BOTRYTIS BLIGHT OF GLADIOLUS IN MYMENSINGH AND ITS MANAGEMENT

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ABSTRACT

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Botrytis blight symptoms appeared on Gladiolus grown in Mymensingh regions of Bangladesh during 2014-2015. The disease caused spots on leaves, stems/ spikes, buds and flowers. In severe infection, the disease caused both flower and leaf blight. In cool and moist weather Botrytis blight incidence was recorded up to 100% in some fields . The causal pathogen identified as *Botrytis gladiolorum* The effect of temperature on mycelial growth, sporulation and sclerotial production of *B. gladiolorum* was investigated in different temperatures. The maximum radial was found 20 $\pm1^0$C . An excellent degree of conidial and sclerotial production also took place at 20 and 25 ± 1^0C. The optimum spore concentration for disease development on the leaf tissue was at $4x10^4$ conidia/ml of water that was identical as recorded from the field. *Trichoderma harzianum* (2%) significantly reduced the growth of *B. gladiolorum.* Maximum plant height, total number of leaves, number of spikes, rachis length, and number of florets, floret diameter and yield (flower stalk /ha) were obtained with the application of 2.0% *Trichoderma harzianum* followed by Bavistin (0.2%) in the field experiment.

Key words : Botrytis blight, gladiolus and management.

INTRODUCTION

Gladiolus (*Gladiolus communis*) is very popular flower and grown throughout the world in a wide range of climatic conditions. Its magnificent inflorescence with various colour have made it attractive in Bangladesh also. Income from gladiolus flower production is six times higher than from that of rice (Momin 2006). Gladiolus was introduced in Bangladesh around 1992 from India (Mollah *et al*. 2002). It has recently been become popular in Bangladesh. Its demand has been increasing day by day with the advancement of aristocracy and modernization of Bangladesh. But the flower suffers from many diseases such as corm rot, leaf spot and leaf blight. Now a days, leaf blight which is caused by *Botrytis gladiolorum* become severe in the farmer's field of Mymensingh region that thrives in high humidity and cool weather. No attention has been given on the diagnosis of botrytis blight and its control in Mymensingh region earlier. On the other hand, chemical fungicide is harmful for human being which is also hazardous to our environment. Therefore, this research work was undertaken to diagnose/identify the leaf blight of gladiolus and management of this disease under field condition.

MATERIALS AND METHODS

The experiments were conducted in the farmers field of Bhabokhali and Sutiakhali of Mymensingh district; Horticulture field, Plant Disease Clinic of Bangladesh Agricultural University, Mymensingh during December 2014 to March 2016. The variety "Mount Everest" was used in this experiment. Percent disease incidence and severity were calculated by using the formula of Mansoor (2007) surveying of the diseased field. Weather data was collected from weather office (Weather report, 2015), BAU during this period. Botrytis infected leaf and flowers were collected from the infected field, kept in polythene bag and brought to the Plant disease clinic for diagnosis. Several temporary slides were prepared from the infected samples by picking method, and Botrytis spore identified following Robert (2008). The collected disease sample was washed into tap water to make them free from soil and sand. To get pure culture of this pathogen, Tissue planting method was followed and inoculated plates on PDA were incubated at 20 ± 1^0C with blower for 12 days. Botrytis blight associated fungus was identified based on morphology as described by Mirzaei *et al*. (2008) and Sung *et al.* (2003). Botrytis growth, it's sporulation and sclerotial production were checked in different temperature viz.10, 15, 20, 25 and 30 ± 1^0C in

incubator for 20 days as described by Sehajpal and Singh (2014). The culture of the fungus was raised on PDA medium at 20±1^{0}C. The concentration of spores was standardized at 1x10^4, 2x10^4, 3x10^4 and 4x10^4 conidia/ml of water for pathogenicity test by artificial inoculation on healthy leaves of gladiolus seedling grown in pot in CRD design. The observations on % severity of the disease were recorded after inoculation of 4, 8 and 12 days, respectively (Sung *et al*. 2003). A Poison Food Technique was followed (Singh and Milne 1973) against *Botrytis* pathogen by using Tilt (0.1%, 0.2%), Bavistin (0.1%, 0.2%) and *Trichoderma harzianum* (1%, 2%) at 20±1 °C. Colony diameters were recorded in every 3 days after inoculation. Land was prepared as described by Nabi (2010); cow dung and fertilizers were applied as recommended by BARC (BARC, 2012). A total of three treatments were used viz.T_0 = Control, T_2 = Bavistin @ 0.2% and T_3= *Trichoderma harzianum* @ 2%. The experimental plot was prepared where corms planted at a depth of 5 cm adopting with ridges and furrows system (Mirzaei,. Ten corms were planted in each row spaced 30 cm apart, corm spacing within rows was 15 cm. One row was considered as one replication. Each treatment was replicated 3 times with Randomized Complete Block Design (RCBD). Data on plant height (cm), % plant infection, total no. of leaves/plant, total no. of healthy leaves/plant, total no. of infected leaves/plant, rachis length (cm), no. of floret /spike, floret diameter and yield (flower stalk/ha) were recorded. The collected data were tabulated and analyzed through a standard computer package statistical procedure by Wasp-2.

RESULTS AND DISCUSSION

Botrytis blight was recorded from all the surveyed 5 (five) fields of Sutiakhali and Babukhali of Mymensingh region and significantly the highest incidence (100%) was recorded at 75 days after sowing on 21 January, 2015 and lower incidence (6%) was found at the younger plants of 45 days on December, 2014 The similar trend was observed in case of disease severity that ranged from 8-60% (Table 1). Significantly the highest incidence (100%) and severity (60%) of Botrytis blight (Plate: 1) was observed at older plants than the younger ones after the 3rd week of January 2015. Sung *et al*. (2003) also found that the Botrytis gray mold *(B. gladiolorum*) reached up to 50% in damaged fields in Korea and *B. gladiolorum* spores produced gray mold on older plants drifted onto the flowers before harvest. However, severe outbreaks of Botrytis blight in mature stage were induced may be due to low temperature 17.9 ^{0}C, high humidity (89%), Rainfal (15 mm) with wind speed (3.06 kmph) and no sunshine at that time. This is supported by Sehajpal *et al*. (2015) who revealed that the progression of botrytis blight disease was more in cool weather and towards the winds and wind direction during January-February. This is the first report of Botrytis blight of gladiolus caused by *B. gladiolorum* in Mymensingh region in Bangladesh. The severely infected leaves become reddish-brown with grayish conidial masses and dried from the tips. As the disease progressed, the lesions developed and blighted completely the spike, petal, flower bud with grey rot of flowers (Fig. 1). This result is supported with the findings of Sung *et al*. (2013) and Siddique *et al*. (2013). Conidia were ellipsoidal or obovoid, unicellular, pale brown, smooth and measured 9.0-18.8 x 7.4-15.0 µ in diameter (Fig. 2). The morphological characteristics of mycelia, conidiophores, conidia and sclerotia of *B. gladiolorum* recorded in this present investigation (Fig. 2) are almost similar to the descriptions of Wang *et al*. (1996), Kishi (1998), Sung *et al*. (2003) and Mirzaei *et al*. (2008). The highest aerial mycelial growth (90 mm in dia) was recorded at 20±1^{0}C, the conidial and sclerotial formation also occurred at temperatures of 15, 20 and 25±1^{0}C, respectively. No conidial and sclerotial production was recorded at 10 and 30±1^{0}C (Table 2). This results are in agreement with the findings of Ahmed *et al*. 2007, Hosen 2010, Sehajpal and Singh 2014 who found that temperature of 20±1^{0}C was optimum for growth of *B. cinerea* in some other hosts and PDA medium supported good colony growth and excellent sporulation of *B. gladiolorum.* Singh and Arora (1994); Shakir *et al*. (1998) also found that *B. gladiolorum* grew well at 15^0 C to 25^{0}C : while its growth was decreased with the increase of temperature. The disease severity on the foliar tissue was observed at 5%, 15% and 40 % when 1 x10^4 (CFU/ml) conidia was sprayed and maximum disease severity (100 %) was recorded in the highest spore concentration, i.e. 4x 10^4 CFU/ml of water after 12 days of inoculation (Table 3). About 52% disease severity was observed at an inoculum load of 4x10^4 conidia/ml after 4 days of inoculation and100% at 12 days of inoculation. The infection has generally been reported to high at higher spore concentrations by many workers (Last and Hamley 1956, Stewart and Mansfield 1984).

Pathogenicity tests revealed that conidial suspension of *B. gladiolorum* @ 4x10^4 conidia/ml caused blight symptoms on leaves and flowers. The disease incidence (DI) was first appeared at 15-20 days after spraying. Conidia were isolated from the infected leaves and flowers. This is in agreement with the

findings of many researchers who reported that *B. gladiolorum* infected gladiolus in North America, Europe, Africa, New Zealand, China, and Japan (Kishi 1998, Mckenzie 1990, Wang *et al.* 1996, Sung *et al*. 2003, Mirzaei *et al*. 2008). Siddique *et al*. (2013) also reported that *Botrytis* blight caused by *B. gladiolorum* regularly attacked the gladiolus plants in Jessore regions of Bangladesh. However, this results regarding isolations, pathogenicity are in confirmity with those of Mirza and Shakir (1991) and Sohi (1992). In vitro bioassay of *Botrytis gladiolorum* against chemicals and bioagent showed that the highest growth was inhibited by Bavistin and *T. harzianum* than nontreated treatment (Table 4). These results are in conformity with those of Shakir *et al*. (1998), Singh and Arora (1994) and Singh *et al*. (2005) who observed that Bavistin proved its performance against *Botrytis* and *Fusarium oxysporum.* Tesfaye and Kapoor (2010) reported that *T. harzianum* could effectively control *Botrytis gladiolorum.* Hermosa *et al*. (2000) also reported that *Trichoderma harzianum* reduces mycelial growth of plant pathogens. Tesfaye and Kapoor (2004) indicated that *In vitro* treatment of *Trichoderma harzianum, T. viride,* and *Gliocladium* species reduce mycelial growth of Botrytis corm rot (*Botrytis gladiolorum*).

Field experiment revealed that Bavistin @ 0.2% and Tricho-suspension @ 2.0% significantly reduced the blight disease (14.2 and 12.5%) where control yielded 42.8% disease incidence. The height of plants, number of leaves/plant, rachis length, no. of floret/spike, floret length and diameter of florets significantly increased with the application of *Trichoderma harzianum* @ 2% followed by Bavistin. (Table 5). *Trichoderma harzianum* was found superior in terms of yield/ha (2.42 lac flower stalk) followed by Bavistin (1.90 lac flower stalk/ha) where control yielded 1.78 lac flower stalk/ha. Tesfaye and Kapoor (2007, 2010) have shown that i*n vivo* evaluation of *Trichoderma* species against *Botrytis corm* rot (*Botrytis gladiolorum*) drastically reduced the disease incidence and severity and simultaneously obtained maximum yield of Gladiolus. Spraying Bavistin or chemical was impractical because concentrated or frequent sprays injured and stained the petals (Mirzaei *et al*. 2008) but *Trichoderma* was not only effective in controlling the *B. gladiolorum* infection, but also increased the yield of flowers as well. Jegathambigati *et al.* (2009) also reported that the *Trichoderma* treatment enhenced plant growth, leading to a significant increase in plant height and weight in relation to untreated control.

Table 1. Occurrence of Botrytis blight of Gladiolus in Mymensingh area during 2014 -2015

Plant age	% Botrytis blight incidence	% Botrytis blight severity
45 days	6.0	8.0
60 days	15.0	12.0
75 days	100.0	60.0
Lsd (0.05)	2.018	2.021

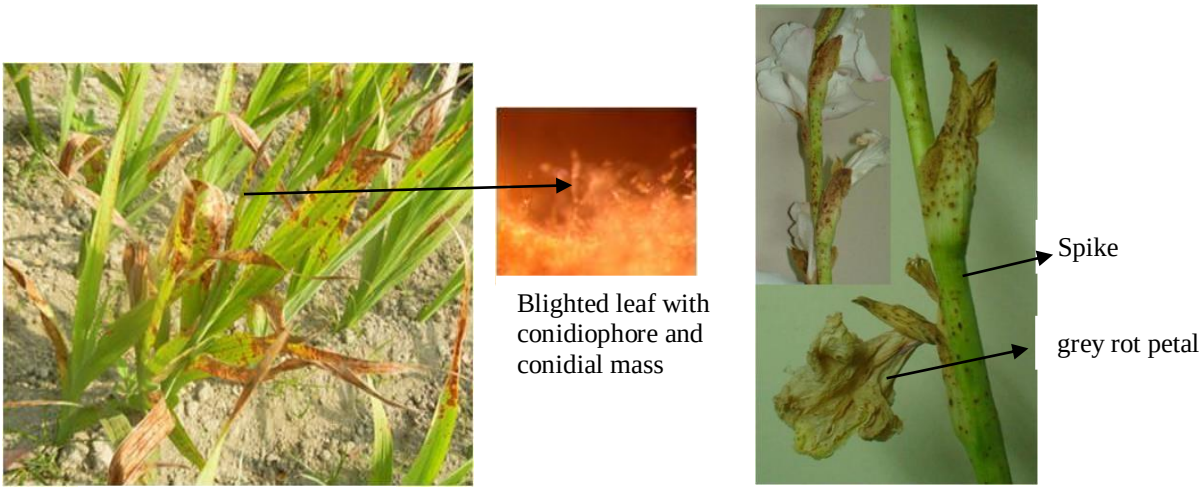

Fig. 1: Botrytis Blight symptoms on infected leaf, spike and flower

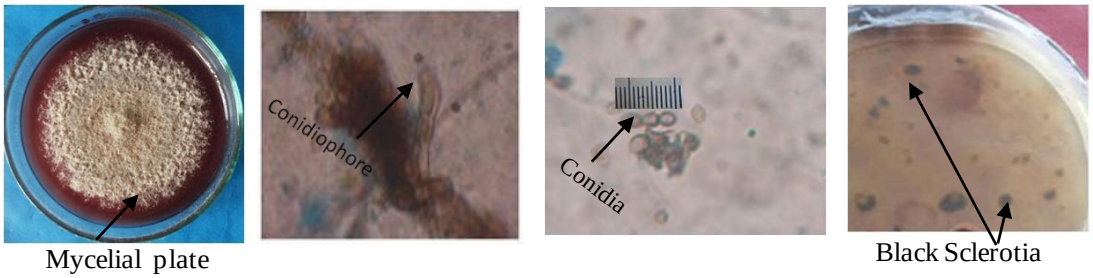

Fig. 2: Morphological characters of *B. gladiolorum*

Table 2. *In vitro* assay of *Botrytis gladiolorum* at different Temperatures

Temperature ($\pm 1^0$C)	Culture medium	Colony growth (mm)			Conidial production	Sclerotial production
		4 days	8 days	12 days		
10	PDA	18	40	55	-	-
15	PDA	26	58	80	+	+
20	PDA	36	74	90	+++	++
25	PDA	21	55	72	++	+
30	PDA	18	52	68	-	-
LSD (0.05%)		1.897	2.776	8.692		

Indices : – = No, + = Poor, ++ = Good; and +++ =Excellent.

Table 3. Effect of inoculum levels of *Botrytis gladiolorum* on botrytis blight severity of gladiolus in 2015

Sl. No.	Spore concentration (Conidia/ml)	Botrytis blight Severity (%)		
		4 days	8 days	**12** days
1.	$1x10^4$	5.0	15.0	40.0
2.	$2x10^4$	12.2	38.0	66.0
3.	$3x10^4$	35.0	55.0	85.0
4.	$4x10^4$	52.0	80.00	100.00
LSD (0.05)		4.501	11.1	8.076

Table 4. *Invitro* assay of *B. gladiolorum* against chemicals and bioagent

Treatments	**Growth of *B. gladiolorum* in diameter (mm)**	
	7 days after inoculation	12 days after inoculation
Control	62.0	90.0
Tilt (0.1%)	16.0	19.5
Tilt (0.2%)	12.0	17.0
Bavistin (0.1%)	9.5	11.5
Bavistin (0.2%)	7.5	8.0
Trichoderma harzianum (1%)	8.0	15.5
Trichoderma harzianum (2%)	8.0	8.0
LSD (0.05)	1.061	6.415

Table 5. Effect of different treatments on the growth parameters and yield of gladiolus in the field

Treatment	Plant height (cm)	Total no.of leaves/ plant	Total no. of infected leaves/plant	Rachis length (cm)	No. of floret/ spike	Length of floret (cm)	Dia of floret (cm)	Yield/ha in lac flower stalk
T_0	72	7	3	50	8	10	7.6	1.78
T_1	78	7	1	56	8	11	8.0	1.90
T_2	86	8	1	64	10	14	9.0	2.42
Lsd(0.05)	9.29	NS	1.66	5.32	NS	2.01	NS	0.847

T_0 = Control, T_1 = Bavistin@0.2% and T_2 = *Trichoderma harzianum* @2%

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