.0b	20.0a
Jute	0.0b	0.0c	0.0c	0.0b	6.5bc	5.5b	0.0c	0.0b	0.0b
Red amaranth	1.0b	3.5b	0.5bc	0.0b	2.0c	5.5b	8.0b	0.0b	0.0b
Swamp cabbage	0.0b	12.0a	2.0b	0.0b	8.5b	3.0b	18.0a	0.0b	0.0b
Spinach	0.0 b	0.0c	0.0c	10.0a	3.5bc	26.0a	1.0c	2.0a	0.0b
Cabbage	0.0b	0.5c	0.0c	0.5b	3.5bc	1.0b	0.5c	0.0b	0.0b
Amaranth	0.0b	4.5b	1.0bc	1.0b	2.0c	1.0b	8.5b	0.0b	0.0b

Figures within the same column having a common letter (s) do not differ significantly (P=0.05).

Seed-borne prevalence of fungi varied with the variation in types of cucurbitaceous vegetables. Only 3% seeds of snake gourd and cucumber var. shosa yielded *Alternaria*. Seeds of other cucurbits were free from the fungus. In case of *A. flavus*, the maximum prevalence was found in bitter gourd followed by bottle gourd. The lowest prevalence was found in seeds of snake gourd. Other cucurbitaceous vegetables were free from *A. flavus.* The maximum prevalence of *A. niger* was recorded from bitter gourd seeds followed by snake gourd, ridge gourd and sweet gourd. Seeds of other four cucurbits were free from the fungus. The occurrence of *Curvularia* spp. was found only in seeds of cucumber var. shosa and cucumber var. khira. The prevalence of *F. oxysporum* was recorded from seeds of all of the cucurbits within the range of 0.5-13.0%. The highest prevalence was found in snake gourd and bottle gourd and the lowest in cucumber var. khira seeds. Seeds of cucumber var. khira, cucumber var. shosa and bitter gourd were free of *F. moniliformae*. Its prevalence was 9.0-40.0% in seeds of other cucurbits with the maximum in snake gourd and minimum in sweet gourd. Except cucumber var khira, 2.0-24.0% seeds of cucurbitaceous vegetables yielded *Penicillium* spp. The highest incidence was found in wax gourd, and the lowest in cucumber var shosa. *Phoma* was recorded from seeds of bottle gourd, wax gourd, sweet gourd and bitter gourd within the range of 5.0-15.0%. *Rhizopus* was recorded from only sweet gourd seeds showing 26.0% prevalence. *Chaetomium* spp. was found in seeds of bitter gourd and ridge gourd showing 34 and 29% prevalence, respectively (Table 2).

The prevalence of *A. niger* was the highest in radish seeds and lowest in carrot seeds (0.5%). The maximum seed-borne prevalence of *F. oxysporum* was recorded in radish and minimum in carrot. The incidence of *F. moniliforme* was found only in carrot seeds. *Penicillium* and *Rhizopus* were recorded in radish by 2.0% and 15.5%, respectively (Table 3).

Prevalence of fungal infection of seeds varied depending on different vegetables. The seed-borne fungus, *Alternaria* was found only in cauliflower and bean seeds. The prevalence of *A. flavus* was 51% in seeds of bean but seeds of tomato, brinjal and cauliflower were free from the fungus. The highest incidence of *A. niger* was recorded in yard long bean and lowest in tomato. *Curvularia* spp. was found in brinjal and bean seeds but the prevalence was only 1-2%. Seeds of yard long bean, tomato and cauliflower were free from *Curvularia* spp. The incidence of *F. oxysporum* was the highest in bean and lowest in brinjal (0.5%). In case of *F. moniliforme*, the maximum prevalence was recorded in bean seeds and minimum in tomato seeds. Prevalence of *Penicillium* spp. was 33% in yard long bean but seeds of tomato and brinjal were free from the fungus. The incidence of *Phoma* spp. was recorded only from tomato seeds, while *Rhizopus* was recorded only in yard long bean. Maximum mean association of seed-borne fungi was *A. flavus* followed by *Penicillium* spp., *A. niger*, *F. moniliforme*, *F. oxysporum*, *Rhizopus*, *Phoma*, *Alternaria* and *Curvularia* (Table 4).

Germination and seedling vigor: Germination of seeds of leafy vegetables varied from 11 to 90%. In case of cucurbit vegetable seeds, germination varied from 12 to 92% depending on vegetables. Germination ranged 57 to 76% in different root vegetables. Germination of other vegetable seeds varied from 15 to 72% depending on vegetable crops. Seedling vigor ranged 78.43 to 3030.3. The highest vigor index was found in snake gourd, while the lowest was recorded in cabbage. The highest germination of 92% was recorded in cucumber var. khira and the lowest in cabbage. Shoot length was the highest in bean and lowest in red amaranth. Root length was the highest in bottle gourd and lowest in red amaranth. There was variations in vegetables in respect of seed germination, shoot and root length, and vigor index (Table 5).

Table 2. Prevalence of seed-borne fungi associated with seeds of cucurbit vegetables collected from northern region of Bangladesh

Cucurbitaceous vegetables	Seed-borne infection (%)									
	Alternaria spp.	*Aspergillus flavus*	*Aspergillus niger*	*Curvularia* spp.	*Fusarium oxysporum*	*F. moniliforme*	*Penicillium* spp.	*Phoma* spp.	*Rhizopus* spp.	*Chaetomium* spp.
Snake gourd	3.0a	1.0c	11.0b	0.0c	13.0a	40.0a	9.0bc	0.0c	0.0b	0.0c
Khira	0.0b	0.0c	0.0c	4.0a	0.5e	0.0d	0.0d	0.0c	0.0b	0.0c
Shosha	3.0a	0.0c	0.0c	1.0b	4.0cde	0.0c	2.0d	0.0c	0.0b	0.0c
Bottle gourd	0.0b	17.0b	0.0c	0. 0c	13.0a	13.0bc	3.0cd	9.0b	0.0b	0.0c
Wax gourd	0.0b	0.0c	0.0c	0.0c	6.0bcd	17.0bc	24.0a	15.0a	0.0b	0.0c
Sweet gourd	0.0b	0.0c	4.0c	0.0c	2.0de	9.0cd	22.0a	5.0bc	26.0a	0.0c
Bitter gourd	0.0b	27.0a	18.0a	0.0c	8.0bc	0.0d	15.0b	7.0b	0.0b	34.0a
Ridge gourd	0.0b	0.0c	9.0b	0.0c	9.0ab	21.0b	15.0b	0.0c	0.0b	29.0b

Figures within the same column having a common letter (s) do not differ significantly.

Table 3. Prevalence of seed-borne fungi of root vegetables grown in northern Bangladesh

Root vegetables	Seed-borne infection (%)				
	Aspergillus niger	*Fusarium oxysporum*	*F. moniliforme*	*Penicillium* spp.	*Rhizopus* spp.
Radish	5.0a	12.5a	0.0b	2.0a	15.5a
Carrot	0.5b	2.0b	10.0a	0.0b	0.0b

Figures within the same column having a common letter (s) do not differ significantly.

Richardson (1990) reported prevalence of only *Alternaria amaranthi* in the seeds of *Amaranthus* spp. Peregrine *et al*. (1984) and Peregrine and Ahmed (1983) recorded *Aspergillus*, *Rhizopus*, *Cladosporium*, *Helminthosporium*, *Alternaria*, *Fusarium*, *Curvularia*, *Penicillium*, *Botrytis*, *Verticillium*, *Cylindrocephalum*, *Colletotrichum*, *Corynespora* and *Ascochytula* from seeds of cucurbits. Sultana (2009) reported *Aspergillus*, *Curvularia*, *Colletotrichum*, *Fusarium*, *Penicillium* and *Botrytis* as seed-borne fungi of vegetables. From the findings of the present study, it may be concluded that the seed samples of different vegetables collected from northern region of Bangladesh were of good quality except sweet gourd, bitter gourd, bean, cabbage and cauliflower.

The seed-borne fungi recorded in the present study have also been reported by other investigators. Hossain *et al.* (2014) reported seed-borne fungi of leafy vegetables collected from Mymensingh region of Bangladesh. Good number of seed-borne fungi of leafy vegetables has also been reported by Richardson (1990). *Alternaria*, *Aspergillus*, *Fusarium* and *Penicillium* were found in seeds of red amaranth. Among the seed-borne fungi of leafy vegetables, *Aspergillus*, *C. capsici*, *Fusarium*, *Penicillium* and *Rhizopus* were detected by Islam (2005).

Table 4. Prevalence of seed-borne fungi on seeds of other vegetables collected from northern part of Bangladesh

Other vegetables	Seed-borne infection (%)								
	Alternaria spp.	*Aspergillus flavus*	*Aspergillus niger*	*Curvularia* spp.	*Fusarium oxysporum*	*Fusarium moniliforme*	*Penicillium* spp.	*Phoma* spp.	*Rhizopus* spp.
Yard long bean	0.0c	25.0b	40.0a	0.0b	1.0bc	2.0c	33.0a	0.0b	13.0a
Tomato	0.0c	0.0c	0.5c	0.0b	4.5b	0.5c	0.0c	6.50a	0.0b
Brinjal	0.0c	0.0c	0.0c	1.0ab	0.5c	2.0c	0.0c	0.0b	0.0b
Cauliflower	1.0b	0.0c	0.0c	0.0b	1.5bc	18.5b	13.0b	0.0b	0.0b
Bean	2.0a	51.0a	31.0b	2.0a	12.0a	33.0a	28.0a	0.0b	0.0b

Figures within the same column having a common letter (s) do not differ significantly.

Table 5. Germination and seedling vigor of different vegetable seeds collected from northern region of Bangladesh

Name of vegetables	Germination (%)	Shoot length (cm)	Root length (cm)	Vigor index
Laffa	52.00±2.5	6.15	2.85	468.00
Mustard	79.00±1.5	12.60	3.05	1236.35
Indian spinach	36.00±2.0	9.65	2.90	451.80
Jute	90.00±1.0	5.40	2.00	666.00
Red amaranth	59.00±3.0	3.90	1.25	303.85
Swamp cabbage	83.00±3.0	11.50	5.40	1402.70
Spinach	67.00±1.5	10.20	3.10	891.10
Cabbage	11.00±1.5	5.19	1.94	78.43
Amaranth	41.00±4.5	4.05	1.50	227.55
Snake gourd	74.00±5.0	28.70	12.25	3030.30
Cucumber (khira)	92.00±3.0	19.35	5.70	2304.60
Cucumber (shosa)	86.00±4.0	20.40	5.90	2261.80
Bottle gourd	76.00±7.0	19.65	12.35	2432.00
Wax gourd	30.00±3.0	12.10	5.50	528.00
Sweet gourd	58.00±6.0	20.95	6.00	1563.10
Bitter gourd	12.00±1.0	20.50	8.50	348.00
Ridge gourd	26.00±12.0	17.35	6.90	630.50
Radish	76.00±2.0	13.750	5.05	1428.80
Carrot	57.00±4.0	5.65	2.25	450.30
Yard long bean	58.00±11.0	26.80	11.70	2233.00
Tomato	56.00±4.5	8.10	2.80	610.40
Brinjal	72.00±7.5	4.50	2.70	518.40
Cauliflower	15.00±1.5	8.10	1.55	144.75
Bean	46.00±4.0	41.10	11.80	2433.40

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LITERATURE CITED

Anonymous. 1976. International Rules for Seed Testing. International Seed- Testing Association, Seed Sci. Tech. 24:39-42.

Anonymous. 1996. International Rules for Seed Testing. International Seed- Testing Association. Seed Sci. Tech. 24:39-42.

Anonymous. 2012. The Yearbook of Agricultural Statistics of Bangladesh. Bangladesh Bureau of Statistics, Ministry of Planning, Government of the Peoples' Republic of Bangladesh, Dhaka. p. 305.

Baki, A. A. and Anderson. J. D. 1972. Physiological and biological deterioration of seeds. *In* Seed Biology, Academic Pess, New York. 2:283-315.

Fakir, G. A., Rahman, G. M. M., and Islam, M. F. 1991. Occurrence of diseases on lentil and country bean germplasm and their reaction to the selected major pathogens. Proc. Res. Prog. Bangladesh Agric. Univ. 5:1-10.

Gomez, K. A. and Gomez, A. A. 1984. Statistical Procedures for Agricultural Research. 2nd Edn. John Willey and Sons., New York. 97-111 pp.

Hossain, I., Dey, P. and Dilruba, K. 2014. Quality of vegetable seeds collected from Mymensingh region in Bangladseh. *Intl. J. Appl. Sci. Biotech*. 2(1):103-108.

Islam, M. R. 2005. Studies on the health and seedling diseases of some leafy vegetables. M.Sc. Thesis, Department of Plant pathology, BAU, Mymensingh. 67 pp.

Mathur, S. K., Mathur S. B. and Neergard, P. 1975. Detection of seed borne fungi in sorghum and location of *Fusarium moniliforme* in the seed. Seed Sci. Tech. 3:683-690.

Mandol, P. 2013. Study of marketing system and shelf life extension of brinjal. Department of Horticulture. Bangladesh Agricultural University, Mymensingh. pp.1-3.

Peregrine, W. T. H., Ahmad, K. B. and Momin, M. 1984. Controlling anthracnose in water melon. World Crops. 36(5):184-185.

Peregrine, W. T .H. and Ahmad, K.B. 1983. Chemical and cultural control of anthracnose (*Colletotrichum lagenarium*) in water melon. Trop. Pest Mang. 29(1): 42-46.

Richardson, M. J. 1990. An annotated list of seed-borne diseases. 4th ed. The International Seed Testing Association, Switzerland. 338 pp.

Sultana, L. 2009. Assessment of health status of Truthfully labeled seeds (TLS) in the markets of Bangladesh. M. Sc. Ag. Thesis, Department of Plant Pathology, Bangladesh Agricultural University, Mymensingh. 39 pp.

EVALUATION OF FUNGICIDE FOR SEED TREATMENT TO CONTROL STEM CANKER AND BLACK SCURF DISEASE (*RHIZOCTONIA SOLANI*) OF POTATO

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[Part of Ph.D dissertation of the first author]

ABSTRACT

M. M. Rahman, M. A. Ali[2], M. U. Ahmad and T. K. Dey. 2014. Evaluation of fungicide for seed treatment to control stem canker and black scurf disease (*Rhizoctonia solani*) of potato. Bangladesh J. Plant Pathol. 30(1&2):23-27.

A field experiment was conducted to evaluate the effectiveness of Amistar 325 SC (Azoxystrobin) @ 0.10%, Provax-200 (Carbixin+Thiram) @ 0.20%, Boric acid at 3.00% and Bavistin 50 WP (Carbendazim) @ 0.10% as pre-plant seed tuber treating fungicides against stem canker and black scurf disease (*Rhizoctonia solani*) of potato. Infested seed tubers were treated by spraying with the fungicidal suspensions over the tuber surface. Two additional treatments, one with infested tubers and other one with healthy seed tubers were maintained in the experiment for comparison. Efficacy of Provax to reduce disease incidence and to increase plant growth and yield was not considerable. Pre-plant treatments of infected seed tubers with Amistar 325 SC at 0.10%, Boric acid at 3.00% and Bavistin 50 WP at 0.10%, and planting of healthy seed tubers increased germination by 9.17, 7.86, 7.47 and 8.56%; number of stem per hill by 7.54, 13.07, 12.56 and 9.30%; plant height by 2.31, 10.42, 9.23 and 10.02; and tuber yield by 64.15, 22.06, 20.06 and 7.23% respectively over control-2. The treatments reduced incidence of stem canker and black scurf by 59.45, 54.05, 48.65 and 51.35 and disease severity, in terms of PDI, was reduced by 65.87, 56.09, 48.77 and 48.77% respectively over control-2. Results of the present investigation reveal that Amistar 325 SC is the most effective fungicide followed by Bavistin 50 WP to improve plant growth, tuber yield and to reduce incidence and severity of stem canker and black scurf disease of potato.

Key words: Fungicide, Seed treatment, stem canker, *Rhizoctonia solani*, control

INTRODUCTION

Potato (*Solanum tuberosum*) is a leading staple food crop of the world and it ranks next to wheat and rice. In Bangladesh, it is the first leading vegetable crop and grown in almost all areas of the country. During 1999-2000, the production of potato was 2.93 million metric tons from 0.243 million hectares of land. In Bangladesh, average tuber yield is lower compared to other potato growing countries of the world. The major constraint of potato production is prevalence of epidemic diseases and lack of supply of quality seed potato to the farmers. In Bangladesh, a total of 39 diseases of potato have been recorded (Ali and Khan 1990). One of the major diseases is stem canker and black scurf caused by *Rhizoctonia solani* (Kuhn). It is the most common and widespread disease throughout the country (Ali and Dey 1994).

Rhizoctonia solani is a soil-borne fungus and causes 'black scurf' on potato tubers and canker on stems. Severe stem canker can kill shoots and delay crop emergence. On the surface of infected tubers the fungus forms dark brown to black sclerotia which vary in size. The sclerotia are resting structures that allow the fungus to survive for long periods under stress conditions. Tuber infested with sclerotia is a significant problem in seed tubers as the sclerotia can act as a primary source of infection for the new plant. The tuber borne nature of *R. solani* is an important component of the disease cycle. Many researchers from different countries reported efficacy of pre-plant seed tuber treatment with fungicides to control stem canker and black scurf diseases (Bolkan 1976, Wenham *et al.* 1976, De and Sengupta 1992, Akhilash *et al.* 1996, Anon. 2005, Himel *et al.* 2007, Djelbali and Bathassel (2010).

Development of effective as well economic control methods against stem canker and black scurf disease is essential in Bangladesh. One of the transmission methods of the disease is infected seed tubers bearing sclerotia of *R. solani*. Effective seed treatment methods can minimize the primary spread of the disease from seed to progeny tubers; thereby reduce incidence and severity of the disease. So, the present investigation was undertaken to evaluate three promising fungicides for pre-plant treatment of potato seed tubers to control stem canker and black scurf under Bangladesh conditions.

MATERIALS AND METHODS

Fungicides tested in the experiment were Amistar 325 SC (Azoxystrobin) @ 0.10%, Provax-200 (Carbixin+ Thiram) @ 0.2%, Boric acid @ 3.0% and Bavistin 50 WP (Carbendazim) @ 0.10%. Two controls consisted of healthy tubers (control-1) and infected tubers carrying sclerotia (control-2) were used in the experiment for comparison. Infected as well as healthy

seed tubers of variety Diamant were collected from Breeder Seed Production Centre of Bangladesh Agricultural Research Institute (BARI) at Panchagarh, Bangladesh. The fungicides were suspended in tap water at desired concentrations. The infected seed tubers were spread on the floor and the fungicidal suspensions were sprayed over the seed tubers for treatment. For proper coverage of the surface, the tubers were rotated frequently at the time spray. The treated tubers were air dried overnight and stored in a room until planting.

The experiment was conducted at the Tuber Crops Research Sub-Centre (TCRSC) of BARI in Bogra during 2008-2009. The experiment was laid out in a randomized complete block design with four replications. The unit plot size was 3.0 m x 3.0 m. Block to block and plot to plot distances were 100 cm and 50 cm, respectively. Row to row distance was 60 cm. Recommended doses of fertilizers and manures were applied as suggested by Tuber Crops Research Centre, BARI, Gazipur (Anon. 2005). Cowdung was incorporated to the soil during land preparation at the rate of 10 t/ha. Urea, Triple super phosphate (TSP), Muriate of potash (MOP), Gypsum, Zinc sulphate and Boric acid were applied at the rate of 360, 220, 250, 120, 14 and 6 kg per hectare, respectively. The entire amount of TSP, MOP, Gypsum, Zinc sulphate, Boron and half of urea were applied at the time of final land preparation. Treated seed tubers were planted on 29 November 2008 maintaining 25 cm seed to seed distance. The 2nd half of urea was applied at 30 days after planting (DAP). Weeding was done at 25 DAP and earthing up was done at 30 DAP. Irrigation was applied at 20 and 40 DAP. The insecticides Dursban (0.5%) and Admire (0.1%) were applied respectively, to control cutworms and aphids. Secure (0.1%) was sprayed at 10 days interval as preventive measures against late blight disease of potato. The crop was harvested on 28 February 2009.

Data on germination, number of stem per hill, plant height were recorded from the field. After harvest, data on healthy and infected tubers were collected. The black scurf infected tubers were separated into three groups such as russet, deformed and sclerotia infected. Number and weight of tubers under each group were recorded. Number and weight of healthy tubers harvested from each plot were also recorded.

Disease related data such as disease incidence, percent disease index (PDI) were recorded. To record disease incidence, 20 plants were randomly selected from each unit plot at 70 DAP, uprooted carefully, washed with running tap water and checked for infection. Numbers of infected and healthy plants were counted and percent disease incidence was calculated based on total number of plants checked. At 70 DAP, severity of stolon infection was indexed on a 0-6 indexing scale (Dey 2010), where 0= No symptom on stolon, 1 = minute brown lesion on stolon or root, 2 = moderately brown lesion on stolon and curling tendency on central leaf, 3 = stolon symptom discolored accompanied by brown discoloration on roots, 4 = brown to black discoloration on underground parts, tissue discoloration and curling of growing leaves, 5 = profuse emerging of auxiliary leaves, leaf size reduced markedly and pale green on leaf margin, and 6 = production of aerial tuber with green colour. Twenty plants in each plot were randomly selected and uprooted carefully from soil, washed with water. The plants were checked individually and severity of stolon infection was indexed. Number of stem canker infected plants under each grade (0-6 scale) was recorded and the percent disease index (PDI) was calculated using standard formula as shown below:

$$PDI = \frac{\text{Class frequency}}{\text{No. of plants checked X highest score}} \text{ X } 100$$

The black scurf infected tubers were separated into russet, deformed and sclerotia infected tubers. Number and weight of tubers under each symptom category per plot were recorded. Collected data were analyzed statistically following MSTAT-C computer program Differences among treatment means were determined following Duncan's Multiple Range Test using the same computer program.

RESULTS AND DISCUSSION

Germination

Range of germination of potato seed tubers under different treatments including two controls (1 and 2) was 85.54 to 93.38%. The lowest germination was recorded when infected seed tubers were planted (control-2) without treatment, which was statistically similar to the treatment with Provax-200 @ 0.20%. The highest germination was recorded from the treatment with Amistar 325 SC at 0.10%, which was statistically similar to Boric acid at 3.00, Bavistin 50 WP at 0.10% and planting of healthy seed tubers. The four treatments increased germination by 9.17, 7.86, 7.47 and 8.56%, respectively and the increase was significant compared to control-2 (Table 1).

Stem number and plant height

Under different treatments, number of stem per hill varied from 3.98 to 4.50 and plant height varied from 57.08 to 63.88 cm. The lowest number of stem per hill and plant height was found under control-2. Treatments with Amistar 325 SC at 0.1%, Boric acid 3.00% and Bavistin 50 WP at 0.1% and planting of healthy tubers increased stem number/hill by 7.54, 8.79, 13.07, 12.56 and 9.30%; and plant height by 2.31, 11.91, 10.42, 9.23 and 10.02, respectively over control-2. The increase in both the parameters was not significant compared to control-2 (Table 1).

Incidence and severity of disease

The highest incidence of 46.26% stem canker and black scurf disease of potato was recorded from control-2. Treatments of infected seed tubers with fungicides and use of healthy tubers caused significant reduction in disease incidence. The reductions obtained with Amistar 325 SC at 0.10%, Boric acid at 3.0%, use of healthy seeds, Bavistin at 0.1% and Provax-200 at 0.20% were 59.45, 54.05, 51.35, 48.65 and 45.95% over control-2. The maximum reduction was obtained with Amistar 325 SC which was statistically similar to two treatments with Bavistin and healthy seeds (Table 1).

The severity, in terms of PDI values of stem canker and black scurf disease of potato was 46.26% under control-2. Treatment of seed tubers with fungicides and planting healthy tubers caused significant reduction in the parameter within the ranged 48.77-65.87 over control-2. However, the efficacy of all fungicidal treatments and planting healthy seeds to reduce disease severity was statistically similar (Table 1).

Number of russet, deformed and sclerotia infested tubers

Per plot maximum number of russet, deformed and sclerotia infested tubers were 26.50, 10.50 and 65.25, respectively, when infected seed tubers were planted without any treatment (control-2). All fungicidal treatments of seed tubers and use of healthy seeds significantly reduced number of russet, deformed and sclerotia infected tubers over control-2. Treatments with Amistar 325 SC at 0.10%, Provax-200 at 0.20%, Boric acid 3.00 and Bavistin at 0.10% and use of healthy seeds reduced number of russet tuber by 64.15, 21.70, 51.89, 41.51 and 61.32%, deformed tubers by 40.48, 21.43, 66.67, 66.67 and 33.33% and sclerotia infested tubers by 55.56, 42.15, 48.28, 48.28 and 91.15%, respectively over control-2 (Table 2).

Weight of russet, deformed and sclerotia infested tubers

The maximum of 580 kg/plot of deformed tubers were harvested from the plot planted with black scurf infected seed tubers without any treatment (control-2). Application of Amistar 325 SC at 0.1%, Provax-200 at 0.2%, Boric acid at 3.0% and Bavistin at 0.1% reduced the parameter to 37.93–450 kg/plot. The reductions under above five treatments were 37.93, 37.93, 39.66 and 39.66%, respectively and the reduction was significant compared to control-2. Difference in weight of deformed tubers harvested from plots planted with infected tuber without treatment (control-2) and healthy tubers was not significant (Table 2).

Maximum of 1280 and 4800 kg/plot of russet and sclerotia infested tubers were found under control-2. Both the parameters were significantly reduced over control due to treatments with fungicides and use of healthy seeds. Treatment of black scurf infected seed tubers with Amistar 325 SC at 0.1%, Provax-200 at 0.2%, Boric acid at 3.0%, and Bavistin at 0.1% and use of healthy seeds caused 45.31, 39.06, 58.59, 58.59 and 35.16% reduction in russet tubers and 62.71, 37.93, 22.41, 45.80, 57.71 and 91.67% reduction in sclerotia infested tubers, respectively (Table 2).

Table 1. Effect of seed tuber treatment with three fungicides on germination, plant growth and incidence of black scurf disease of potato caused by *Rhizoctonia solani*

Fungicide with dose (%)	Germination (%)	Stem/ hill	Plant height (cm)	Incidence (%)	Severity (PDI)
Amistar 325 SC (0.1%)	93.38 a* (9.17)**	4.28 a (7.54)	58.40 a (2.31)	18.75 c (59.46)	5.83 b (65.87)
Provax-200 (0.2%)	86.46 b (1.08)	4.33 a (8.79)	63.88 a (11.91)	25.00 b (45.95)	9.58 b (43.91)
Boric acid (3.0%)	92.26 a (7.86)	4.50 a (13.07)	63.03 a (10.42)	21.25 bc (54.05)	7.50 b (56.09)
Bavistin 50 WP (0.1%)	91.93 a (7.47)	4.48 a (12.56)	62.35 a (9.23)	23.75 bc (48.65)	8.75 b (48.77)
Healthy tuber (Control-1)	92.86 a (8.56)	4.35 a (9.30)	62.80 a (10.02)	22.50 bc (51.35)	8.75 b (48.77)
Infected tuber (Control-2)	85.54 b	3.98 a	57.08 a	46.25 a	17.08 a

*Means within the same column with a common letter(s) do not differ significantly (P=0.05).

**Values within parentheses are % increase in first 3 columns and percent decrease in last 2 columns over control-2.

Tuber yield

The lowest tuber yield of 16.32 t/ha was harvested from plots planted with black scurf infected seed tubers without treatment (control-2). Treatment of black scurf infected seed tubers with Amistar at 0.1%, Provax-200 at 0.2%, Boric acid at 3.0%, Bavistin at 0.1% and use of healthy seeds increased tuber yield by 64.15, 12.13, 22.06, 20.06 and 7.23% over control-2. The increase was significant in case of Amistar @ 0.1%, Boric acid @ 3.0% and Bavistin @ 0.1% (Table 2).

Results of the present investigation reveal that pre-plant treatment of seed tubers with the fungicides Amistar 325 SC at 0.1%, Provax-200 at 0.2%, Bavistin 50 WP 3.0% and Bavistin 50 WP at 0.1% is effective to control stem canker and black scurf disease of potato. The treatments increased germination, plant growth and tuber yield but decreased incidence and severity of the disease. The fungicides also reduced number and weight of russet, deformed and sclerotia infested tubers. Planting of healthy seed tubers without treatment gave similar results as recorded from fungicidal treatments. Among the fungicides Amistar 325 SC (0.1%) was the most effective one followed by Bavistin 50 WP (1.0%) to improve plant growth parameters, tuber yield and to reduce incidence and severity of stem canker and black scurf disease of potato. The findings of the present investigation are in agreement with the findings of other researchers (Bolkan 1976, Wenham *et al.* 1976, De and Sengupta 1992, Akhilash *et al.* 1996, Anon. 2005, Himel *et al.* 2007, Djelbali and Bathassel (2010). Djelbali and Bathassel (2010) agreed that seed tuber treatment and in furrow application of Amistar gave effective control of stem canker and black scurf of potato.

Table 2. Effect of seed tuber treatment with fungicides on black scurf of potato (*Rhizoctonia solani*) to reduce diseased tubers.

Fungicide with dose (%)	Number of infected tuber/plot			Weight of infected tuber (g/plot)			Yield (t/ha)
	Russet	Deformed	Sclerotia infested	Russet	Deformed	Sclerotia infested	
Amistar 325 SC 0.1%	9.50d* (64.15)**	(6.25b) (40.48)	29.00c (55.56)	700 bc (45.31)	360 c (37.93)	1790.0d (62.71)	20.43a (64.15)
Provax-200 (0.2%)	20.75b (21.70)	8.25ab (21.43)	37.75b (42.15)	780b (39.06)	450bc (22.41)	2600.0b (45.80)	18.30abc (12.13)
Boric acid (3.0%)	12.75cd (51.89)	3.50c (66.67)	33.75bc (48.28)	530c (58.59)	350c (39.66)	2030.0cd (57.71)	19.92ab (22.06)
Bavistin 50 WP (0.1%)	15.50c (51.89)	7.75b (66.67)	35.50b (48.28)	750bc (58.59)	380c (39.66)	2480.0bc (57.71)	19.68ab (20.06)
Healthy tuber (Control-1)	10.25d (61.32)	7.00b (33.33)	5.75d (91.19)	830b (35.16)	670a (15.52)	400.0c (91.67)	17.50bc (7.23)
Infected tuber (Control-2)	26.50a	10.50a	65.25a	1280a	580ab	4800.0a	16.32c

*Means within the same column with a common letter(s) do not differ significantly (P=0.05).
**Values within parentheses are per cent reduction over control-2.

LITERATURE CITED

Akhilesh, S., Tyagi, P. D. and Singh, A. 1996. Evaluation of chemicals dip treatments for the control of black scurf of potato. Himachal J. Agril. Res. 22(1-2): 142-145.

Ali, M. S. and Dey, T. K. 1994. Pathological research on tuber crops in Bangladesh. *In:* Proc. Workshop on Tubers Crops on Transfer of Technology of CDP crops under Research Extension LinkageProgramme. pp. 159-165.

Anonymous. 2005. Annual report 2004-2005. Tuber Crops Research Centre, Bangladesh Agricultural Research Institute (BARI), Gazipur, 75 pp.

Bolkan, H. A. 1976. Seed tuber treatment for the control of black scurf disease of potatoes. New-Zealand J. Exp. Agril. 4 (3): 357-361

De, B. K and Sengupta, P.C. 1992. Evaluation of different chemicals to control of black scurf of potato in the West-Bengal Plains. J. Indian Pot. Assoc. 19: (1): 81-83.

Dey, T. K. 2010. Tuber crops diseases-their identification, method of recording, rating scale and grading system. Oilseed Res. Center, Bangladesh Agril.Res. Inst. Gazipur. pp.28-31.

Djelbali, N. and Bethassel, T. 2010. Field study of the relative susceptibility of eleven potato *(Solanum tuberosum L.)* varieties and the efficacy of two fungicides against *Rhizoctonia solani*. Tunisia Crop Prot. 29:9.

Himel, M. K., Akand, A. M. and Khair, A. 2007. Controal of *Rhizoctonia solani* infecting potato. Abstract of Annual Botanical Conference, March 7-9, 2008. Jahangir-nagar University, Savar, Dhaka.

Wenham, H. T., MacKintosh, B. L. and Bolkan, H. A. 1976. Evaluation of fungicides for control of potato black scurf disease. New-Zealand J. Exp. Agric. 4 (1): 97-101.

PLANT PARASITIC NEMATODES ASSOCIATED WITH BRINJAL (*SOLANUM MELONGENA*) IN SOME AREAS OF BANGLADESH

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ABSTRACT

Sharmin Afrose, I. H. Mian, M. Z. Alam and Rayhanur Jannat. 2014. Plant parasitic nematodes associated with brinjal (*Solanum melongena*) in some areas of Bangladesh. Bangladesh J. Plant Pathol. 30(1&2):27-35.

A survey was conducted during 2011-2014 to identify plant parasitic nematodes associated with rhizosphere soils and roots of brinjal (*Solanum melongena*). Soil and root samples were collected from brinjal fields in the districts of Gazipur, Tangail, Kishoregonj, Manikgonj and Mymensingh. The nematode was extracted following Baermann Funnel technique. Temporary mounts of the extracted nematodes were prepared and morphological characters and necessary morphometrics were determined uncer a compound microscope. Based on morphological characters and morphometrics, nematodes associated soil and roots of brinjal were identified up to genera or species level using appropriate key books. The plant parasitic nematodes associated with rhizosphere soils and roots of brinjal were *Aphelenchus avenae*, *Cephalenchus emerginatus*, *Helicotylenchus indicus*, *Hoplolaimus indicus*, *Meloidogyne incognita*, *Pratylenchus pratensis*, *Pratylenchus zeae*, *Tylenchorhynchus claytoni*, *Tylenchus* sp., *Ditylenchus melongena*, *Xiphinema americanum* and *Zygotylenchus guevarai*. Except *M. incognita* other nematode species of brinjal have been reported first time from Bangladesh. Morphological characteristics have been described.

Key words: Nematode, parasite, brinjal

INTRODUCTION

Brinjal or eggplant *(Solanum melongena)* is a popular vegetable and important cash crop in Bangladesh. Per hectare yield of the crop is very low in comparison to China, India and world average (Anon. 2004a). Brinjal is attacked by more than 40 species of plant parasitic nematodes throughout the world (Romero and Arias 1969, Patel *et al*. 2007, Haidar *et al.* 2008, Zakir and Bora 2009, Anwar and McKenry 2012).

In India, *Ditylenchus melongena*, *Helicotylenchus*, *Hoplolaimus*, *M. incognita*, *Pratylenchus*, *Radopholus*, *Rotylenchus*, *Rotylenchulus reniformis*, *Trichodorus* and *Tylenchorhynchus* have been identified as pests of brinjal (Patel *et al*. 2007, Verma *et al*., 2013). In Pakistan, *Helicotylenchus* spp., *M. incognita*, *M. javanica*, *Meloidogyne* spp., *Pratylenchus* spp. and *Xiphinema* spp. have been recorded as major nematode pests of brinjal (Anwar and Chaudhry 1973, Shakeel *et al.* 2012). A study was conducted by Audamou *et al.* (2013) in Niger and identified *Meloidogyne*, *Tylenchorhynchus*, *Helicotylenchus*, *Scutellonema*, *Rotylenchulus*, *Pratylenchus* and *Xiphinema* as nematode pests of brinjal. From Syria, Haidar *et al.* (2008) reported ten plant parasitic nematodes of brinjal namely *Meloidogyne*, *Pratylenchus*, *Paratylenchus*, *Tylenchus*, *Rotylenchus*, *Helicotylenchus*, *Tylenchorhynchus*, *Longidorus*, *Ditylenchus* and *Xiphinema*. Available reports reveal that plant parasitic nematodes cause considerable damage to brinjal in India (Anowar *et al*. 1986, Patel *et al*. 2007, Verma *et al*. 2013), Pakistan (Shakeel *et al.* 2012), Niger (Audamou *et al.* 2013), Nigeria (Bhatti *et al.* 2013) and Spain (Romero and Arias 1969).

Reports on nematode pests of brinjal are scanty in Bangladesh. Occurrence of only root-knot nematode (*Meloidogyne* spp.) has so far been recorded in the country (Timm and Ameen 1960, Mian 1986, Mian and Zahid 1986). More than 50 species of plant parasitic nematodes have been identified on different crop plants in Bangladesh (Sam 1979, Mian 1986, Rahman and Mian 2010). Reports so far discussed above indicate that the plant parasitic nematodes identified on various crops in Bangladesh may also attack brinjal. Under the above circumstances, a research project was undertaken to identify plant parasitic nematodes associated with soils and roots of brinjal in some intensively brinjal growing areas of Bangladesh.

MATERIALS AND METHODS

The survey was conducted in the districts of Kishoregonj, Mymensingh, Tangail, Manikgonj and Gazipur in Bangladesh during of 2011-2014. Brinjal is extensively grown in those areas. Rhizosphere soil and root samples of brinjal were collected from the fields of the selected districts from 20 places of each field at a depth of 15-20 cm. All 20 soil samples of each field was mixed together to have a composite sample. About 2.5 kg subsamples were drawn from each composite sample. Nematodes were extracted from soil and root samples following Baermann funnel method (Mian 1994). Temporary mounts of the extracted nematode were prepared and observed under a compound light microscope. Morphological characters and morpho-metrics of individual nematode specimens were recorded. For identification, related sets of CIH

Descriptions of Plant-parasitic nematodes and other available key books, journal articles and internet descriptions as well as pictures were used. Female perineal pattern was studied to identify the species of root-knot nematodes (Taylor and Sasser 1980, Eisenback 2010).

RESULTS AND DISCUSSION

Nematodes identified

During the survey, a total of 12 nematode species under 11 genera were associated with rhizosphere soils and roots of brinjal. The nematode species were *Aphelenchus aveneae*, *Cephalenchus emerginatus*, *Ditylenchus melongena*, *Helicotylenchus indicus*, *Hoplolaimus indicus*, *Meloidogyne incognita*, *Pratylenchus pratensis*, *Pratylenchus zeae*, *Tylenchorhynchus claytoni*, *Tylenchus* sp., *Xiphinema americanum* and *Zygotylenchus guevarai*.

Aphelenchus aveneae

For identification of *Aphelenchus aveneae*, CIH Description of Plant-parasitic nematodes was consulted (Hooper 1974a). Female body is cylindrical with slight arcuate when killed by heat, maximum wide at vulval point, gradually tapering towards ends, posterior body behind vulva is nrrower than anterior region. Morphometrics of female are L = 120-127 µm, a = 30.4, b = 5.7, V = 70-75%. Stylet length is 13.5-15.5 µm. Head is bluntly rounded and not offset from the body. Vulva is a transverse slit. At mid-body lateral field is widest. Esophagus with cyndrical procorpus, median bulb well developed squarish, conspicuous, rectangular to oval shape having refractive cresentic valve plate. Ovary is monodelphic, prodelphic and outstretched. Esophageal lumen of dead nematode is strait and dorsal esophageal gland orifice is not visible because it opens in side the median bulb. Tail is 1.77 times of anal body diameter and tail tip is bluntly rounded (Plate I).

Cephalenchus emerginatus

The nematode was identified based on characteristics described by Hooper (1974b). Body of adult female is relatively slender, small, only 500-580 µm long, vermiform, narrow only a little anterior but posterior to the bulb it tapers sharply forming a narrow long tail with a pointed terminus. Cuticle is fairly thick and annules slightly coarse. Cephalic framework mode rately set off and round. Lateral field has six lines. The stylet is relatively long compared to body length with rounded knobs. Stylet is well developed with round knobs. Esophagus is tylenchoid, basal bulb is not overlapping the intestine. Vulva is a transverse slit at 70-72% of body from the anterior end. Ovary is monodelphic prodelphic and outstretched (Plate II).

Helicotylenchus indicus (**Spiral nematode**)

The species of spiral nematode was identified consulting morphological and morphometrical characteristics described by Siddiqi (1972). Adult female body is vermiform with tapering towards both terminuses, length is 660-850 µm, a=28-33, b = 5.3-6.2. Posterior region of the female body forms a coil on thermal death. Annulations are distinct, not interrupted by lateral lines. Lateral fields are about one-sixth of body width, continuing to tail terminus, marked with four incisures. Lip region is hemispherical, cephalic framework is well developed, head slightly set off and conoid. Stylet is well developed, anteriorly tapering and knob is rounded. DGO is situated at less than half of stylet length behind the knob. Esophagus is tylenchoid with oval shape median bulb. Basal bulb overlaps anterior part of intestine from ventral side. Tail is, conoid with a narrow terminus and mucronate. Ovary is didelphic amphidelphic and outstretched. Vulva is transverse slit like at 65-70% of the body from anterior**.** Male is similar to female except less coiled body shape. Tail is elongate with mucron. Spicule is 20-25 µ (Plate III).

Ditylenchus melongena (**Stem nematode**)

The nematode genus was identified consulting the description of Hestling (1974) and Haider *et al*. (2008). Females are slender transparent, almost strait when relaxed or killed by heat. Labial framework is not well developed; lip region flattened and cap-like, not offset. Stylet is 111-13 µm long with distinct basal knobs. Lateral fields with four incisures. Tail terminus is sharply pointed. Measurements of female showed that L = 1020-1400 µm, a = 24.0, b = 5.2-39.5, V = 76 - 87%. Dorsal esophageal grand orifice (DGO) is very close to stylet knob. Median bulb is fusiform. Basal bulb overlaps intestine mostly dorsally. Ovary is monodelphic and prodelphic. Tail shape is more or less conoid with pointed tip (Plate IV).

Tylenchorhynchus claytoni (**Stunt nematode**)

Descriptions of the species used for identification by Loof (1974b) were also used in the present investigation to identify the nematode. Adult female body is cylindrical, with weakly curve ventrally when relaxed. The body is more tapering towards posterior end. Body length is 680- 750 µm with coarse transverse striae. Lateral field consists of 4 longitudinal lines. Lip region of the nematode is rounded and offset by slight constriction. Stylet is slender with round basal knobs. Median bulb is oval; its valve located halfway along esophagus. Isthmus is long and narrow, terminal bulb pyriform, with conspicuous dorsal gland nucleus. Vagina is less than one-half body width long. Ovary is didelphic, amphi-delphic, outstretched. Rectum is about one-half o anal body width. Tail is tapering towards end (Plate V).

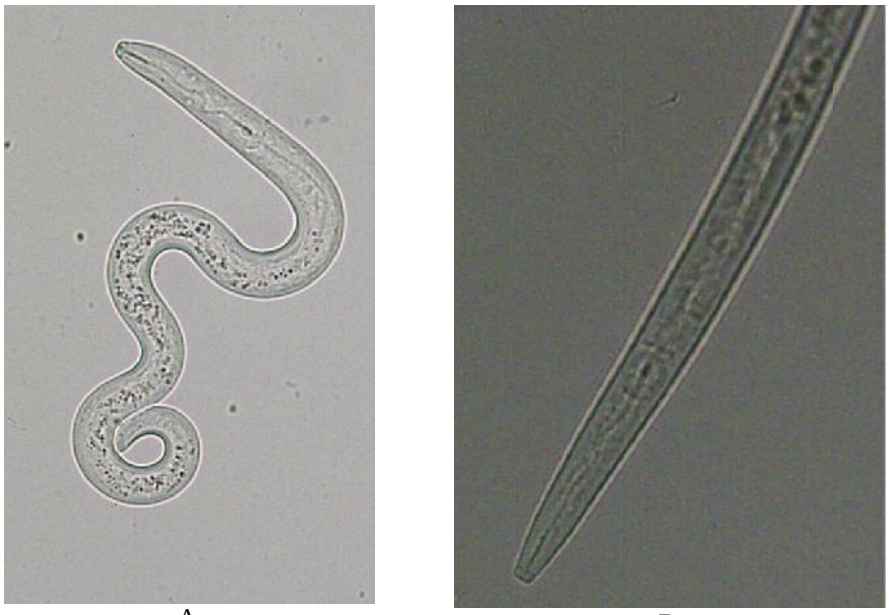


Plate I. Microphotograph of *Aphelenchus aveneae* [A:Adult female; B: Anterior virew of female]

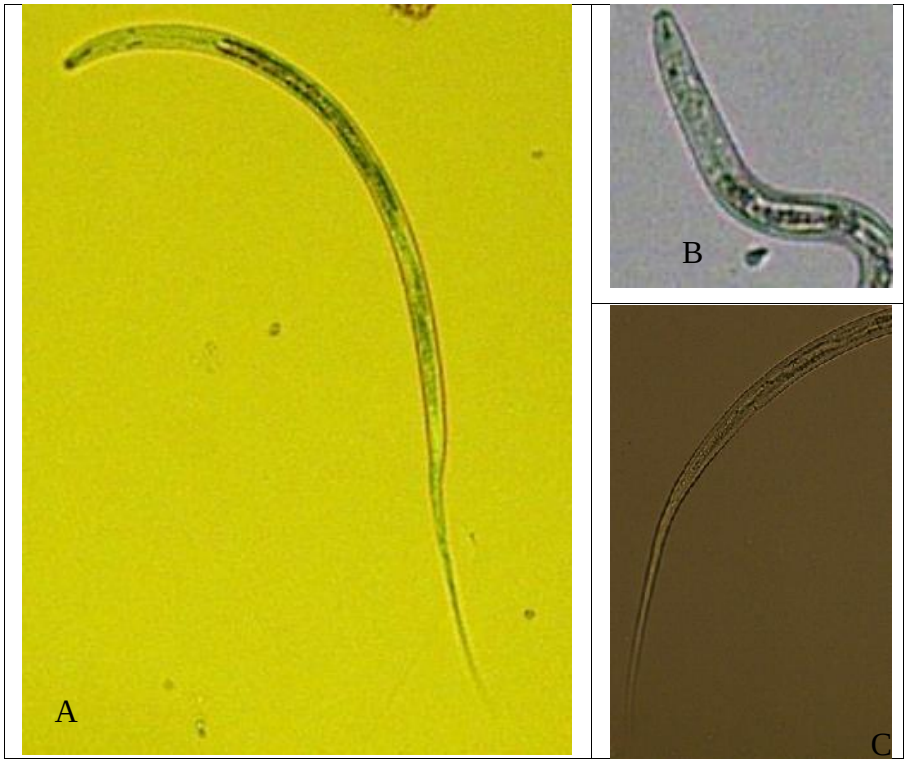
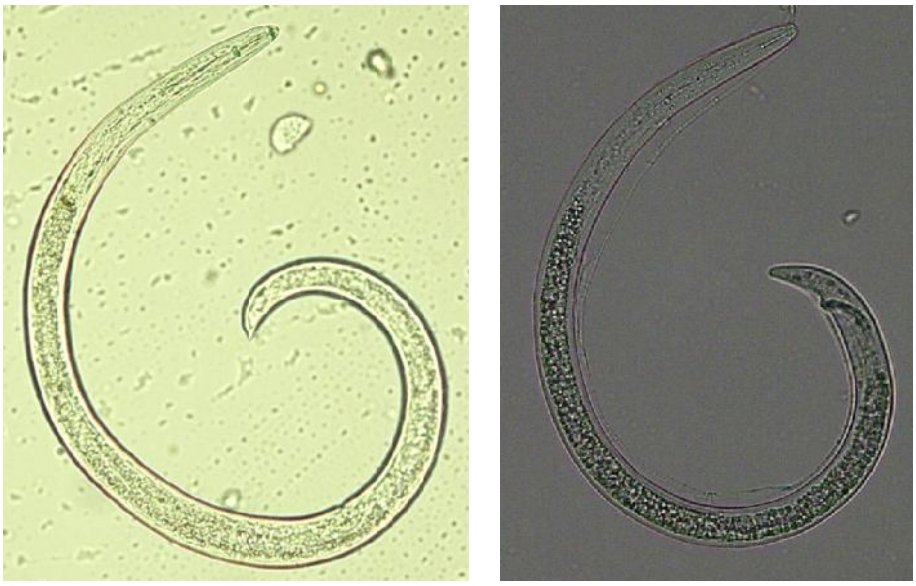


Plate IV. Microphotgraph of *Cephalenchus emerginatus* [=Adult female, B = Anterior gerion and Posterior part of the nematode].



A B

Plate III. Microphotographs of *Helicotylenchus dihystera* [A: Adult Female and B: Adult Male]

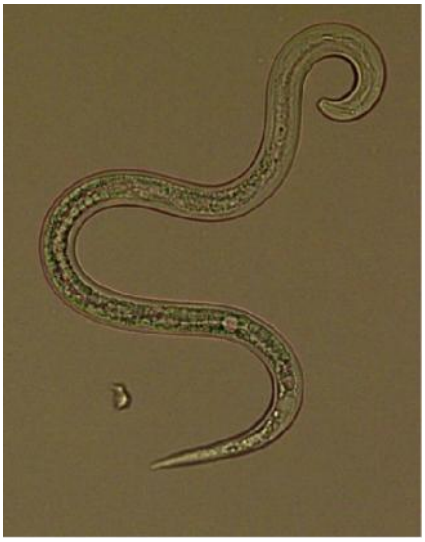


Plate IV. Microphotographs showing adult female of *Ditylenchus melongena*

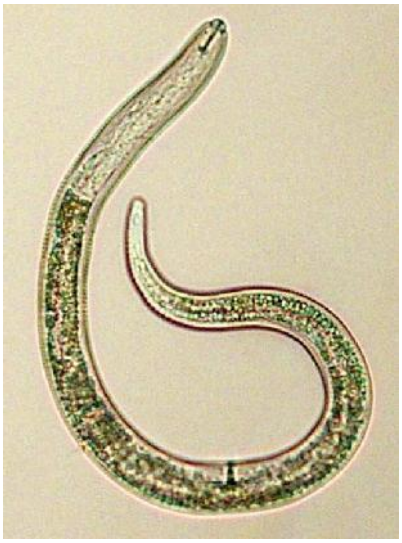


Plate V. Microphotographs showing adult female of *Tylenchorhynchus claytoni*

Hoplolaimus indicus **(Lance nematode)**

Hoplolaimus indicus was identified consulting the description of Khan and Chowla (1975). Adult female body is vermiform, cylindrical, tapering towards both ends, especially towards the anterior end and an open C-shaped after death due to heat. Female body length is 1020-1302 µm, 'a' and 'b' values are 23-28 and 7.1-8.2, respectively. Esophagus tylenchoid, stylet strong with backwardly projected knobs. Ovary is didelphic and outstretched. Head is distinctly set off. Tail is shorter (22.1-22.4 µm) than anal body diameter (25.5-26.5 µm) with round tip. Anus is circular, 2.0-2.2 µm diameter, present on 18-20th annule from tail terminus. Annules on the tail terminus are anastomosed irregularly. Lateral field represented by two very indistinct striae, annulation continues round the body. Vulva is conspicuous, transverse slit, 10.0-12.0 µm wide with two distinct lips (Plate VI).

Plate VI. Microphotographs of *Hoplolaimus galeatus* [A = Adult female, B = Adult male, C = Anterior region and D = Posterior region showing short hemispherical tail]

Root-knot nematode (*Meloidogyne incognita*).

The morphological and morphometrical characteristics of egg, larvae, the mature females, and perineal pattern of mature female were the basis of identification (Taylor and Sasser 1980, Barker *et al*. 1985, Eisenback 2010). Matured eggs are elliptical in shape containing J_1 larvae (Plate VII A). Second stage larvae with prominent fat droplets, 210 to 3950 µm long, head region not offset. Stylet 15 to 25 µm in length, basal knobs set off, rounded to transversely elongated. Tail 25 to 65 µm in length, tip rounded and lateral lines four with incisures (Plate VII B). Third (J_3) and fourth stage (J_4) and immature females were found in the root galls of brinjal (Plate VII C). The perineal pattern is oval to rounded 90 to 100 µm in length and 82 to 100 µm in width. Generally high with dorsal arch. Anus anteriorly located 15 to 30 µm distance from vulval slit and anus located 14 to 20 µm distance from tail terminus. Lateral lines are not pronently demarcated by breaks and forked striae. Striae are distinct and wavy. Lateral fields weakly demarcated and not disrupted by the lateral lines. Tail terminus is smooth (Plate VII D).

Xiphinema americanum (**Dagger nematode**)

The genus was identified comparing the features described by Siddiqi (1973). Female body is vermiform, slightly tapering towards both ends, cuticle finely annulated. Adult female is 1400 to 2000 µm long. The most important identifying character is long stylet with flanges extension and more than 100 µm long. Lip region is hemispherical in shape and slightly expanded. Tail rounded with greater curvature dorsally and terminus conoid. It is needle like and about 72 µm long. Esophagus is divided into two parts having narrow corpus and cylindrical basal region. Vagina at right angles to body axis covering 40% of the body width. Vulva is situated at 50-54% of body length (Plate VIII).

Pratylenchus pratensis (**Lesion nematode**)

The species of lesion nematode was identified consulting the description of Loof (1974a). Female body is vermiform, more or less straight when killed by heat or on relax and 50-68 µm long; 'a' = 26-35, 'b' = 6.5-7. Stylet has three well separated knobs. DGO is present at less than one-third of stylet length behind the knobs. Esophageal lumen is not straight, median bulb is broadly oval in shape. Basal bulb overlaps the intestine ventratally. Excretory pore is located at anterior to the esophago-intestinal junction. Ovary is monodelphic prodelphic; uterus with large oval to rectangular spermatheca filled with sperm received from the male partner. The vulva is transverse and about 72-77% from the head. Post-uterine sac is slightly longer than body width. Tail with 25-28 annules and the terminus is slightly curve ventrally (Plate IX).

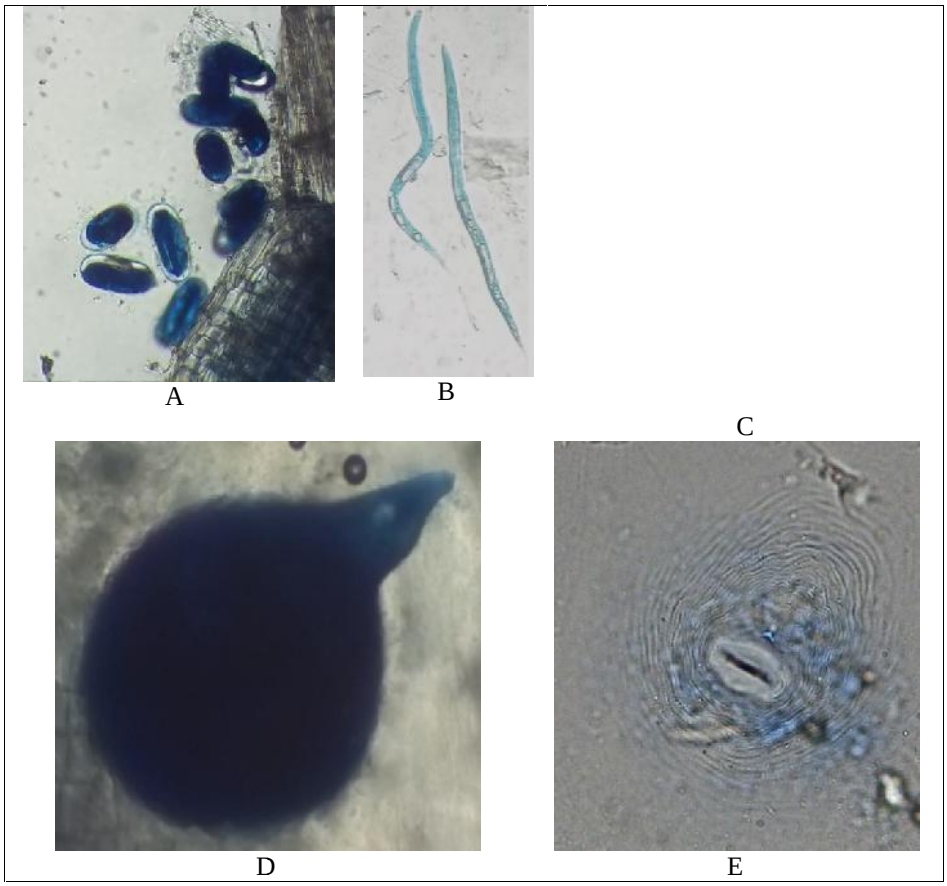


Plate VII. *Meloidogyne incognita* (A: Egg, B: 2nd stage larvae, C: 3rd stage larva, D: Egg laying female and E: perennial pattern)

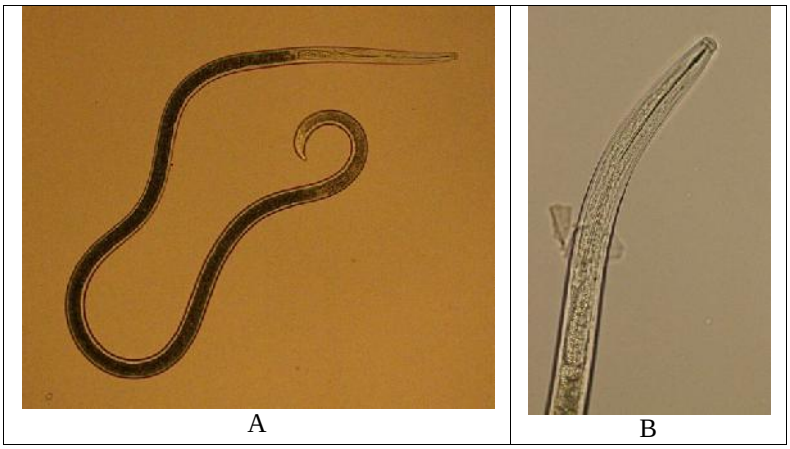


Plate VIII. Microphotograph of *Xiphinema americanum* [A = Adult female and B = Anterior region showing stylet and esophagus]

Pratylenchus zeae (**Lesion nematode**)

The nematode was identified consulting the description of Fortuner and Tom (1976). Female body is vermiform, almost straight on death by heat, marked by very faint annules and 450-475 µm long. Lateral field with 4 incisures. Lip region is not set off from the body. Tip of the head is bluntly round. Stylet is 16-17 µm long, basal knob anteriorly flattened. DGO locate 3

µm behind stylet base. Basal bulb of esophagus is overlaps intestine ventrally. Ovary single and prodelphic. Post-uterine sac is short, 2 body width long. Vulva is at 70-76%. Tail tapering, terminus is almost pointed and narrowly sub-acute (Plate IX).

Zygotylenchus guevarai

The genus was identified comparing the characters described by Siddiqi (1974). Female is vermiform, slightly tapering towards both extremities, slightly arcuate ventrally when relaxed due to thermal death. Cuticle with distinct annulations, lateral field with 4 incisures, without arealation. Esophagus is tylenchoid, basal bulb elongate, lobed, and extending over intestine ventrally and laterally. Style is strong with round basal knobs. Cephalic frame-work is sclerotized, round, low, anteriorly flattened, conoid, labial disc squire. Tail elongate, cylindrical, terminus broadly rounded. Ovary two, vulva sub median, characterized by a transverse slit with slightly raised lips (Plate X).

Tylenchus sp.

Tylenchus was identified using description of Andrassy (1977). It is a small nematode body length less than 1.0 mm. Tail is elongate conoid to filiform. Stylet with distinct knobs. Median bulb is prominent with vulvular apparatus. Basal bulb is abutting with cardia over the intestine. Ovary is monodelphic prodelphic. Vulva lip is raised and well posterior to middle of body. Tail filiform with pointed terminus (Plate IX).

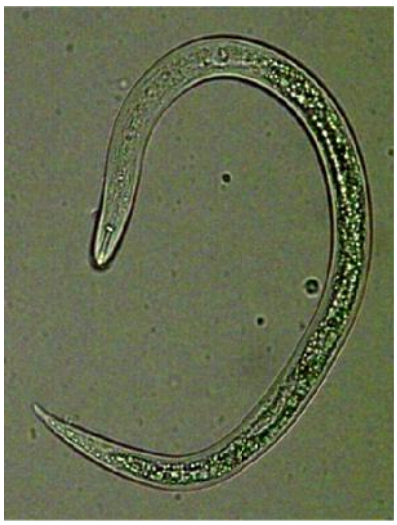


Plate IX. Microphotographs of adult female of *Pratylenchus pratensis*

Plate X. Microphotographs of adult female of *Pratylenchus zeae*

Plate XI. Microphotgraph of an adult *Zygotylenchus guevarai*

Results of the present survey reveal that at least 12 species of plant parasitic nematodes are associated with soils and roots of brinjal may attack brinjal in Bangladesh. Most of them have also been reported from India (Anwar *et al*. 1986, Patel *et al*. 2007, Vermam *et al*. 2013), Pakistan (Bhatnagar *et al*. 1969, Anwar and Choudhry 1973, Maqbool 1986, Shakeel *et al*. 2012) and other countries (Romero and Arias 1969,

Audamou *et al*. 2013, Bhatti *et al*. 2013) as pests of brinjal. Available reports from Bangladesh reveal that all of the genera of nematodes were found to be associated with soils and roots have been reported on other crops of the country (Timm and Ameen 1960, Mian 1986, Mian and Zahid 1986, Mian 1987, Mian and Tsuno 1988). Except root-knot nematode, existence of other 11 plant parasitic nematodes of brinjal has not yet been reported earlier from Bangladesh. So, these may be reported from the country for the first time.

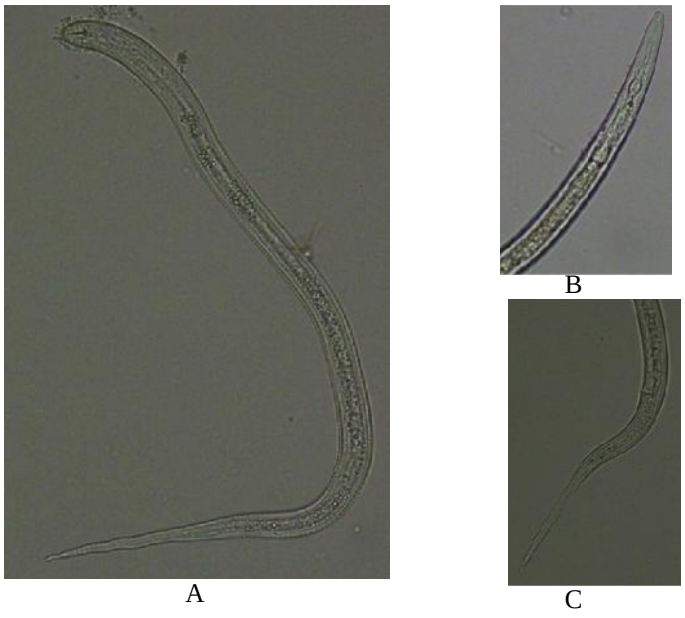


Plate XII. Microphotographs showing adult female (A), anterior region (B) and posterior region (C) of *Tylenchus* sp.

LITERATURE CITED

Andrassy, I. 1977. *Tylenchus* sp. CIH Descriptions of Plant Parasitic Nematodes set 7 No. 97.

Anonymous. 2004a. Hand book of agricultural technology. Bangladesh Agricultural Research Institute (BARI), Joydebpur, Gazipur-1701. pp. 330-33.

Anwar, S. A. and Chaudhry, G. Q. 1973. Nematodes associated with egg plant (*Solanum melongena*) roots. J. Agriil. Res. Punjab. 11(4):93. In Helminthological Abstract – Series B. 1977. Vol. 66 No. 3.

Anwar, S. A. Kalu, M. A., Javid, M. A. and Khan, S. H. 1986. Nematode parasites of brinjal. A. Agric. Res. 24:124-126.

Anwar, S. A. and McKenry, M. V. 2012. Incidence and population density of plant-parasitic nematodes infecting vegetable crops and associated yield losses in Punjab, Pakistan. Pakistan J. Zool. 44(2): 327-333.

Audamou, H., Audamou, B., Oumarou, A. H., Seyni, S. R., D., and Toudou. 2013. Characterization of plant parasitic nematode communities associated with tomato, eggplant and pepper in the suburban area of Niamey (Niger). Intl. Agri. Crop Sci. 5(20): 2488-2494.

Barker, K. R., Carter, C. C. and Sasser, J. N. 1985. An advanced treatise on *Meloidogyne*. Volume II. North Carolina State University Graphics, Raleigh, North Carolina: Methodology. A cooperative publication of the Department of Plant Pathology and the United States Agency for International Development. 1985. pp. 69–77.

Bhatti, K. H., Kausar, N., Rashid, U., Hussain, K., Nawaz, K. and Siddiqi, E. H. 2013. Effects of biotic stresses on eggplant (*Solanum melongena* L.). World Appl. Sci. J. 26 (3): 302-311.

Bhatnagar, R.D.S. and Kadyan, A.S. 1969. A preliminary survey of the plant-parasitic nematodes of brinjal from the Punjab. J. Res. - Punjab Agri. Univ. (1):281-289.

Eisenback, J. D. 2010. A New Technique for Photographing Perineal Patterns of Root-knot Nematodes. J. Nematol. 42(1): 33–34. PMCID: PMC3380516.

Fortuner, R.1976. *Pratylenchus zeae*. CIH Descriptions of Plant Parasitic Nematodes. Set 6 No.77.

Haidar, A., Al-Assas, K. and Al-Ashkar, K. 2008. Survey of phytoparasitic nematode and vesicular arbuscular mycorrhizal fungi genera associating eggplant in Reef Damascus governorate, Syria. Arab Journal of Plant Protection. 26(2):123-128.

Hestling, J. J. (1974). *Ditylenchus myceliophagus*. CIH Descriptions of Plant-parasitic Nematodes. Set 3 No 36.

Hooper, D. J. 1974a. *Aphalenchus avenae*. CIH Descriptions of Plant Parasitic Nematodes. Set 4 No.50.

Hooper, D. J. 1974b. *Cephalenchus emarginatus*. CIH Descriptions of Plant Parasitic Nematodes. Set 3 No.35.

Khan, E. and Chawla, M. I. 1975. *Hoplplaimus indicus*. Plant Parasitic Nematodes. Set 35 No.68.

Loof, P. A. A. 1974. *Tylenchorynchus claytoni*. Plant Parasitic Nematodes.set3 No.39.

Loof, P. A. A. 1974a. *Pratylenchus pratensis*. CIH Descriptions of Plant Parasitic Nematodes. Set 4 No.52.

Loof, P. A. A. 1974b. *Tylenchorynchus claytoni*. Plant Parasitic Nematodes. Set 3 No.39

Maqbool, M.A. 1986. Classification and distribution of plant nematodes in Pakistan. Natl. Nematol. Res. Cen., Uni. Pak., Karachi, Pakistan.

Mian, I. H. 1986. Plant parasitic nematodes associated with some crop species of Bangladesh. Bangladesh J. Plant Pathol. 2(1&2):7-13.

Mian, I. H. 1987. Survey of potato nematodes in Bangladesh. Bangladesh Hort. 15(1):17-22.

Mian, I. H. and Zahid, M. I. 1986. Plant parasitic nematodes associated with betelleaf in Bangladesh. Bangladesh Hort. 14 (2): 49-51.

Mian, I. H. and Tsuno, K. 1988. Identification of major species of *Meloidogyne* and location of their females in root galls using scanning electron microscope. Bull. Inst. Trop.Agr., Kyushu Univ. 11: 15-22.

Mian, I. H. 1994. Introduction to Nematology. Publication No. 23, IPSA-JICA Project. BSMRAU, Gazipur-1706. pp. 64-86.

Patel, A. D., Panickar, B. K., Patel, B. A., and Patel, D. J. 2007. Community analysis of plant parasitic nematodes associated with agricultural crops in Junagadh district of Gujarat and Diu-union territory. Indian J. Nematol. 37(1): 68-71.

Rahman, M. M. and I. H. Mian. 2010. Isolation, and identification of ten genera of plant parasitic nematodes. Bangladesh J. Sci. Ind. Res. 45(3): 267-270.

Romero, M. D. and Arias, M. 1969. Nematodes of solanaceous plants cultivated in the southern Spanish Mediterranean area. I. Tylenchida. [Boletin de la Real Sociedad Espanola de Hostoria Natural (1969) Vol. 67 pp. 121-142]. *In:* Helminthological Abstracts-Series B, 1977, Vol. 46 No. 1. Abstract No. 27.

Sam, L. P. 1979. An assessment of the importance of and control of plant parasitic nematodes of vegetables in Bangladesh. Imperial College, Field statin, Silwood Park, Berkshire, England, 36 pp.

Shakeel, Q., Javed, N., Iftikhar, Y., Haq, I. U., Khan, S. A. and Ullah, Z. 2012. Association of plant parasitic nematodes with four vegetable crops. Pak. J. Phytopathol. 24(2):143-148.

Siddiqi, M. R. 1972. *Helicotylenchus indicus*. CIH Descriptions of Plant Parasitic Nematodes. Set 5 No.68

Siddiqi, M. R. 1973a. *Xiphinema americanum*. CIH Descriptions of Plant Parasitic Nematodes. Set 2: No.29

Siddiqi, M. R. 1975. *Zygotylenchus guebarai*. CIH Descriptions of Plant Parasitic Nematodes. Set 5 No.65

Taylor, A. L. and Sasser, J. N. 1980. Biology, identification and control of root-kot nematode (*Melooidogyne* species). International *Meloidogyne* project. A cooperatibe publication of the Department of Plant Pathology, North Carolina State University and United State Agency for International Development. North Carolina State University, Raleigh, NC 27607, USA. 111 pp.

Timm, R. W. and Ameen, M. 1960. Nematode associated with commercial crops of East Pakinstan. Agric. Pak. 11:355-363.

Verma, D., Kumar, M. and Nazeri, M. 2013. Collection and population frequency of phyto-parasitic nematodes associated with brinjal (solanum melongena) in North Uttar Pradesh region: A field study. J. Cur.Res. Sci. 1(3):169-172. Available at www.jcrs010.com

Zakir, H. and Bora, B. C. 2009. Interrelationship of *Meloidogyne incognita* and *Ralstonia solana-cearum* complex in brinjal. Indian J. Nematol. 39(1): 41-45.

RESPONSE OF PUMPKIN BREEDING LINES TO *PAPAYA RINGSPOT VIRUS*-W

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ABSTRACT

Fatema Begum, M. A. T. Masud, A. M. Akanda, and I. H. Mian. 2014. Response of pumpkin breeding lines to *Papaya Ringspot Virus*-W. Bangladesh J. Plant Pathol. 30(1&2):37-42.

A field experiment was conducted under field condition to screen 29 breeding lines of pumpkin against *Papaya ring spot virus*-W (PRSV-W) under artificially inoculated condition. Five lines (Pk67-1-9-10, Pk13-1-1-9, Pk01-10-9-4-7, BARI mistikumra 1 and Pk102-5) showed characteristic mosaic symptoms of PRSV-W infection. Seventeen lines (Pk05-4-1-1, Pk67-1-9-10, Pk67-1-9-6, Pk37-1-4-6, Pk13-1-1-9, Pk20-2-1-9, Pk02-2-1-6, Pk55-2-2-10, Pk61-1-1-5, Pk54-4-12-9, Pk54-4-12-1, Pk05-1-2-4, Pk05-1-2-10, Pk01-10-9-4-7, BARI mistikumra 1, Pk102-5 and Pk105-2) manifested positive reaction in ELISA to PRSV-W infection. Of these, 7 lines showed resistant reaction having disease incidence of 13.3 to 24.4% and 12 lines exhibited negative reaction to PRSV-W which was graded as highly resistant. Maximum disease incidence (58.3%) was recorded on Pk67-1-9-10 while minimum on Pk13-1-1-9 (9.0%). Significantly the highest disease severity and AUDPC were found on line BARI mistikumra 1. The disease severity ranged 13.3-15.0% and AUDPC ranged 46.7-52.5 in three lines (Pk13-1-1-9, Pk20-2-1-9 and Pk02-2-1-6). Pumpkin lines which showing highly resistance and resistance reactions to PRSV-W may be used in breeding program for pumpkin improvement program.

Key word: PRSV-W, pumpkin line, response, plant growth, yield

INTRODUCTION

At least 35 viruses infect cucurbits including pumpkin (Lovisolo 1980). Among the viruses *Papaya Ringspot Virus* - Water melon strain (PRSV-W) is the most destructive one causing significant reduction in plant growth and yield (Rezende and Pacheco 1998). The virus was first described in papaya (PRSV-P) by Jensen (1949) and in cucurbits (PRSV-W) by Webb (1965) and Webb and Scott (1965). Akanda (1991) reported the virus from Bangladesh and he found that it may cause 70-100% yield reduction of cucurbits depending upon the stage of infection. PRSV-W belongs to the family Potyviridae and is transmitted by aphids in a non-persistent manner (Purcifull *et al.* 1984).

Major virus control strategies include the use of insecticides to eliminate virus vectors, herbicides to remove alternative hosts for the virus and genetic resistance (Provvidenti 1993), which is often pathogen-specific (Grumet 1989). Of those control strategies, the most economical method is genetic resistance. Virus resistance may also be accomplished through virus coat proteins transferred into existing cultivars (Namba *et al.* 1992, Quemada *et al.* 1990), or by screening of germplasm.

The present investigation was undertaken to determine the response of pumpkin lines to PRSV-W and to determine the effect of PRSV-W on plant growth and yield.

MATERIALS AND METHODS

A field experiment was conducted to screen 29 pumpkin lines against PRSV-W under artificially inoculated condition. The experiment was conducted in the experimental field of Bangabandhu Sheikh Mujibur Rahman Agricultural University (BSMRAU), Gazipur during October 2011 to April 2012 to screen. PRSV-W infected pumpkin plants of previous year experiment were selected on the basis of visible symptoms and Enzyme-Linked Immuno Sorbent-Assay (ELIZA). Leaf samples were collected from the selected PRSV-W infected plants. Inoculum of PRSV-W was prepared by grinding the infected leaves using mortar and pestle in 0.02 M phosphate buffer, pH 7.0. Leaf sample to buffer ratio was 1:10 (1 g infected leaf and 10 ml buffer). The sap of infected pumpkin leaves obtained after passing through double-ply cheese cloth was used as inoculum. Mechanical inoculation method using carborundum powder (800 meshes, Fisher Scientific, Fair Lawn, NJ) was followed (Daryono 2006).

Seedlings of the selected lines were raised in polyethylene bags. Before development of the true leaf, both cotyledons of seedlings were rubbed with carborundum powder to make minor injuries.

The inoculum sap was soaked with cotton and gently rubbed on the injured areas of leaf. After inoculation, carborundum powder was washed off with sterilized distilled water. Inoculated pumpkin seedlings were kept in aphid-proof cages for 10 days and transplanted in the main field. Standard procedures were followed for cultivation of land, preparation of bed and pits, application of manures and fertilizers, transplanting of seedlings, intercultural operations (Razzaque *et al*. 2000).

Pumpkin plants grown in the experimental field were checked at 55 days after transplanting to record the incidence of PRSV-W. Disease incidence was identified based on visible symptoms followed by serological test using PRSV-W antiserum (Wei *et al.* 2001). The lines which showed positive reaction to PRSV-W antiserum were graded as susceptible and those showed negative reaction to the antiserum were graded as resistant according to Daryono (2006).

Data on disease incidence, disease severity and area under disease progress curve (AUDPC) of virus disease in experimental field were recorded through frequent visit after appearance of symptoms. Disease incidence was estimated using a standard formula ofAgrios (2005):

$$Disease\ incidence\ (\%) = \frac{Number\ of\ diseased\ plant\ (or\ parts)}{Total\ number\ of\ plants\ (or\ parts)\ checked} \times 100$$

According to Begum and Khan (1996), the lines were graded based on degree of disease incidence as highly resistant (HR=0.0% disease incidence), resistant (R=0-25% disease incidence), moderately resistant (MR= 26-50% disease incidence), moderately susceptible (MS= 51-75% disease incidence) and susceptible (S= 76-100% disease incidence). Disease severity was expressed in percent disease index (PDI). The PDI was computed using a standard formula (Piper *et al.* 1996) as shown below:

$$PDI = \frac{\sum Disease\ grade\ x\ number\ of\ plants\ in\ grade}{Total\ number\ of\ plants\ x\ highest\ disease\ grade} \times 100$$

The severity of virus disease of pumpkin was indexed on a 0-5 indexing scale, where 0 = no visible symptoms, 1 = slightly mosaic on leaves, 2 = mosaic patches and/or necrotic spots on leaves, 3= leaves near apical meristem deformed slightly, yellow, and reduced in size; 4= apical meristem with mosaic and deformation, and 5= extensive mosaic and serious deformation of leaves (Xu *et al*. 2004).

Area under the disease progression curve (AUDPC) wad calculated according to Tooley and Grau (1984) using the formula shown below:
AUDPC=$\sum$ $([x_i+x_{i+1})/2]$ $(t_{i+1}-t_i)$, where, Xi= cumulative disease incidence, expressed as a proportion of observation, ti= time (days after planting) the ith observation and n= total no. of observation.

The experiment was laid out following randomized complete block design with three replications. Each replication was consisted of four plants. Data con growth parameters (branches/plant and vine length), yield (relative to fruits/plant, yield/plant) and total soluble solids (%TSS) were recorded. Correlation of plant growth parameters and yield with PRSV-W incidence determined using MSTAT-C software and mean separation was done by DMRT.

RESULTS AND DISCUSSION

Development of symptoms of PRSV-W in inoculated plants

Upon inoculation, pumpkin lines tested in the present experiment exhibited various degrees of symptoms of PRSV-W infection such as mosaic, vein clearing, leaf curling, blistering, deformation of the leaf leading to the formation of fern leaf or shoe string. Mild mosaic with vein clearing and vein banding were also observed on infected leaves as associated symptoms. At later stage of disease development, entire leaves became deformed and small. The infected plants produced deformed, smaller fruits having green raised spots scattered on the surface. The older leaves were small and deformed fern leaf like appearance (Plate 1).

Reaction of pumpkin lines to PRSV-W

On inoculation of pumpkin leaves with PRSV-W, lines Pk67-1-9-10, Pk13-1-1-9, Pk01-10-9-4-7, BARI mistikumra 1 and Pk102-5 showed mosaic symptoms at cotyledon stage. Symptoms developed after cotyledon stage, were severe mosaic on the young leaves, crinkling and blisters on the older leaves (Plate I). ELISA test was performed using one antiserum (PRSV-W) to detect presence of PRSV-W in inoculated leaves showing no visible symptoms of virus infection.

Out of 29 pumpkin lines tested, 17 (Pk05-4-1-1, Pk67-1-9-10, Pk67-1-9-6, Pk37-1-4-6, Pk13-1-1-9, Pk20-2-1-9, Pk02-2-1-6, Pk55-2-2-10, Pk61-1-1-5, Pk54-4-12-9, Pk54-4-12-1, Pk05-1-2-4, Pk05-1-2-10, Pk01-10-9-4-7, BARI mistikumra 1, Pk102-5 and Pk105-2) showed positive reaction against PRSV-W antiserum. These were graded as susceptible. Remaining 12 lines did not show any visible symptoms of PRSV-W infection or positive reaction against PRSV-W antiserum and graded as resistant to the virus.

Disease incidence, severity and AUDPC

Significantly the highest incidence PRSV-W (54.3-58.3%) was recorded from pumpkin lines Pk67-1-9-10, BARI mistikumra 1, Pk67-1-9-6, which were graded as moderately susceptible (MS). In lines Pk105-2, Pk37-1-4-6, Pk05-1-2-10, Pk102-5, Pk05-4-1-1 and Pk05-1-2-4, the disease incidence was 26.0-46.3% and these were graded s moderately resistant (MR). The incidence of PRSV-W was 9.0-24.3% on lines Pk01-10-9-4-7, Pk02-2-1-6, Pk55-2-2-10, Pk61-1-1-5, Pk01-10-9-4-7, Pk54-4-12-9, Pk20-2-1-9 and Pk13-1-1-9 and these were graded as resistant (R). Rest of the 12 pumpkin lines were free from infection with PRSV-W and graded as highly resistant (HR) (Table 2).

The percent disease index (PDI) value ranged from 13.3 to 58.3%. The highest PDI of was found on BARI mistikumra1 followed by Pk37-1-4-6. The lowest PDI was observed on Pk13-1-1-9 which was statistically similar to that of Pk20-2-1-9 and Pk02-2-1-6 (Table 2).

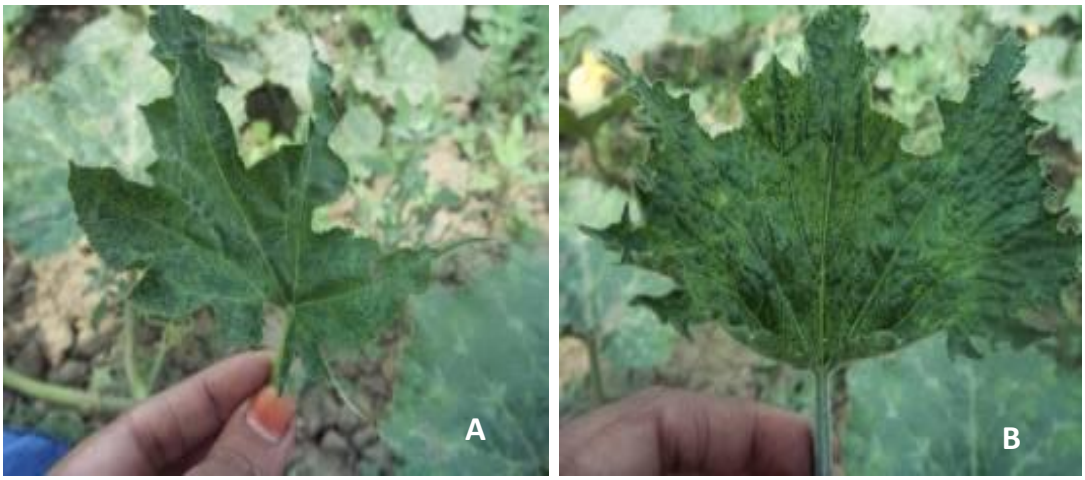

Plate 1. Crinkling (A) and mosaic (B) symptoms in PRSV-W inoculated pumpkin leaves

Table 1. Reaction of pumpkin lines to PRSV-W checked by DAS-ELISA test.

Pumpkin lines	Serological test	Grading
Pk31-2-4-10, Pk34-4-3-8, Pk07-4-7-1, Pk13-1-1-1, Pk20-2-1-3, Pk19-4-1-10, Pk04-7-12-3-4, Pk05-7-11-8-3, BARI mistikumra 2, Pk101-8, Pk106-4, Pk107-4	-	Resistant =R
Pk05-4-1-1, Pk67-1-9-10, Pk67-1-9-6, Pk37-1-4-6, Pk13-1-1-9, Pk20-2-1-9, Pk02-2-1-6, Pk55-2-2-10, Pk61-1-5, Pk54-4-12-9, Pk54-4-12-1, Pk05-1-2-4, Pk05-1-2-10, Pk01-10-9-4-7, BARI mistikumra 1, Pk102-5, Pk105-2	+	Susceptible=S

Mean AUDPC ranged 46.7 - 198.3 at 55 days after inoculation. The highest AUDPC was recorded from BARI mistikumra1 followed by Pk37-1-4-6. The lowest AUDPC was observed on line Pk13-1-1-9, which was statistically similar to that of Pk20-2-1-9 and Pk02-2-1-6 (Table 2).

Relationship of plant growth and yield parameters with disease incidence and severity

The relationship of plant growth and yield parameters such as branches/plant, vine length, fruit number/plant, yield /plant TSS% with incidence of PRSV-W was linear and negative but not significant (Fig. 1 A, B and Fig. 2A, B & C).

The relationship of fruit number/plant, fruit yield/plant and %TSS of pumpkin with PRSV-W was liner and negative. The correlation coefficients of the relationship of fruit/plant (r=-0.019) and yield/plant (r=-0.067) with disease incidence was negative but not significant. The correlation co-efficient (r) was (- 0.019 and -0.067) and the contribution of regression (R^2 =0.140 and R^2 =0.175)

was 14% and 17.55, respectively (Fig. 2A&B). The correlation co-efficient (r) was - 0.008 and the contribution of regression (R^2 = 0.019) was only 19% (Fig. 2C). Virus in infected plant hampered the physiology and nutrient uptake of plants. So, yield and yield contributing characters are also affected. In virus-stressed plants, there was decreased yield in infected plant compared to healthy plant.

The symptoms recorded from pumpkin lines inoculated with PRSV-W are identical with the symptoms developed on other cucurbits due to viruses infection as reported by other investigators (Purcifull *et al.* 1984). Results of the present investigation reveal that disease incidence, severity, AUDPC and response of the tested pumpkin lines to PRSV-W are varied with the vitiations of the pumpkin lines. The variations may due to genetically variations in the pumpkin lines tested. Sherwood *et al.* (1986) also reported that differences in the pattern of incidence and degree of severity among cultivars may be due to variation in genetic make-up of the tested cultivars as well as the strain of the virus and possible co-infection with other viruses. Masud (1995) tested 27 pumpkin genotypes under field condition. Of them three were resistant and nine moderately resistant to pumpkin viruses. The resistance observed in those lines could be related to the existence of mechanisms that inhibit movement of virus from inoculated leaves to healthy leaves. Resistance could involve cellular membrane changes that impede the diffusion transport of infective virus particle from cell to cell, or an inhibition of virus particle replication in the leaf tissue of resistance plants (Gray *et al.* 1988).

Finding of the present experiment show that plant growth and yield of pumpkin are affected by the PRSV-W. The findings are consistent with findings of previous researchers (Fortun and Lopez 1982, Tattini *et al.* 1990, Huang 2003). They found that disease incidence was used and positive effects on plant growth could be proved. The positive effect of growth on the nutrients uptake was also proved with tomato, cucumber and other plants.

Table 2. PRSV-W incidence, severity and AUDPC in pumpkin during 2011-2012

Pumpkin lines	Disease incidence (%)	Disease severity (%)	AUDPC[Z]	Reaction
Pk67-1-9-10	58.3 a	38.3 d	134.2 d	MS
BARI mistikumra 1	56.0 a	58.3 a	198.3 a	MS
Pk67-1-9-6	54.3 a	33.3 e	116.7 e	MS
Pk54-4-12-1	46.3 b	38.3 d	134.2 d	MR
Pk105-2	44.3 b	46.7 c	163.4 c	MR
Pk37-1-4-6	40.7 bc	51.1 b	188.6 b	MR
Pk05-1-2-10	35.0 cd	30.0 f	105.0 f	MR
Pk102-5	34.0 d	35.0 e	122.5 e	MR
Pk05-4-1-1	30.3 de	47.8 c	171.1 c	MR
Pk05-1-2-4	26.0 ef	30.0 f	105.0 f	MR
Pk01-10-9-4-7	24.3 ef	26.7 g	93.4 g	R
Pk02-2-1-6	15.3 gh	15.0 h	52.5 h	R
Pk55-2-2-10	12.7 h	33.3 e	116.7 e	R
Pk61-1-1-5	12.3 h	26.7 g	93.35 g	R
Pk54-4-12-9	20.0 fg	40.0 d	140.0 d	R
Pk20-2-1-9	14.7 gh	15.0 h	52.5 h	R
Pk13-1-1-9	9.0 h	13.3 h	46.7 h	R
BARI mistikumra 2, Pk101-8, Pk07-4-7-1, Pk13-1-1-1, Pk20-2-1-3, Pk19-4-1-10, Pk31-2-4-10, Pk34-4-3-8, Pk04-7-12-3-8, Pk05-7-11-8-3, Pk106-4, Pk107-4	0.0 i	0.0 i	0.0 i	HR

[Z]Mean of the disease progress assessed as an area under the disease progression Curve (AUDPC) following the formula $\sum ([x_i+x_{i+1})/2] (t_{i+1}-t_i)$ using 0-5 scale.
MS=Moderately susceptible, MR=Moderately resistant, R=Resistant, HR=Highly resistant.

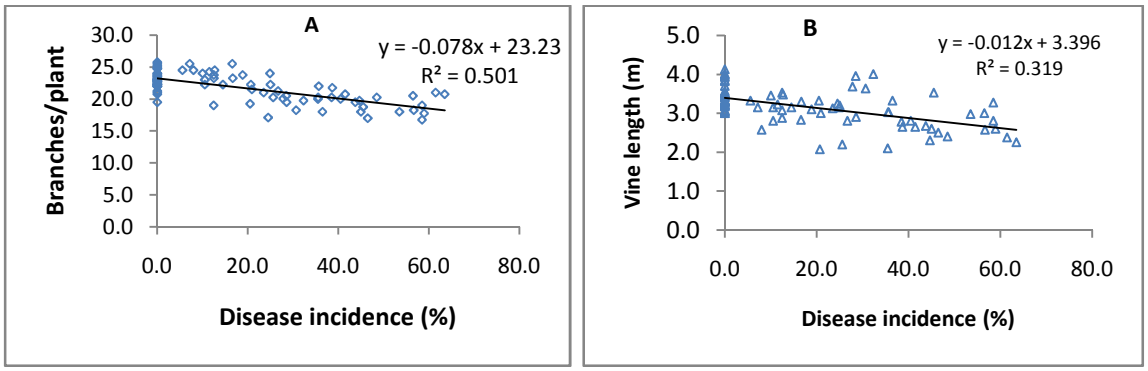

Fig. 1. Relationship between disease incidence with growth characters (A) Branches/plant (B) Vine length

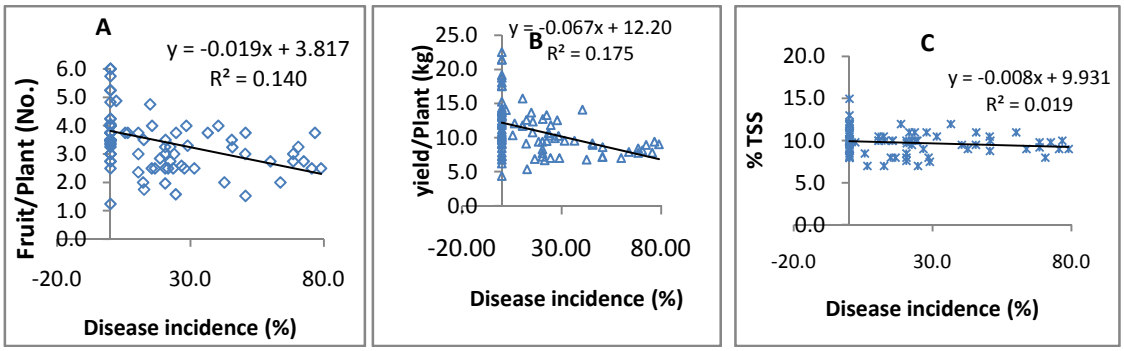

Fig. 2. Relationship between disease incidence with Yield and yield contributing characters (A) Fruits/Plant, (B) Yield/Plant and (C) %TSS.

Finding of the present experiment show that plant growth and yield of pumpkin are affected by the PRSV-W. The findings are consistent with findings of previous researchers (Fortun and Lopez 1982, Tattini *et al.* 1990, Huang 2003). They found that disease incidence was used and positive effects on plant growth could be proved. The positive effect of growth on the nutrients uptake was also proved with tomato, cucumber and other plants.

LITERATURE CITED

Agrios, G. N. 2005. Plant Pathology. 5th Edn., Academic Press, Burlington, 992 pp. ISBN:0120445654.

Akanda, A. M. 1991. Studies on the virus and mycoplasma disease of crops in Bangladesh. A Thesis submitted to the Faculty of Agriculture, Kyushu University, Japan for the partial fulfillment of Doctor of agriculture. 181pp.

Begum, S.N. and Khan, M. A. 1996. Tomato Leaf curl virus in Bangladesh. Proceedings of the Phase-1 final workshop of the South Asia Veg. Res. Network, Katmandu, Nepal, pp. 210-215.

Daryono, B. S. 2006. Resistance to cucurbit viruses in several genotypes of melon (*Cucumis melo* L.). Berkal Ilmiah Biol. 5 (1): 1-12.

Fortun, C. and Lopez, C. 1982. Influence of humic acid on the mineral nutrition and the development of the maize roots, cultivated in normal nutritive solutions and lacking of Fe and Mn. Anades-de dafologia-y-agrobiologia (Spain). 41(1-2): 335-349.

Gray, S. M., Moyer, J. W. and Kennedy, G. G. 1988. Resistance in *Cucumis melo* to watermelon mosaic virus 2 correlated with reduced virus movement within leaves. Phytopathology78:1043-1047.

Grumet, R., 1989. Genetically engineered plant virus resistance. Hort. Sci. 25(5):508–513.

Huang, C.A. 2003. The partial characterization of melon vein-banding mosaic virus, a newly recognized virus infecting cucurbits in Taiwan Plant Pathol. 42(1):100-107.

Jensen, D. D. 1949. Papaya virus diseases with special reference to papaya ring spot. Phytopathology 39:191-211.

Lovisolo, O. 1980. Virus and viroid diseases of cucurbits. *Acta Horticulturae*. 88: 3-90.

Masud, M. A. T. 1995. Variability association and genetic diversity in pumpkin. M.S. Thesis, Dept. of Hort., BAU, Mymensingh.

Namba, S., Ling, K., Gonsalves, C., Slightom, J. L. and Gonsalves, D. 1992. Protection of transgenic plants expressing the coat protein gene of watermelon mosaic virus II or zucchini yellow mosaic virus against six potyviruses. Phytopathology 82: 940-946.

Piper, J. K., Handley, M. K. and Kulakow, P. A. 1996. Incidence and severity of viral disease symptoms on eastern gamagrass within monoculture and polycultures. Elsevier. Agric. Eco. Environ., 59:139-147.

Provvidenti, R. 1993. Resistance to viral diseases of cucurbits, In: M.M. Kyle (ed.). Resistance to viral diseases of vegetables: genetics and breeding. Timber Press, Portland. 843pp.

Purcifull, D. E., Edwardson, J. R., Hebert, E. and Ganosalves, D. 1984. Papaya Ring spot Virus. CMI/AAB Descriptions of plant viruses, No. 292.

Quemada, H., Sieu, L. C., Siemieniak, D. R., Gonsalves, D. and Slightom, J. L., 1990. Watermelon mosaic virus II and zucchini yellow mosaic virus: cloning of 30-terminal region, nucleotide sequences, and phytogenetic comparisons. J. Gen. Virol. 71:1451–1460.

Razzaque, M. A., Sattar, M. A., Amin, M. S., Quayum, M. A. and Alam, M.S. 2000. Krishi Projukti Hatbai (in Bangla). BARI Gazipur, Bangladesh. 464 pp.

Rezende, J. A. M. and Pacheco, D. A. 1998. Control of *Papaya ringspot virus*- type W in zucchini squash by protection in Brazil. *Plant Dis.* 82:171-175.

Shukla, D. D., Ward, C.W., and Brunt, A. A. 1994. The potyviridae. Cab International. Wallingford, Oxon UK.

Tattini, M., Chiarini, A., Tafani, R. and Castagneto, M. 1990. Effect of humic acids on growth and nitrogen uptake of container-grown olive (*Olea europaea* L. 'Maurino'). Internat. Symp. on Olive Growing, Proceeding, Wageningen (Netherlands). pp. 125-128.

Tooley, P. W. and Grau, C. R. 1984. Field characterization of rate reducing resistance to *Phytophthora megasperma* f.sp. *glycinea* soybean. Phytopathology 74:1901-1908.

Webb, R. E. 1965. *Watermelon mosaic virus* in cucumber, melon and squash. Plant Disease. 75: 203-207.

Webb, R. E and Scott, H. A. 1965. Isolation and identification of watermelon mosaic viruses 1 and 2. Phytopathol. 55: 895-900.

Wei, S., Jiao, K. and Zhang, S. 2001. Electrochemical ELISA for the detection of *Cucumber mosaic virus* using opheneyl-enediamine as substrate Talanta. 55(6):1211-1218.

Xu Y., Kang, D. Shi, Z. Shen, H. and Wehner, T. 2004. Inheritance of Resistance to Zucchini Yellow Mosaic Virus and Watermelon Mosaic Virus in Watermelon. J. Heredit. 95(6):498–502.