Screening of Selected Garlic Varieties against Fusarium Rot Caused by *Fusarium proliferatum*

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ABSTRACT

An experiment was conducted in central farm and plant pathology laboratory of Sher-e-Bangla Agriculture University (SAU) design with RCBD in field and CRD in lab to study the screening of selected garlic varieties in field condition against Fusarium rot disease caused by Fusarium proliferatum and study the pathogenicity test by inoculation of Fusarium proliferatum in one selected garlic varieties. In field, the entire experimental plot was divided into three blocks, each of which then divided into 24-unit plots. A total eight varieties viz. BAU Rashun-1, BAU Rashun-2, BARI Rashun-1, BARI Rashun-2, BARI Rashun-3, BARI Rashun-4, Local Deshi and Local Indian were selected to conduct the study. In this study, the highest preemergence mortality was observed in BAU Rashun-1 garlic variety with 31.35% germination failure and the lowest were found in BARI Rashun-1 with 14.19% germination failure. The maximum result was found for the post-emergence mortality in BARI Rashun-3 with 13.22% mortality rate after germination and minimum result were found in Local Indian variety 6.15%. In case of disease incidence and disease severity, BARI Rashun-4 showed the highest (25.00%) and (15.57%) susceptibility to Fusarium rot, respectively. Significant growth and yield parameters variations were showed during field experiment. After isolation of Fusarium proliferatum from infected leaf, maximum radial mycelial growth was observed in BARI Rashun-3 (9 mm) compared to others in PDA medium. Pathogenicity test was done with selecting one susceptible variety viz-BARI Rashun-4 in in-vivo condition by inoculating Fusarium proliferatum as soil inoculation. In this condition, 29.62% pre emergence mortality and 49.37% dead garlic seedling was recorded after germination.

Keywords: Fusarium rot, Fusarium proliferatum, Incidence and Severity, Pathogenicity test.

I. INTRODUCTION

Garlic is a species in the onion genus, *Allium*. Its close relatives include the onion, shallot, leek, chive, and Chinese onion. Garlic has been used since the

ancient times for food flavoring and as a traditional medicine. It is the second most widely cultivated and used Allium species and possesses second position after onion [1]. This bulb plant is widely used as a spice in a variety of processed foods and also used as a base for herbal medicine [2-8]. It is one of the oldest species and its positive effect on human has been known for thousand years. Today, garlic cultivation is distributed throughout most regions of the temperate world. The earliest indication of the use of garlic is in clay models in Egyptian cemeteries, dated to as early as 3,750 B.C. [9]. Garlic is also mentioned in the literature of Ancient Israel (The Talmud) and in the Bible during the time of the exodus. The Romans also extolled the virtues of garlic. Pliny the Elder, a Roman naturalist, described in his Historia Naturalis how garlic could be used for gastrointestinal disorders, dog and snake bites, scorpion stings, asthma, madness, convulsions, tumors and constipation.

Presently, China is the leading producer of garlic accounting for 20.0 million tons followed by India with 1.25 million tons per year. The other three top producing countries of garlic include; South Korea, Egypt, and Russia producing 0.35, 0.26 and 0.26 million tons respectively [10]. But the highest national yield is recorded from Armenia (40 t/ha), and the major garlic producing countries are china, South Korea, India, Spain, Egypt, United States, Thailand, Turkey and Mexico. The average yield of garlic in this country is only 2.89 t/ha [10]. The total production of garlic in Bangladesh is 425401 metric tons [11], but the requirement is 85,000 metric tons [12]. Garlic is known to be thermo- photosensitive [13] and its vegetative growth and bulb development are greatly influenced by growing environment [12].

The yield and quality of garlic bulb depend on several factors like diseases, insects, soil and climatic condition. The incidences of fungal diseases of garlic reported in Bangladesh were purple blotch complex (*Alternaria porri* and *Stemphylium vesicarium*), black mould of onion (*Aspergillus niger*), grey mould rot (*Botryotinia fuckeliana* and *Botrytis aclada*), leaf spot (*Colletotrichum dematium*), anthracnose (Glomerella cingulata), charcoal rot (Macrophomina phaseolina), cottony soft rot (Sclerotinia sclerotiorum), basal rot (Fusarium oxysporum) and fusarium rot (Fusarium proliferatum). Among these diseases, the purple blotch for garlic, and Fusarium rot and blast were more damaging than others. But, these diseases were reported as minor diseases of onion and garlic and caused damage with low infection intensity in Bangladesh [14].

The increasing of garlic production is closely related to the availability of land and the presence or absence of plant pests and diseases. One of the diseases that are often found in the field of garlic is stem or root rot caused by microorganisms, both by fungi, bacteria and viruses. One of the fungi that causes stem or root rot is *Fusarium* This fungus lives cosmopolite on the soil and can attack on roots, bulb and stems of garlic plants [5, 15-18]. As a result the plant will wither and end in death.

Fusarium is a group of the most common plant pathogens occurring all over the world and damaging crop yield. Fusarium proliferatum is a very important representative among Fusarium genus and the species has the ability to infect a wide range of hostplants, including asparagus, maize, garlic, wheat, pea, pineapple, banana and many more [19]. It is mainly transferred and spread by seeds and crop residues. F. proliferatum biosynthesizes multiple mycotoxins, such as: beauvericin, moniliformin, fusaric acid and highly toxic fumonisins from group B. Moreover, this pathogen can survive in many ecological niches, but the optimal environment is a warm and humid climate as well as loam soils with pH of about 5 [20]. Such characteristics may indicate a high plasticity and excellent adaptability of F. proliferatum to environmental challenges. Morphological characteristics based on shape and size of microconidia and macroconidia, phialides and the formation of chlamydospores [21] are the main form of identification and taxonomic description of Fusarium species [22]. Additionally, pathogenicity test considering disease incidence and severity are faster and more reliable to obtain proper identification of F. proliferatum. Varietal screening could be performed against some selected varieties of garlic to measure disease incidence and severity. In 2000 and 2001, from forty-one field samples of wilting onion and garlic plants in Serbia, F. proliferatum as the predominant fungal species was isolated from root and bulbs. Adequate identification of this pathogen is necessary to apply appropriate measures to reduce its negative effects [23]. Management of Fusarium infections is very difficult because no effective fungicides against Fusarium are available and multi-stage actions including plant and soil protection as well as inoculum reduction are necessary [24, 20].

Many fungi limit garlic production in Bangladesh. This study aims to identify the main *Fusarium proliferatum* species associated with garlic plant in field in order to determine disease intensity of Fusarium rot, characterization of its isolates and pathogenicity test.

The objectives of this study are-

- 1. To evaluate different garlic verities against fusarium rot caused by *Fusarium proliferatum* at field conditions.
- 2. To isolate and identify *Fusarium proliferatum* in lab; and
- 3. To study the pathogenicity test of fusarium rot disease of garlic by inoculation of *Fusarium proliferatum*.

II. MATERIALS & METHODS

A. Description of the study area

The experiment was conducted in Central Farm and Plant Pathology Lab, Sher-e Bangla Agricultural University, Dhaka-1207, Bangladesh from October' 2018 to October' 2019. Geographically the experimental area was located at 23°41'N latitude and 90°22'E longitudes at the elevation of 8.6 m above the sea level. The experiment was conducted by using RCBD (Randomized Complete Block Design) and 8 varieties (treatments) were experimented with 3 replications. The entire experimental plot was divided into three blocks, each of which then divided into 24unit plots. The size of the unit plot was 2.40 m^2 . Two adjacent unit plots and blocks were separated by 0.25 m, plant-to-plant distance 15 cm and line-to-line distance of 20 cm. The treatments (8 Varieties) were distributed randomly among the unit plots of each block.

B. Intercultural operation

Several intercultural operations were done during the field experiment and the parameters were considered in the study were- % Germination, % Pre-emergence mortality, % Post-emergence mortality, % Total mortality, Disease incidence (%), Disease severity (%), No. of leaf, Leaf length (cm), Bulb length (cm), Bulb diameter (cm), Bulb neck diameter (mm), Clove length (mm), Clove diameter (mm), No. of clove bulb⁻¹, Bulb dry weight plant⁻¹ (gm), average bulb weight plant⁻¹ (gm), Dry matter percentage, total Yield (Kg/ha). A list of formula given below which were used in the experiment for data calculation-

%	Pre	emergence	mo	ortalit	y=
Number	of cloves p	$\frac{1}{2}$ X 100		[25]	-
Total se	edlings en	nerged A 100		[23]	
%	Post	emergence	mortality		=
Number	of dead se	edlings v 100		[25]	
Total se	edlings en	nerged A 100		[23]	
Disease Numb	er of disea	Incidence sed plants V 100	(DI)		=
Number	of total pla	ints observed A 100			
[26]					

Disease	Severity	(DS)	=
Total p	oint score v	100	
Total no of plan	t x maximum grade $^{\Lambda}$	100	
[26]	-		

C. Isolation and determination of mycelial growth of Fusarium on PDA media

Eight isolates of *Fusarium proliferatum* were obtained from Fusarium rot infected garlic each variety. Isolations were made from Fusarium rot infected garlic leaf part on Water agar (WA) medium (Distilled water=1L, Dextrose=20g, Agar=20g) and after 3 days when mycelia was grown then it transferred into potato dextrose agar (PDA) medium (Potato=200g, Dextrose=20 g, Agar=20 g and Distilled water=1 L). In order to get a huge amount of inoculum of *Fusarium proliferatum* f. sp. isolates each isolate was sub-cultured on PDA medium and incubated at least 10 days of incubation, inoculum (mycelial mat and spores) were scraped by a plastic scrapper, wrapped with aluminum foil and preserved in the room temperature.

Pathogenicity test was conducted in *in vivo* inoculation of pathogen on growth media (soil) to see the mortality of selected garlic variety (BARI Rashun 4).

D. Statistical Analysis

The data obtained for different parameters were statistically analyzed to observe the significant difference among the treatment by using the MSTAT-C computer package program. The mean values of all the characters were calculated and analyses of variance were calculated. DMRT test was performed to determine the level of significant differences and to separate the means within the parameters at 5% level of probability [27].

III. RESULTS

A. Evaluation of selected garlic varieties on percent germination and seedling mortality in field

Significant variations of percent germination, pre and post emergence mortality and total mortality were found in eight varieties of garlic collected from different sources (Table 1). In the study, the highest number of germination percentage was observed at BARI Rashun-1 (85.81%) and the lowest number of germination percentage was found in BAU Rashun-1 (68.98%). The highest pre-emergence mortality was observed at BAU Rashun-1 garlic variety with 31.35% germination failure and the lowest for BARI Rashun-1 garlic variety at 14.19% germination failure, for the post-emergence mortality for BARI Rashun-3 with 13.22% mortality rate after germination and the lowest result showed in local Indian variety with 6.15%. Maximum emergence mortality was found in BAU Rashun-1(44.04%) and minimum emergence mortality was found in local Indian variety (20.44%).

Table	1:	Evalua	tion o	f sele	cted g	arlic	varie	eties	on
percer	ıt g	germina	tion a	nd see	edling	morte	ality	in f	ield
agains	st F	Tusariun	n proli	feratı	ım				

Variety	Germ	Pre	Post	Total
	inatio	emerge	emerge	mortali
	n	nce	nce	ty (%)
	(%)	mortali	mortali	
		ty (%)	ty (%)	
BAU	68.98	31.35 a	13.02 a	44.04 a
Rashun-1	d			
BAU	77.39	22.62	11.41 a	34.68
Rashun-2	с	b		bc
BARI	85.81	14.19	6.470	20.66 e
Rashun-1	а	с	bc	
BARI	79.17	20.83	12.62 a	33.45
Rashun-2	bc	b		bcd
BARI	83.61	16.39	13.22 a	28.61
Rashun-3	ab	с		cde
BARI	72.23	21.11	11.03	38.80
Rashun-4	d	b	а	ab
Local	84.38	15.62	8.640	24.26
Deshi	а	с	b	de
Local	85.71	14.29	6.150	20.44
Indian	а	с	с	e
LSD	4.592	3.24	2.340	8.934
CV	3.29%	9.47%	12.95%	16.66%

CV = Coefficient of variance; In a column mean values having similar letter(s) are statistically similar and those having dissimilar letter(s) differ significantly as per 0.01 level of significance.

B. Percent Disease Incidence of Fusarium Rot among selected garlic varieties

Disease incidence of Fusarium rot in every plant was counted in the field also counted the infected plants and then expressed in percentage (Table 2). At 30 DAS, BARI Rashun-4 (15.00%) showed the highest percentage of disease incidence and then at 45 DAS, the highest disease incidence percentage observed in BARI Rashun-3 (18.22%) but at 60 DAS, BARI Rashun-4 (25.00%) again showed the highest disease incidence which is most likely severe than other varieties. On the contrary, Local Indian variety showed the lowest incidence all the three times like at 30 DAS (2.23%), at 45 DAS (2.78%) and at 60 DAS (3.80%).

Table 2: Percent disease incidence of Fusarium rotat 30, 45 and 60 DAS among selected garlicvarieties

Variety	Disease Incidence					
	30 DAS	45 DAS	60 DAS			
BAU	6.67 f	8.33 f	12.22 f			
Rashun-1						
BAU	8.89 e	11.48 c	14.45 d			
Rashun-2						
BARI	10.00 d	10.167 e	13.33 е			
Rashun-1						
BARI	8.89 e	10.56 d	11.67 f			
Rashun-2						
BARI	10.55 c	18.22 a	20.33 c			
Rashun-3						
BARI	15 .00 a	16.82 b	25.00 a			
Rashun-4						
Local	12.78 b	16.67 b	22.20 b			
Deshi						

Local Indian	2.23 g	2.78 g	3.80 g
LSD	0.706	0.1767	0.7066
CV	0.04	0.86	2.66
		_	

CV=Coefficient of variance; In a column mean values having similar letter(s) are statistically similar and those having dissimilar letter(s) differ significantly as per 0.01 level of significance.

C. Percent Disease severity of Fusarium rot disease among selected garlic varieties

For measuring of disease severity of selected garlic variety 60 plants were considered and results are shown in Figure-1 and Figure-2. At 30 DAS, 45 DAS and 60 DAS BARI Rashun-4 showed the highest disease severity which was 5.33%, 11.33% and 15.67%, respectively Whereas Local Indian variety showed the lowest percentage of disease severity at 30, 45 and 60 DAS which was 0.33%, 2.67% and 3.33%, respectively.





(Vertical bars represent the standard errors of the treatment means at $P \le 0.01$)

Figure-2: Disease severity measurement of *Fusarium* proliferatum

Figure 1: Disease severity (%) of Fusarium rot at 30, 45 and 60 DAS among selected garlic varieties

D. Effect of Fusarium rot disease on growth and yield parameters among selected garlic varieties

i. Growth Parameter

Significant differences were found in growth parameters such as Leaf length (cm) and Number of leaf during experiment are shown in Table 3.

ii. Leaf length (cm) and Number of leaf

The longest leaf at 45 DAS was recorded from same variety Local Indian (26.34 cm) but the shortest one observed from BAU Rashun-2 (17.07cm). Number of leaf was recorded at 45 DAS. At 45 DAS BARI Rashun-1 gave the highest no. of leaves (7) BARI Rashun-4 variety gave the lowest number of leaves (5.14).

iii. Yield Parameters

Significant variations were found in yield parameters like Bulb length (cm), Bulb diameter (cm), Clove length (cm) and No of clove bulb⁻¹ during experiment are shown in (Table 3).

⁷ Bulb length (cm), Bulb diameter (cm), Clove length (cm) and No of clove bulb⁻¹

The present experiment indicates BARI Rashun-1 as the longest bulb length of 2.76 cm and for shortest local Indian with 2.10 cm was found. Higher bulb diameter indicates better yield. BAU Rashun-2 showed the largest diameter of 1.53 cm and the lowest diameter was found in BARI Rashun-1 variety. The longest clove was obtained from BARI Rashun-1 and the shortest was collected from local Indian of 2.10 cm which is statistically similar to Local Deshi variety. The highest no. of clove producer in a single bulb was BARI Rashun-1 of 16 cloves and the lowest was from BARI Rashun-4 of 9 cloves per bulb only. The highest fresh weight was collected from BAU Rashun-1 which is statistically similar to BARI Rashun-3. The lowest one was collected from Local Indian variety as it was failed to

produce any bulb. The highest fresh weight was collected BARI Rashun-3. Highest dry matter found in of BARI Rashun-1 (71.67%) and the lowest from Local Indian variety (Table 3). Highest yield producing variety was BARI Rashun-3 of 3307.5 kg/ha production.

Variety	Leaf length (cm)	No. of leaves	Bulb length (cm)	Bulb diameter (cm)	Clove length (cm)	No. of clove/ bulb
BAU Rashun-1	18.05 de	6.84 b	2.433 ab	1.20 ab	2.300 a	11.333 ab
BAU Rashun-2	17.07 e	6.070 f	2.167b	1.53 a	2.233 a	13.333 ab
BARI Rashun-1	19.14 bcd	7.00 a	2.767 a	1.40 ab	2.467 a	11.667 ab
BARI Rashun-2	17.74 de	6.57 cd	2.367 ab	1.00 ab	2.400 a	14.000 a
BARI Rashun-3	19.81 bc	6.50 d	2.233 b	1.43 ab	2.233 a	16.333 a
BARI Rashun-4	18.20 cde	5.14 g	2.267 b	1.23 ab	2.400 a	7.667 b
Local Deshi	20.55 b	6.80 bc	2.267 b	1.13 ab	2.467 a	13.667 ab
Local Indian	26.34 a	6.30 e	2.1 b	1.33 ab	2.1 a	14.667 a
LSD	1.6909	0.115	0.4593	0.4290	0.4593	6.0375
CV	4.98	0.0884	11.41	19.12	11.41	27.18

Table 3: Effect of Fusarium rot on growth and yield parameters for selected garlic variety

CV=Coefficient of variance; in a column mean values having similar letter(s) are statistically similar and those having dissimilar letter(s) differ significantly as per 0.01 level of significance.

E. Effect of fusarium rot on bulb fresh weight (kg/ha), dry weight (kg/ha), (%) dry matter and total total yield (kg/ha) of selected garlic varieties in field condition

Significant variations were found among different varieties of garlic in different yield parameters due to the effect of Fusrium rot, are shown in figure 3, 4, 5 and 6.

i. Bulb fresh weight (kg/ha)

The average mature bulb fresh weight per bulb was measured. The highest fresh weight was collected from BAU Rashun-1 (579.49 gm). BARI Rashun-1, BARI Rashun-2, BARI Rashun-3, BARI Rashun-4 and Local Deshi variety showed statistically similar result. The lowest one was collected from Local Indian variety (400.33 gm).

ii. Bulb dry weight (kg/ha)

The average mature bulb dry weight per bulb measured and found statistically significant. The highest fresh weight was collected BAU Rashun-1 (315.333 gm). The lowest one was collected from BARI Rashun-3 (218.667 gm).

iii. Dry matter percentage

Dry matter percentage of garlic is positively correlated with its nutritious value and yield. The dry matter percentage of selected garlic variety was measured and found statistically significant. Highest dry matter found in of Local Indian variety (70.930%) and the lowest from BARI Rashun-1 (38.353%).

iv. Total yield (Kg/ha)

Total bulb yield of plants grown in three central rows was measured after bulbs were cured or exposed for ten days to sunlight. The yields obtained from plots were converted to kilogram per hectare. The calculated data was statistically significant. Highest yield producing variety was BARI Rashun-2 of 2891.820 kg/ha production. Lowest Yield was gathered from BARI Rashun-3 of 2206.347 kg/ha.



Vertical bars represent the standard errors of the treatment means at $P \le 0.01$ Figure 3: Average bulb fresh and dry weight (g) per plant for selected garlic varieties



Vertical bars represent the standard errors of the treatment means at $P \le 0.01$

Figure 5: Yield performance for selected garlic variety



Vertical bars represent the standard errors of the treatment means at $P \le 0.01$

Figure 6: Yield loss for selected garlic variety



Vertical bars represent the standard errors of the treatment means at $P \le 0.01$ *Figure 4: Dry matter percentage for selected garlic varieties*

F. Effect of fusarium rot on total yield and yield loss among different garlic varieties

Significant variations of total yield and yield loss were found in different varieties of garlic in field condition due to Fusrium rot disease. The results of variations are shown in Table 4.Yield loss was compared with expected yield and BAU Rashun-2 (86.08%) showed the poor performance whereas from BARI Rashun-1 (26.04%) best result was obtained.

Table 4: Effect of expected yield, total yield, yield loss (Kg/ha) and (%) yield loss for selected garlic variety due to fusarium rot

Variety	Expecte	Total	Yield	Yield
	d yield	yield	loss	loss
	(kg/ha.)	(kg/ha.)	(kg/ha.)	(%)
BAU Rashun-1	11000	3101.68	7898.32	78.90
BAU Rashun-2	11000	2393.84	8606.16	86.08
BARI Rashun-1	6500	2895.053	3604.94	26.04
BARI Rashun-2	8500	2592.72	5907.28	59.07
BARI Rashun-3	10905	3307.50	7597.50	75.97
BARI Rashun-4	8500	1972.84	6524.16	65.24
Local Deshi	5000	2263.28	2736.72	27.37
Local Indian	7500	1635.48	5865.00	58.64

In a column mean values having similar letter(s) are statistically similar and those having dissimilar letter(s) differ significantly as per 0.01 level of significance.

G. Correlation of leaf height (cm) and yield parameters

To determine the effect of growth parameters on yield of selected garlic varieties correlation of coefficient was considered at 0.01% level of probability. Here leaf height (cm) showed negative correlation with the entire yield parameters considered and become significant at 0.01% level of probability. These values clearly express less leaf height give higher yield and longer leaf cause degradation of garlic yield performance (Figure 7).



Figure 7: Correlation of leaf height (cm) and yield parameters

H. Correlation of no. of leaf with yield parameters

The effect of growth parameters on yield of selected garlic verities correlation of coefficient was considered at 0.01% level of probability. Here no. of leaf showed positive correlation with all the yield defining characters except clove diameter (-.859). These value express better performances of yield can be considered as predeterminer of better yield but narrower cloves (Figure 8).



Figure 8: Correlation of no. of leaf with yield parameters

I. Isolation, Identification and pathogenicity test of Fusarium rot caused by Fusarium proliferatum

Mycelial growth and cultural characters of eight isolates of *Fusarium proliferatum* at 2, 5 and 7 days on PDA media are shown in Figure 9 and Figure 10. After 7 days of inoculation, the maximum radial mycelial growth of *Fusarium proliferatum* was observed in BAR3I (9 mm). The minimum radial mycelial growth was BAR2I (5.25 mm).



Figure 9: Mycelial growth of 8 isolates of F. proliferatum at different days on PDA media



Figure 10. Radial mycelial growth of Fusarium proliferatum on PDA media after 7 days Pathogenicity test for Fusarium rot

During screening of fusarium rot disease in garlic BARI Rashun-4 found highest disease incidence and severity which were selected for pathogenicity test. BARI Rashin-4 showed highly susceptible with *Fusrium proliferatum* that showed 29.62% germinated seedling and 49.37% dead seedling (Figure 11 & Figure 12).



Figure 10. Pathogenicity test of Fusarium proliferatum in pot



Figure 11. Symptom on garlic seeding after soil inoculation

IV. DISCUSSION

In the following study, the highest number of germination percentage was observed at BARI Rashun-1 85.81% and the lowest number of germination percentage was found in BAU Rashun-1 68.98%. The highest pre emergence mortality was observed at BAU Rashun-1 garlic variety with 31.35% germination failure and the lowest for BARI Rashun-1 garlic variety at 14.19% germination failure, for the post emergence mortality for BARI Rashun-3 with 13.22% mortality rate after germination and the lowest result showed in local Indian variety with 6.15% mortality rate after germination. Maximum emergence mortality was found in BAU Rashun-1 with 44.04% and minimum emergence mortality was found in local Indian

variety 20.44%. The finding of the present study reflects the finding of [28] mentioned that garlic is typically propagation via seed cloves but planting of bulb is an alternative means of propagation for cultivars that produce them. We wished to know if the infection rate in bulbils would differ from the infection rate in cloves, when both bulbils and cloves originated from plants whose bulbs were known to be infected by *F. proliferatum* as a major cause of pre emergence and post emergence as well as total mortality of garlic.

Highest disease incidence showed in BARI Rashun-4 at different day's interval and lowest disease incidence showed in Local Indian Variety at different day's interval. At 30 DAS, 45 DAS and 60 DAS BARI Rashun-4 showed the highest disease severity which was 5.33%, 11.33% and 15.67%, respectively whereas Local Indian variety showed the lowest percentage of disease severity at 30, 45 and 60 DAS which was 0.33%, 2.67% and 3.33%, respectively. The following results supports the report of [29] that, garlic clove rot caused by F. proliferatum has become a limiting factor to garlic production indifferent growing areas of Egypt and other countries, such as China, the largest garlic producer in the world and [30] reported that as fresh garlic is consumed worldwide, the production of mycotoxins in cloves infected with F. proliferatum requires serious consideration.

In pathogenicity test of Fusarium proliferatum BARI Rashun-4 showed highly susceptible with Fusrium proliferatum that showed 29.62% germinated seedling and 49.37% dead seedling. This result is similar with the findings of [31] in 2000 and 2001, from forty-one field samples of wilting onion and garlic plants in Serbia, F. proliferatum as the predominant fungal species was isolated from root and bulbs. Seventy isolates were firstly characterized for their sexual fertility and were shown to be mostly members of Gibberella intermedia (sixty-seven of seventy isolates, the remaining three isolates were unfertile), the sexual stage of F. proliferatum (syn. mating population D of G. fujikuroi complex). A selected set of eleven F. proliferatum isolates from both hosts were also tested for their pathogenicity and toxigenicity. Six of the eleven isolates of F. *proliferatum* produced fumonisin B_1 from 25 to 3000 μ g g⁻¹, and beauvericin from 400 to 550 μ g g⁻¹; ten isolates produced fusaric acid from 80 to 950 μ g g⁻¹ and moniliformin from 50 to 520 μ g g⁻¹. Finally, all isolates produced fusaproliferin up to 400 μ g g⁻¹. These results confirm *F. proliferatum* as an important pathogen of garlic and onion in Europe and that there is a potential mycotoxin accumulation risk in contaminated plants of both garlic and onion.

V. CONCLUSION

Fusarium proliferatum has been identified as the main causal agent of bulb rot of garlic (Allium sativum L.). This disease occurs after the drying process and can rot almost 30 % of the bulbs. This particular experiment was done in order to screen the selected garlic varieties against fusarium rot disease caused by Fusarium proliferatum. From this experiment we have recorded the highest pre emergence mortality was observed at BAU Rashun-1 garlic variety with 31.35% germination failure and the maximum result was found for the post emergence mortality for BARI Rashun-3 with 13.22% mortality rate after germination. For disease incident data was measured considering 60 plants and for BARI Rashun-4 showed the highest result at 30 DAS and for the minimum incidence data local Indian variety showed the lowest result at 30 DAS, 45 DAS and 60 DAS, respectively. For measuring of disease severity of selected garlic variety at 30 and 60 DAS maximum result observed in BARI Rashun-4 variety. The maximum radial mycelial growth of Fusarium proliferatum was observed in BAR3I (9 mm).

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Conflict of interest

The authors declare no conflict of interest.

Ethical issues

There are no ethical issues involved in this research work.

Author contributions

N. P. Jannatun conducted the research and wrote the article; U. A. Habiba collected the *Fusarium* samples from field helped author in *in vivo* and *in vitro* experiment and analyzed the data; B. Fatema designed and supervised the study and edited the manuscript; Md. Rafiqul Islam and M. Morshed read the manuscript contributed to the conceptualization, and methodology of the study.

References

- [1] Bose, T.K. and M.G. Som, 1990. Vegetable Crops in India. 1st Edn., Naya Prakash Calcutta, India, pp: 583-601.
- [2] Irkin R and Mihriban K. 2007. Control of Aspergillus niger with garlic, onion and leek extracts. Afr. J. of Biotech. 6(4): 384-87.
- [3] Kumar K.P.S., Dedjit B., Chiranjib, Pankaj T., Rakesh K. 2010. Alium sativum and its health benefits: An overview. J. Chem. Pharm. Res., 2(1): 135-46.: 656-62.
- [4] Bisen P.S. and Mila E. 2016. Nutritional and therapeutic potentioal of Garlic and Oniuon (Allium sp.). Curr. Nutri. & Food Sci. 12: 190-199.

- [5] Alzowahi F.A.M., Ahmed AT. Amani AS and Kadam T.A. 2013. The inhibitory effects of garlic extract and its fractions against some Enterobacteriaceae sp isolated from sprouted Mung bean. Int. J. Curr. Microbiol. App. Sci. 2(7): 104-15.
- [6] Gebreyohannes G. And Mebrahtu G. 2013 Medicine values of garlic: A review. Int. J. of Med. and Medic. Sci. 5(9): 401-408.
- [7] Queiroz Y.S., Ishimoto E.Y., Bastos D.H.M., Geni R.S. and Elizabeth A.F.S.T. 2009. Garlic (Alium sativum L.) and ready-to- eat garlic products: In vitro antioxidant activity. Food Chem. 115: 371-74.
- [8] Neeraj S., Kaura S., Dilbaghi N., Parle M. And Pal. M. 2014. Garlic: A pungent Wonder from Nature. Int. Res. J. Pharm. 5(7): 523-29.
- [9] Woodward, P. D. (1996). Garlic and friends: the history, growth and use of edible *Alliums*. Hyland House, South Melbourne, Australia. P: 34-36.
- [10] Food and Agricultural Organization (FAO), (2003). Global review of area and production of garlic, Year Book. pp: 135-139.
- [11] BBS (Bangladesh Bureau of Statistics), (2018). Yearbook of Agricultural Statistics of Bangladesh, Statistics Division, Ministry of Planning, Government of the People's Republic of Bangladesh, Dhaka, Bangladesh.
- [12] Rahim, M.A. 1992. Spices and plantation crops in national economy. In:Horticulture in national development. Proceedings of the Sixth National Horticulture Convention and Symposium. Bangladesh Soc. Hort. Sci., pp. 24-29.
- [13] Rahim, M.A. and R. Fordham. 1988. Effects of storage temperature on the initiation and development of garlic cloves (Allum sativum L.). Scientia Hort., 37: 25-38.
- [14] USDA. (2014). Garlic, *Allium sativum*. | United States Department of Agriculture (USDA).
- [15] Dugan F.M. 2007. Diseases and disease management in seed garlic: problems and prospects. The Am. J. of P. Sci. and Biotec. 1(1): 47-51
- [16] Palmero D., Miguel D.C., Walid N., Laura G., Alejandra C., Stephen W., Maria T.GJ. and Julio C.T., 2012. Fusarium proliferatum isolated from garlic in Spain: Identification, toxigenic potentioal and pathogenicity on related Allium species. Phytop. Mediter. 51(1): 207-18.
- [17] Amini J. And Dzhalilov F.S. 2010. The effects of fungicides on Fusarium oxysporum f.sp.lycopersici associated with Fusarium wilt on tomato. Journal of Plant Protection Research 50(2): 272-78.
- [18] Chand S.K., Satyabrata N. and Rai K.J. 2016. Regulation of miR394 in response to Fusarium oxysporum f. Sp. Cepae (FOC) infection in Garlic (Allium sativum L.). Frontiers in Plant Science 7: Article 258
- [19] St epie 'n, Ł.; Wa'skiewicz, A.; Urbaniak, M. Wildly growing asparagus (Asparagus officinalis L.) hosts pathogenic Fusarium species and accumulates their mycotoxins. Microb. Ecol. 2016, 71, 927–937. [CrossRef] [PubMed]
- [20] Vujanovic, V.; Hamel, C.; Yergeau, E.; St-Arnaud, M. Biodiversity and Biogeography of Fusarium Species from Northeastern North American Asparagus Fields Based on Microbiological and Molecular Approaches. Microb. Ecol. 2006, 51, 242–255. [CrossRef] [PubMed]
- [21] Rodrigues, A.A.C., Menezes, M. Identification and pathogenic characterization of endophytic *Fusarium* species from cowpea seeds. *Mycopathologia* 159, 79–85 (2005). https://doi.org/10.1007/s11046-004-7138-x.
- [22] Leslie, J.F. and Summerell, B.A. (2006). The Fusarium Laboratory Manual. Blackwell Publishing, Ames, Iowa, USA.
- [23] Takken F, Rep M. The arms race between tomato and Fusarium oxysporum. *Mol Plant Pathol*. 2010;11(2):309-314. doi:10.1111/j.1364-3703.2009.00605.x.
- [24] Pontaroli, A.C.; Camadro, E.L. Increasing resistance to Fusarium crown and root rot in asparagus by gametophyte selection. Euphytica 2001, 122, 343–350. [CrossRef]
- [25] Zagade, S & Deshpande, G & Gawade, Dattatray & Atnoorkar, A. (2013). Studies on pre-and post-emergence

damping off of Chilli caused by *pythium ultimum*. 41:332-337.

- [26] Manandhar, H.K., Timila, R.D., Sharma, S., Joshi, S., Manandhar, S., Gurung, S.B., Sthapit, S., Palikhey, E., Pandey, A., Joshi, B.K., Manandhar, G., Gauchan, D., Jarvis, D.I. and Sthapit, B.R. (2016). A field guide for identification and scoring methods of diseases in the mountain crops of Nepal. NARC, *LI-BIRD and Biover. Int.*, P: 12.
- [27] Gomez, K.A. and Gomez, A.A. (1984). Statistical Procedure for Agricultural Res. (2nd Ed.) John Wiley and Son. Singapore., pp. 188-192.
- [28] Ibrahim, Totic & Stevan, Čanak, 2014. "Production And Economic Specificities In Growing Of Different Garlic Varieties," Economics of Agriculture, Institute of Agricultural Economics, vol. 61(4), pages 1-14, December.
- [29] Dugan F.M., Hellier, B.C. and Lupien, S.L. (2007). Pathogenic fungi in garlic seed cloves from the United States and china, and efficacy of fungicides against pathogens in garlic germplasm in washington state. J. Phytopatho., (155) 437-445.
- [30] Stepien, L., Koczyk, G. y Waskiewicz, A. (2011). Genetic and phenotypic variation of *Fusarium proliferatum* isolates from different host species. *J. App. Gen.*, 52: 487-496.
- [31] S. Stankovic, J. Levic, T. Petrovic, A. Logrieco & A. Moretti. 2007. Pathogenicity and mycotoxin production by *Fusarium proliferatum* isolated from onion and garlic in Serbia *European Journal of Plant Pathology* volume 118, pages165–172.