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OCCURRENCE OF ANTHRACNOSE DISEASE INCIDENCE AND ITS SEVERITY ON GUAVA SEEDLINGS AT DIFFERENT LOCATIONS OF BANGLADESH

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ABSTRACT

Islam MN, Chowdhury MSM, Rahman MM, Alam KM, Arifunnahar M, Alam MM (2015) Occurrence of anthracnose disease incidence and its severity on guava seedlings at different locations of Bangladesh. *Int. J. Sustain. Crop Prod.* 10(3), 21-31.

Guava, a major fruit crop of Bangladesh, was surveyed during the period of July, 2010 to April, 2012 for the incidence of anthracnose disease and its severity. The study was carried out to observe the effect of temperature, relative humidity and rainfall on the incidence of anthracnose disease and its severity at seedling stage. The causal pathogen of anthracnose, *Colletotrichum gloeosporioides* was identified and its incidence and severity on guava at seedling stage was studied. A significant variation in development of the disease was observed in varied weather parameters. Occurrence of seedling anthracnose disease was positively correlated with temperature, rainfall and relative humidity. Therefore there were great variation of disease incidence and its severity on guava was evident from one location to another and season to season. These weather parameters should be critically addressed for the host-pathogen interaction and to find out the most appropriate time for combating the disease at minimum cost.

Key words*: anthracnose, Colletotrichum gloeosporioides, disease incidence, guava and severity etc.*

INTRODUCTION

Guava (*Psidium guajava* L.), is one of the most common fruits in tropical and sub-tropical regions of the world. The tropical guava is the best adapted to a warm climate, thriving in both humid and dry seasons. It is adaptable over a wide range of climatic and soil conditions (Purseglove 1984). It can be used as an important economic resource for local communities with potential for commercialization, boosting food and nutritional security. In rural areas, guava is used to supplement the daily diet and it substitutes exotic fruits. Guava has been found to be beneficial for people suffering from Asthma, high blood pressure, oral ulcers, scurvy, congestion of the lungs, bacterial infection etc. Bangladesh produces less than 30% of the fruits needed to meet the minimum daily requirements for its population. They are the chief sourch of some vitamins, mineral salts and also possess high medicinal values (Kamaluddin 1994). About 80% of families in the country consume less than the minimum recommended daily requirement of fruits. As a consequence widespread nutritional deficiencies in vitamin 'A' and 'C' and other nutrients cause debilitating illness among the people (HKI 2005). The situation can be improved by growing guava fruit trees and increasing the production of guava which may fulfill the requirements of people of Bangladesh.

Bangladesh produces 182 thousand metric tons of guavas annually (BBS 2010). Quantity and quality of guava in this country is far below the world standard. There are several factors responsible for low yield and poor quality of guava in Bangladesh, where diseases infection is the most important. It has been estimated that the production could be increased at least by 28% if the crop could be protected from various seedling diseases (Chowdhury 2009). The climate of Bangladesh harbors plant pathogens and provide luxuriant environment for the growth and reproduction of large number of plant pathogens causing hundreds of different diseases (Fakir 2001).

Like other fruits healthy seedlings are prime need and basic raw material for establishment of orchard for the successful production guava. Seedling diseases are important consideration for guava production. Seedling is frequently affected by physical and physiological disorders as well as diseases caused by fungi, bacteria and viruses (Mittal and Mathur, 1990). Seed borne pathogens affect nursery seedlings and reduce seedling vigor (Abdelmonem and Rasmi, 2003). So, seedling diseases of guava are the important problems in the tropics. Although a huge number of nurseries are engaged in producing seedlings, they fail to produce quality seedlings due to lack of their knowledge about diseases. Seedling of guava are affected by diseases such as wilt (*Fusarium oxysporium*), anthracnose (*Colletotrichum gloeosporioides*), seedling blight, fruit rot, *Phoma* rot, *Rhizopus* rot, collar rot, *Pestalotia* leaf spot, *Cercospora* leaf spot, stem canker , sooty mold, die back (Rahman *et al*. 2003).

The existing technology of guava cultivation in the country is in a stage that needs to be upgraded for successful guava production in order to meet up the national demand. So, studies on the seedling disease of guava are of urgent need in the country. Therefore, the attempts were taken to study the prevalence of anthracnose disease occuring on guava seedlings in some selected nurseries of Dhaka, Gazipur, Barisal and Khagrachari and the effect of temperature, relative humidity and rainfall on incidence and severity of anthracnose disease of guava were undertaken.

Considering the above facts, the present research work was designed with the following objectives : i) to survey on the prevalence of anthracnose disease of guava seedling in some selected nurseries of Dhaka, Gazipur,

Barisal and Khagrachari, ii) to identify the pathogen associated with the disease and iii) to study the effect of temperature, relative humidity and rainfall on the incidence and severity of anthracnose disease of attacking guava.

MATERIALS AND METHODS

Location of survey area

Prevalence of diseases incidence on guava seedlings raised in the selected nurseries were surveyed. The study was carried out in eight nurseries of Dhaka, Gazipur, Barisal and Khagrachari districts. The eight nurseries of four districts were surveyed: Dhaka (Green orchid nursery, Agargaon and Barisal nursery, Savar); Gazipur (Gazipur nursery and Laxmipur nursery); Barisal (Sarchina nursery and Riyad nursery) and Khagrachari (Ramghor Hill Research Center and Ramghor nursery).

Observation of the symptoms

Symptoms of the diseases were studied by visual observation. Sometimes hand lens was used for critical observation of the disease and sometimes a disease was identified based on matching the observed symptoms in the infected plants with the symptoms published in Ber and other guava disease compendium.

Identification of causal organisms

Collection of diseased specimen

Diseased leaves were collected from the infected plants representing the different areas of survey. The specimens were preserved in the laboratory following standard procedure of preservation of disease specimens until isolation was made. Isolation of causal organisms was made by two methods as follows:

Moist blotter method

The pathogen associated with the diseased plant parts (leaves) were cut into pieces by scissors and placed on the moist filter paper (Whitman no.1). Three pieces of filter paper were moistened by dipping in sterile water. The Petri dishes with the diseased specimens were incubated at 22±2°C under 12/12 alternating cycles of NUV and darkness in the incubation room of the Seed Pathology Lab (SPL) for three to five days. After incubation the plates were examined under stereomicroscope for primary identification of the organisms (fungi). The fungi were transferred to PDA plates for proper sporulation and purification.

Agar plate method

The diseased plant parts (leaves) were surface sterilized by dipping them in 0.001% $HgCl_2$ solution for 1.5 minutes and washed three times with sterile water and there after placed on PDA (Potato = 200g, Dextrose = 17g, Agar = 17-20g, Water = 1000 ml) plates aseptically. The plates were incubated at 28°±1°C for 15-20 days and examined daily for any fungal growth.

Epidemiology of disease incidence and severity

Survey period

Altogether eight surveys were made during the period from July, 2010 to April, 2012. Where First, second, third, fourth, fifth, sixth, seventh, and eighth surveys were made in July, 2010; October, 2010; January, 2011; April, 2011; July, 2011; October, 2011; January, 2012; and April, 2012, respectively.

Data collection during survey

During the survey in the nurseries, total numbers of guava seedlings as well as number of diseased seedlings in the nurseries were recorded. Then 30 seedlings were randomly selected for counting diseased leaves and disease free leaves. Moreover, five leaves per plant were randomly selected to determine the disease severity.

Determination of disease incidence and disease severity

For calculation of disease incidence every seedling was counted in the nursery and also counted the infected seedlings and then expressed in percentage. The disease incidence of guava seedling was determined by the following formula (Rai and Mamatha, 2005):

$$\text{Percent plant infection} = \frac{\text{Number of diseased plants}}{\text{Number of total plants observed}} \times 100$$

Percent disease incidence(PDI) of foliar diseases was determined by the following formula (Rai and Mamatha, 2005):

$$\text{Percent disease incidence (leaves)} = \frac{\text{Number of diseased leaves on each plant}}{\text{Number of total leaves on each plant}} \times 100$$

Percent disease severity (PDI) was determined by the following formula (Rai and Mamatha, 2005):

$$\text{Percent disease severity (leaves)} = \frac{\text{Area of leaf tissue infected by disease}}{\text{Total leaf area of the plant}} \times 100$$

Meterological data collection

Meterologial data of the experimental period were collected from Meterological Department, Agargaon, Dhaka.

Satistical analysis

Data on different parameters were analyzed in two factor randomized block design (RCBD) through computer software MSTAT-C (Anonymous 1989). Duncan's Multiple Range Test (DMRT) and Least Significant difference (LSD) test were performed for mean separation of different parameters and to determine the level of significant differences.

RESULTS

Survey on nursery diseases of guava

Three different diseases *viz*., anthracnose, scab and bacterial leaf blight were recorded in the survey conducted in eight nurseries of Dhaka, Gazipur, Barisal and Khagrachari districts.

Symptom of the Anthracnose disease of guava and identification of the pathogen

The characteristic symptoms consist of sunken, dark colored, necrotic lesions. Under humid conditions, the necrotic lesions become covered with pinkish spore masses. As the disease progresses, the small sunken lesions coalesces to form large necrotic patches (Plate 1 A & B).

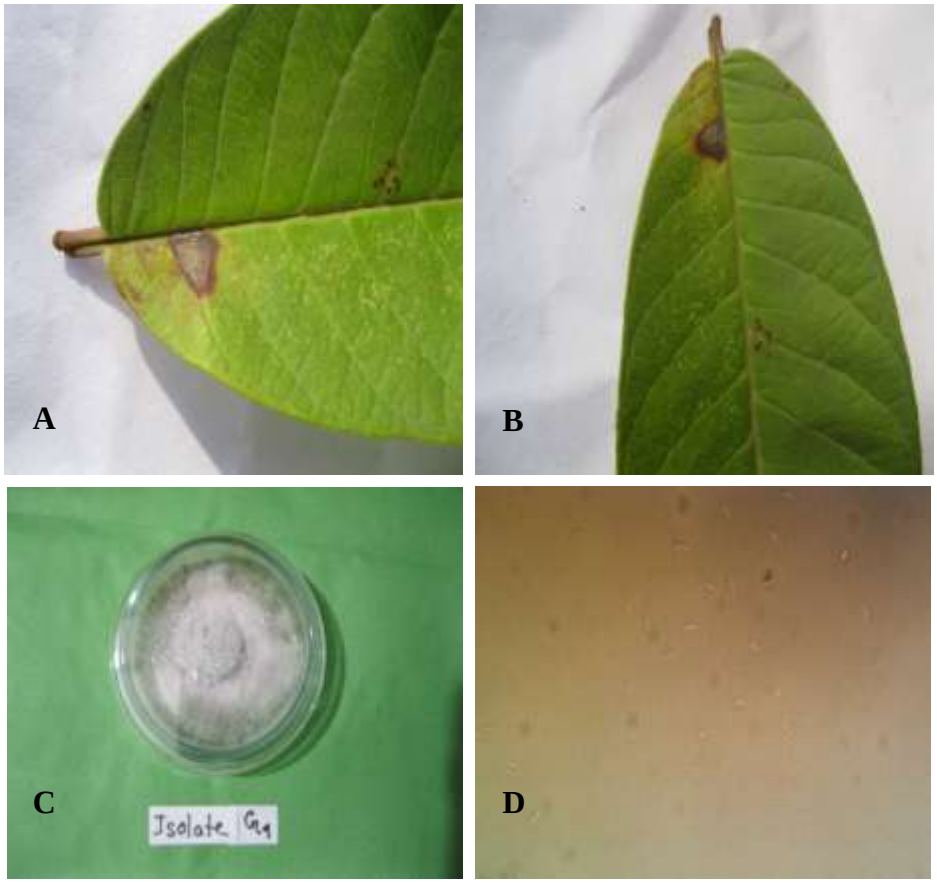

Plate 1. Symptom of the disease and identification of the pathogen. A & B showing symptoms of anthracnose of guava. C. Pure culture of *Colletotrichum gloeosporioides* (Penz.), D. Conidia of *Colletotrichum gloeosporioides* (Penz.) produce in culture

The pathogen isolated from the diseased symptom was identified as *Colletotrichum gloeosporioides* (Plate 1 C & D) by observing the dark coloured conidia with single cell barrel shaped structure.

Epidemiology of disease incidence and severity

Incidence and severity of anthracnose of guava at different locations of Bangladesh from July, 2010 to April, 2012

Incidence of anthracnose of guava varied from location to location and year to year that ranged from 25.53-22.50% in 2010-2011 and 43.93-24.38% in 2011-2012 (Table 1). The highest incidence was recorded at Khagrachari and the lowest was at Gazipur in both the years. The severity of anthracnose of guava also varied from location to location and year to year and was ranged from 19.73-15.82% in 2010-2011 and 26.46-14.62% in 2011-2012. The highest severity was recorded at Dhaka in 2010-2011 and Khagrachari in 2011-2012. The lowest was recorded at Barisal in 2010-2011 and Gazipur in 2011-2012.

Table 1. Incidence and severity of anthracnose of guava at different locations of Bangladesh from July, 2010 to April, 2012

Location	Anthracnose			
	Incidence (%)		Severity (%)	
	2010-2011	2011-2012	2010-2011	2011-2012
Dhaka	25.18 a	31.61 b	19.73 a	18.13 c
Gazipur	22.50 b	24.38 c	17.95 b	14.62 d
Barisal	23.72 b	30.14 b	15.82 c	23.16 b
Khagrachari	25.53 a	43.93 a	18.15 b	26.46 a
$LSD_{(p\geq0.05)}$	1.381	1.600	1.009	1.766
CV%	3.57	4.27	4.82	6.95

Each data represents the mean value of three nurseries

Incidence and severity of anthracnose of guava during July, 2010 to April, 2012

Incidence of anthracnose of guava varied from July, 2010 to April, 2012 and that ranged from 33.19-15.57% in 2010-2011 and 47.90%-16.30% in 2011-2012 (Table 2). Significantly the highest incidence was recorded in July, and the lowest was recorded in January, in both the years. The severity of anthracnose of guava also varied from year to year that ranged from 25.98-10.47% in 2010-2011 and 37.73-9.77% in 2011-2012. Significantly the highest severity was recorded in July, in both the years and the lowest was recorded in January, in both the years.

Table 2. Incidence and severity of anthracnose of guava during July, 2010 to April, 2012

Data recording time(month)	Incidence (%)		Severity (%)	
	2010-2011	2011-2012	2010-2011	2011-2012
July	33.19 a	47.90 a	25.98 a	37.73 a
October	27.34 b	41.19 b	21.67 b	22.27 b
January	15.57 d	16.30 d	10.47 d	9.77 d
April	20.83 c	24.67 c	13.53 c	12.59 c
$LSD_{(p\geq0.05)}$	1.459	2.339	1.455	2.412
CV%	3.57	4.27	4.82	6.95

Each data represents the mean value of three nurseries

Incidence and severity of anthracnose of guava during July, 2010 to April, 2012 at different study area of Bangladesh

Incidence of anthracnose of guava varied significantly from season to season as well as location to location and that ranged from 35.75-12.96% in 2010-2011 and 64.25-10.40% in 2011-2012 (Table 3). Significantly the highest incidence of anthracnose of guava recorded in July, 2010 (35.75%) and July, 2011 (64.25%) at Khagrachari. Again significantly the lowest (12.96% and 10.40%) incidence was observed in January, in both the years at Barisal. The severity of anthracnose of guava also varied significantly from season to season as well as location to location and that ranged from 27.63-9.40% in 2010-2011 and 52.08-5.737% in 2011-2012. Significantly the highest (27.63% and 52.08%) severity of anthracnose of guava observed in July, 2010 at Dhaka and July, 2011 at Barisal, while the lowest (9.40% and 5.737%) was recorded in the month of January, 2011 at Dhaka and January, 2012 at Barisal respectively.

Table 3. Incidence and severity of anthracnose of guava during July, 2010 to April, 2012 at different study areas of Bangladesh

Location	Data recording time(month)	Incidence (%)		Severity (%)	
		2010-2011	2011-2012	2010-2011	2011-2012
Dhaka	July	32.82 b	44.18 d	27.63 a	27.63 c
	October	29.14 c	35.49 e	21.18 d	22.17 d
	January	16.45 g	19.66 hi	9.40 i	9.960 g
	April	22.32 f	27.09 g	14.40 e	12.75 f
Gazipur	July	32.74 b	34.21 ef	23.08 c	19.59 e
	October	25.08 e	25.63 g	15.63 e	13.29 f
	January	15.86 g	16.18 j	11.81 fg	13.64 f
	April	21.21 f	21.50 h	12.76 f	11.94 fg
Barisal	July	31.44 b	48.94 c	27.26 ab	52.08 a
	October	28.48 c	44.00 d	22.69 c	22.84 d
	January	12.96 h	10.40 k	9.653 hi	5.74 h
	April	17.12 g	17.22 ij	12.18 fg	11.98 fg
Khagrachari	July	35.75 a	64.25 a	25.96 b	51.64 a
	October	26.66 d	59.62 b	27.18 ab	30.77 b
	January	17.02 g	18.97 i	11.00 gh	9.767 g
	April	22.68 f	32.87 f	14.79 e	13.68 f
$LSD_{(p\geq0.05)}$		1.459	2.339	1.455	2.412
CV(%)		3.75	4.27	4.82	6.95

Each data represents the mean value of three nurseries

Effect of weather components on the incidence and severity of anthracnose of guava seedling during July, 2010 to April, 2012

In different growing seasons of guava seedlings, the highest incidence (33.19% and 47.90%) and the highest severity (25.98% and 37.73%) of anthracnose disease were recorded in July, in both the years when the average temperature, relative humidity and rainfall were 29.65°C, 81.40% & 7.55cm and 30.50°C, 83.50% & 5.50cm, respectively. On the other hand, lowest incidence (15.57% and 16.30%) and the lowest severity (10.47% and 9.77%) were recorded in January, in both the years when the mean temperature, relative humidity and rainfall were 16.88°C, 73.80% & 0.52cm, and 18.46°C, 76% & 0.60cm, respectively (Figs. 1 & 2).

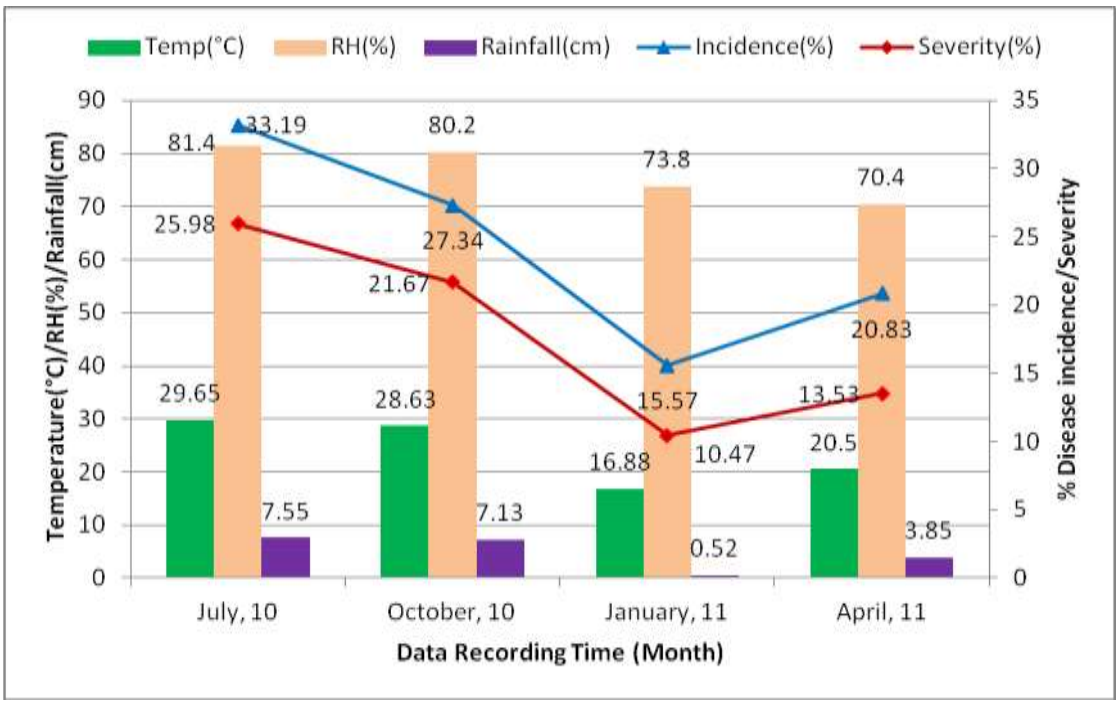

Fig. 1. Effect of different weather parameters on the incidence and severity of anthracnose of guava seedling during July, 2010 to April, 2011

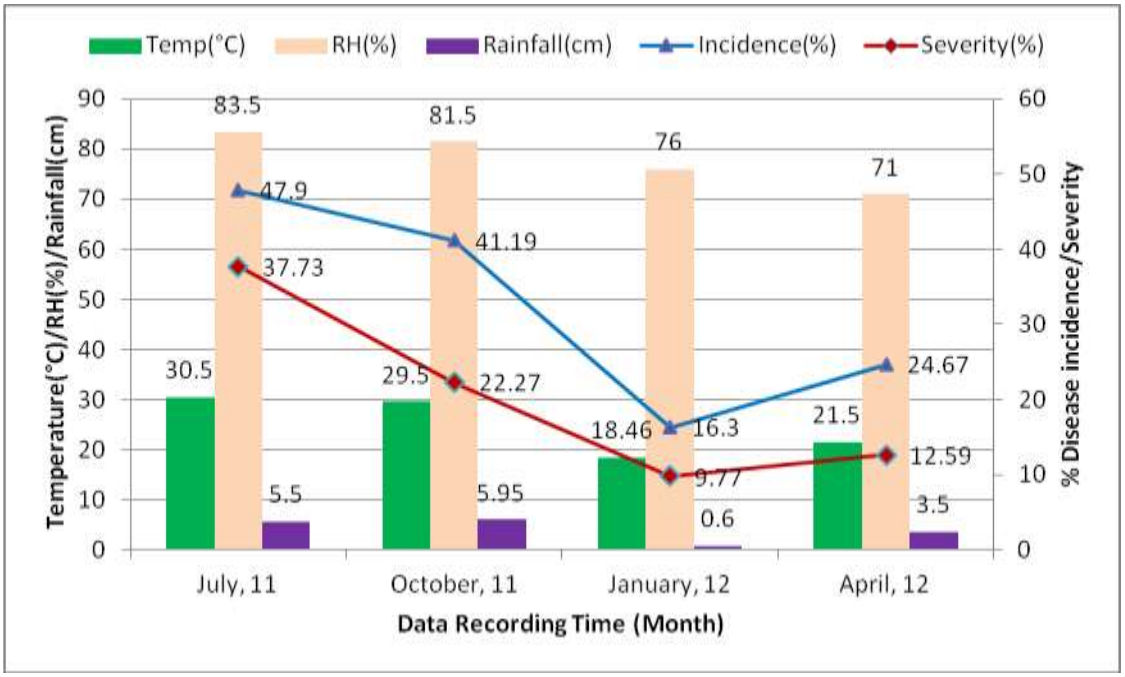

Fig. 2. Effect of different weather factors on the incidence and severity of an anthracnose of guava seedling during July, 2011 to April, 2012

Relationship between anthracnose disease incidence, severity of guava seedlings and temperature

A positive correlation between incidence and severity of anthracnose disease with temperature was observed in both the years (Figs. 3 & 4). The relationship between disease incidence and temperature could be expressed by the equation $Y=1.191x-4.270$, ($R^2=0.935$) and $Y=2.437x-28.38$, ($R^2=0.984$), where x=temperature and y=disease incidence. Here, the R^2 values indicate that the contribution of temperature to the incidence of anthracnose of guava. On the other hand, the relationship between disease severity and temperature could be expressed by the equation $Y=1.129x-9.094$, ($R^2=0.964$) and $Y=1.901x-26.91$, ($R^2=0.798$), where x=temperature and y=disease severity. Here, the R^2 values indicate that the contribution of temperature to the severity of anthracnose of guava.

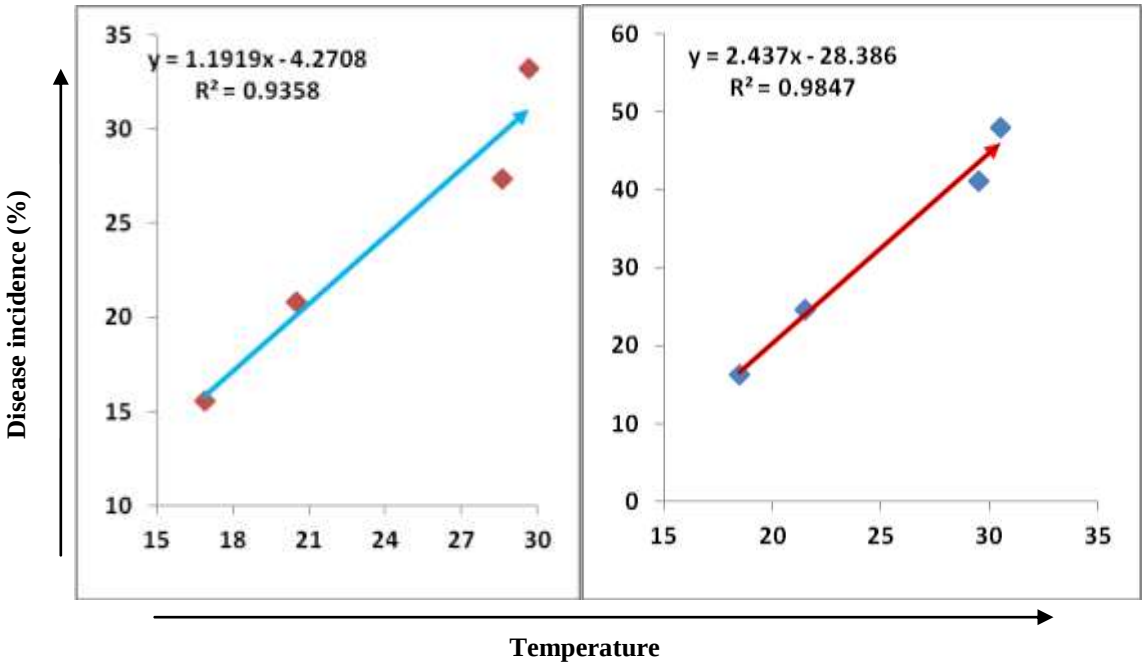

Fig. 3. Linear regression on the effect of temperature on the incidence of anthracnose of guava during July, 2010 to April, 2012

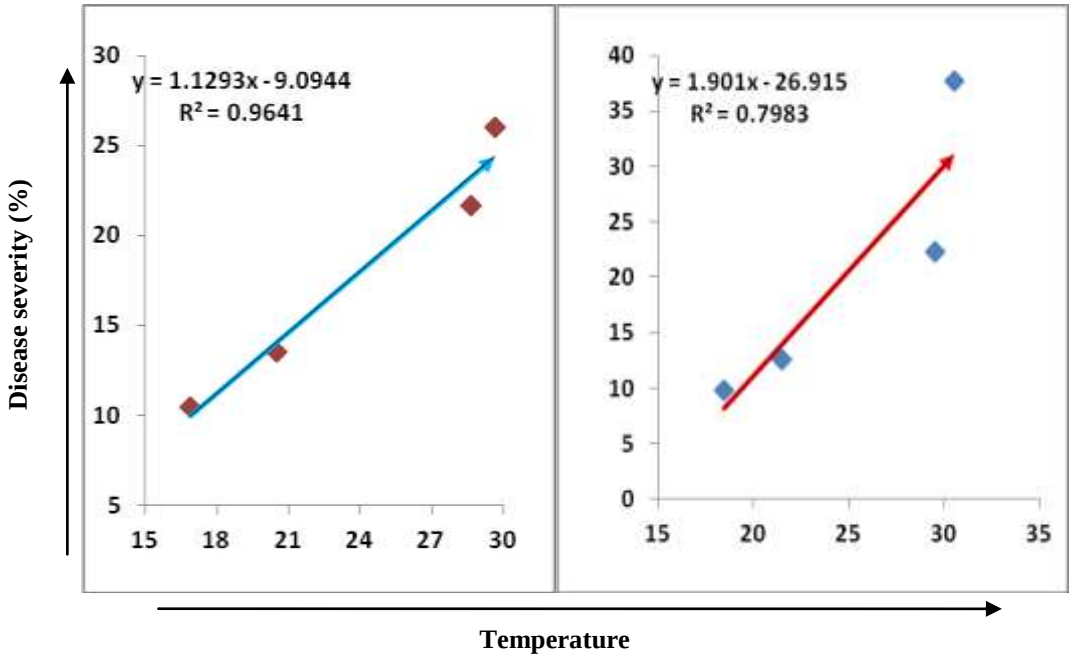

Fig. 4. Linear regression on the effect of temperature on the severity of anthracnose of guava during July, 2010 to April, 2012

Relationship between anthracnose disease incidence, severity of guava seedlings and relative humidity

A positive correlation between incidence and severity of anthracnose disease with relative humidity was observed in both the years (Figs. 5 & 6). The relationship between disease incidence and relative humidity could be expressed by the equation Y=1.211x–68.35, (R^2=0.682) and Y=2.118x–132.7, (R^2=0.673), where x=relative humidity and y=disease incidence. Here, the R^2 values indicate that the contribution of relative humidity to the incidence of anthracnose of guava. On the other hand, the relationship between disease severity and relative humidity could be expressed by the equation Y=1.219x–75.34, (R^2=0.795) and Y=1.861x–124.6, (R^2=0.692), where x=relative humidity and y=disease severity. Here, the R^2 values indicate that the contribution of relative humidity to the severity of anthracnose of guava.

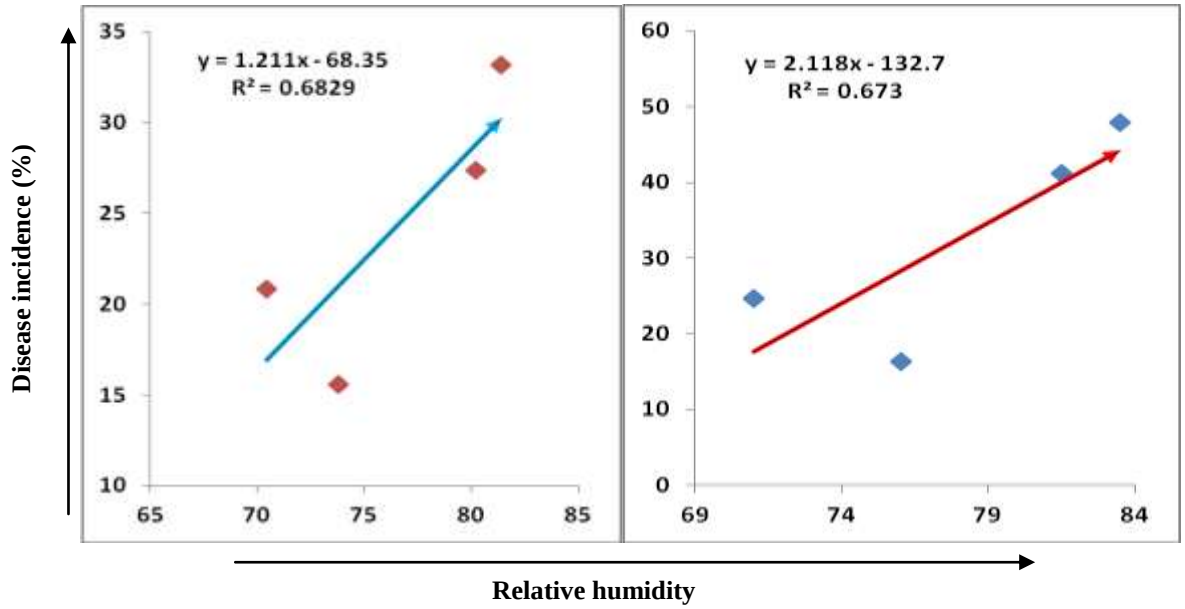

Fig. 5. Linear regression on the effect of relative humidity on incidence of anthracnose of guava during July, 2010 to April, 2012

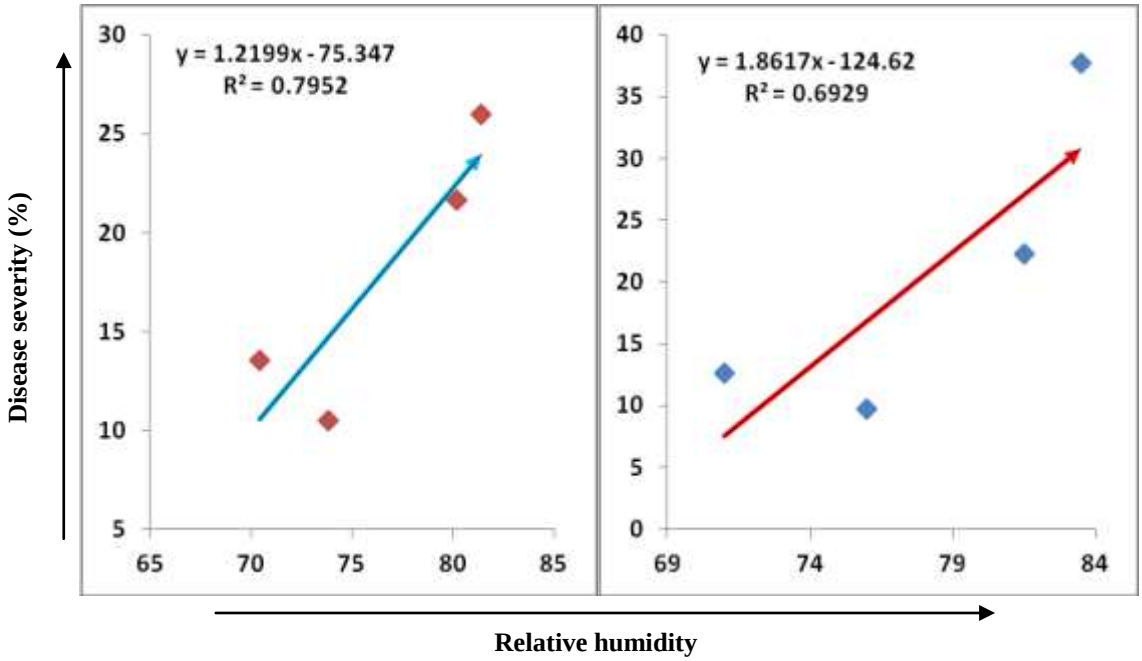

Fig. 6. Linear regression on the effect of relative humidity on severity of anthracnose of guava during July, 2010 to April, 2012

Relation between anthracnose disease incidence, severity of guava seedlings and rainfall

A positive correlation between incidence and severity of anthracnose disease with rain fall was observed in both the years (Figs. 7 & 8). The relationship between disease incidence and rainfall could be expressed by the equation Y=2.241x+13.55, (R^2=0.916) and Y=5.560x+10.89, (R^2=0.865), where x= rainfall and y=disease incidence. Here, the R^2 values indicate that the contribution of rain fall to the incidence of anthracnose of guava. On the other hand, the relationship between disease severity and rain fall could be expressed by the equation Y=2.079x+8.011, (R^2=0.905) and Y=3.917x+5.361, (R^2=0.572), where x=rainfall and y=disease severity. Here, the R^2 values indicate that the contribution of rainfall to the severity of anthracnose of guava.

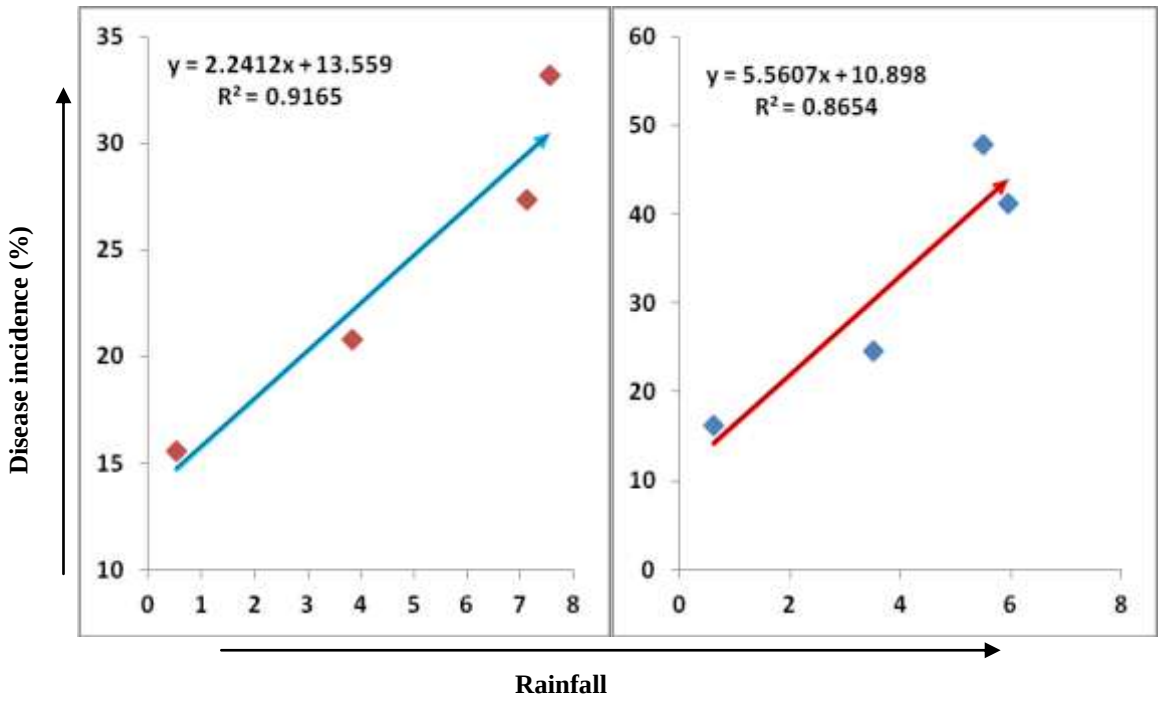

Fig. 7. Linear regression analysis of the effect of rainfall on the incidence of anthracnose of guava during July, 2010 to April, 2012

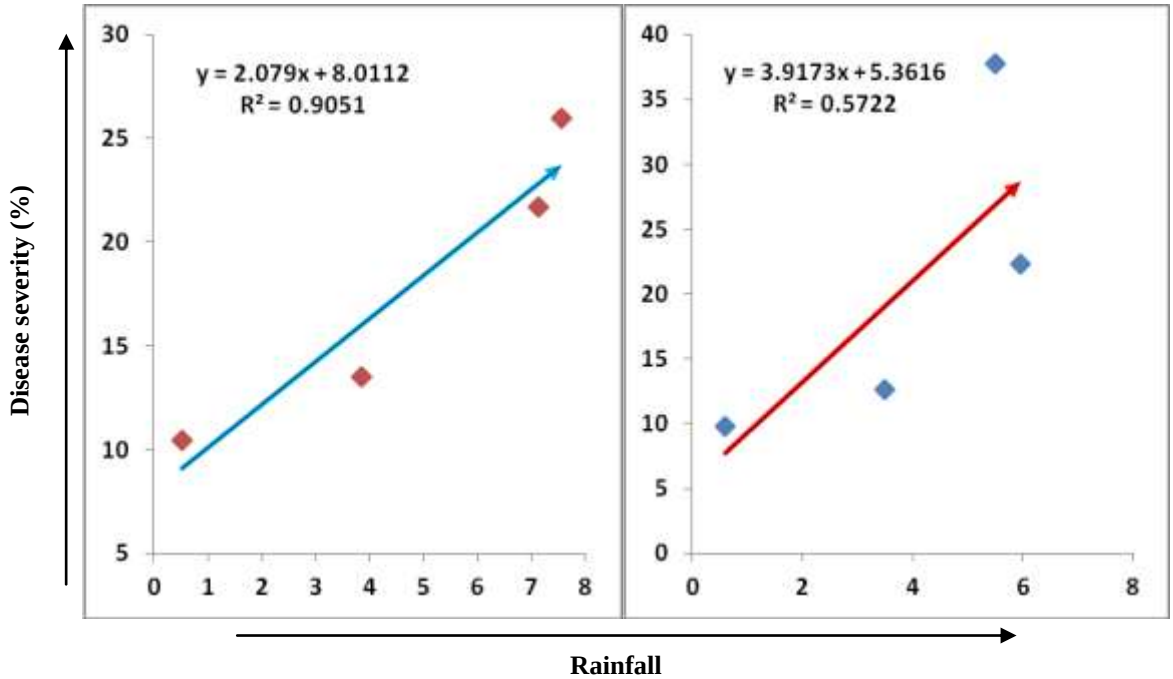

Fig. 8. Linear regression analysis of the effect of rainfall on the severity of anthracnose of guava during July, 2010 to April, 2012

DISCUSSION

Anthracnose disease was recorded as a common disease in all the growing areas. The diseases recorded in the present study have also been reported on guava seedlings from different countries of the world by (Chowdhury 2009; Chowdhury *et al.* 2011; Hossain 2011; Keith *et al*. 2006 and Marques *et al*. 2007). The disease was identified by observing the symptoms on the seedlings during survey and the presence of fungi were made either directly by preparation of slides and examining them under compound microscope or indirectly by isolation to agar culture following keys described by (Pathak 1980; Peterson 1986; Singh 1998 and Ploetz *et al.* 1998). In the investigation, *Colletotrichum gloeosporioides* was isolated from anthracnose diseased leaf. Anthracnose in guava seedling caused by *Colletotrichum gloeosporioides* were reported by researcher from many parts of the world (Gupta and Rai, 1973; Pathak 1986; Rahman *et al*. 2003 and Awasthi *et al*. 2005). Observed symptom of the present study was comparable as reported by Coutinho *et al.* (1998). However, presence of fungus, *Botryodiplodia theobromae* (Hossain and Meah, 1992 and Rahman *et al*. 2003) and *Pestalotiopsis psidii* (Hossain and Meah, 1992; Rahman *et al*. 2003; Lin *et al.* 2003; Keith *et al*. 2006), *Gloeosporium psidii* (Gupta *et al.* 1984) in anthracnose of guava fruits were also been reported.

The prevalence of the recorded disease on guava varied in respect of nursery, location and time. Similar variation in prevalence of anthracnose seedling diseases in respect of nursery location and time was recorded by Chowdhury (2009) in different guava growing areas of the country. It was also observed that the incidence and severity of anthracnose of guava varied from location to location. These variations may be due to effect of environment of different agro-ecological zone. The highest (incidence & severity) of anthracnose of guava was recorded at Khagrachari and the lowest (incidence & severity) was at Gazipur in both the years. This variation in the prevalence may be due to environmental effect of these particular agro-ecological zones.

The effects of temperature, rainfall and relative humidity on the incidence and severity of the disease of guava in selected location was observed. The climate of Bangladesh is characterized by high temperature, heavy rainfall, and often excessive humidity with fairly marked seasonal variations observed (Anonymous 1995). Determining the effects of temperature, rainfall and relative humidity on the incidence and severity of disease in different pathosystems has been focused by many researchers worldwide (Chowdhury 2009; Chowdhury *et al.* 2011; Hossain 2011; Keith *et al*. 2006 and Marques *et al*. 2007).

In the epidemiological study, the disease was recorded eight times during the period of twenty month survey from July, 2010 to April, 2012. The incidence and severity of anthracnose of guava was found to be increased in the month of July and October while the disease decreased in the month of January and April in both the years. Correlation analysis of prevalence of anthracnose disease along with generalized environmental parameters revealed that this increase and decrease were due the effect of temperature, relative humidity and rainfall. A positive correlation was observed between prevalence of anthracnose with temperature, relative humidity and rainfall. With the increase of temperature, relative humidity and rainfall both the incidence and severity increased significantly. In a similar study on anthracnose of guava seedlings, (Chowdhury 2009) recorded the

highest incidence (20.00%) and severity (16.19%) in July, 2007 at temperature, relative humidity and rainfall of 28.87°C, 85.67% and 62.60 cm, respectively while no disease was recorded in the month of January. The finding of present study is in accordance with the results of Chowdhury *et al.* 2011; Koushik *et al.* 1972 and Tandon and Singh, (1969).

CONCLUSION

The present study on the incidence and severity of seedling anthracnose disease of guava in different nurseries reveals that the diseases studied are related to the temperature, relative humidity and rainfall and have a profound effect on the disease. These weather parameters should be critically observed for each host-pathogen interaction and to find out the most appropriate time to combat the disease at minimum cost.

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