ASSESSING SEED HEALTH AND RESISTANCE TRAITS IN WATERMELON AGAINST GUMMY STEM BLIGHT

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Abstract

Water melon is a popular summer fruit, and gummy stem blight (GSB) disease, caused by the fungal pathogen *Didymella bryoniae*, is this fruit. Using resistant sources is the most effective disease management strategy. The seed health test revealed the presence of D. bryoniae and F. oxysporum f.sp. niveum fungi associated with water melon seeds imported from different counties. BARI line 03-144x21 and BARI line 02-07x08 showed the lowest lesion symptom of 25.67 x 25.17mm and 24.17 x 23.67mm in the laboratory detached leaf test in the artificially inoculated with D. bryoniae isolates S002 and S005, respectively. In pot evaluation test, all BARI lines showed lower gummy stem blight disease incidence and severity which indicated moderate resistant (MR) against the disease and all the imported hybrid varieties showed moderate susceptible to susceptible the disease. In the field experiments, four BARI inbreed lines and two commercial varieties (Black Diamond (Metal Seed) and Black Diamond (Alamgir Seed) showed resistance (MR) against the disease. Thus, it can be concluded that BARI inbred lines, viz., BARI line 01-08X07, BARI line 02-07X08, BARI line 03-144X21BARI line 04-21X144, are moderate resistant against stem blight disease in water melon with the commercial varieties and the advanced line is susceptible to stem blight disease in water melon.

Keywords: BARI inbred line, Gummy stem blight (GSB) and Resistant variety

Introduction

Watermelon holds the distinction of being the most widely consumed cucurbit worldwide, with cucumber and melon following closely behind (FAO, 2005). In Bangladesh, watermelon cultivation covers an extensive area of 12,246.59 hectares, resulting in a substantial production of 254,814.25 metric tons (BBS, 2020). This fruit crop carries significant economic importance and is valued for its abundance of lycopene, citrulline, and essential minerals and vitamins. The presence of pathogens in infected seeds can significantly diminish germination rates, weaken plant vigor, and ultimately reduce potential yields by transferring the pathogen from seed to plants. The most severe consequence of seed-borne pathogens is their ability to contaminate disease-free areas,

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serving as the primary source of inoculation for disease outbreaks. Seed infection typically occurs during three distinct phases: seed production, development, and maturation. Pathogens can play a role in each of these growth stages and can be transmitted from one crop to the next, establishing systemic infections that can colonize the seeds (McGee, 1995).

Gummy stem blight, caused by Didymella bryoniae (anamorph Phoma cucurbitacearum (Fr. Fr) Sacc.), is a widely prevalent disease affecting cucurbits globally (Sitterly and Keinath, 1996). Cucurbita spp. are particularly vulnerable to black rot, which directly diminishes both pre and post-harvest yields (Keinath et al., 1995; Zitter and Kyle, 1992). The pathogen resides both on and within the seed coat, transmitting from seed to seedling (Lee et al., 1984). Cotyledons and young leaves of watermelon and are equally susceptible to gummy stem blight. Although the leaves of young squash and cucumber plants initially display resistance, they become susceptible with age, particularly under high temperature and humid conditions (Prasad and Norton, 1967). Vansteekelenburg (1983) investigated the epidemiology of D. bryoniae and the occurrence of ascospores in glasshouses, outdoor environments, and controlled conditions. In a study conducted by Vansteekelenburg (1985), the impact of humidity on the occurrence of D. bryoniae on cucumber leaves and growing tips was examined under controlled settings. It was reported that a 10-fold increase in conidial concentrations was required to achieve the same level of infection as with leaf wetting. The disease is known to spread in greenhouses during the growing season through airborne ascospores and conidia transported by water on plant surfaces, as well as through contact between plants or between plants and humans or tools.

Effectively managing gummy stem blight (GSB) through fungicide applications and suitable cultural practices is challenging, especially during periods of rainfall when high humidity persists for prolonged durations. There's also growing concern among pathologists and breeders regarding the potential development of resistance by D. bryoniae to fungicides. This issue has been a focus of attention since the 1970s, as there's interest in exploring resistance to GSB as an alternative to chemical control. Variations in gummy stem blight (GSB) resistance have been noted among different commercial watermelon cultivars, with 'Congo' displaying the least susceptibility, 'Fairfax' showing intermediate susceptibility, and 'Charleston Gray' being the most susceptible, as reported by Schenck in 1968. Despite these efforts, no watermelon cultivars with high levels of resistance to natural GSB epidemics have been released. The rising challenges posed by GSB outbreaks in the southeastern United States prompted further studies on genetic resistance. Gusmini et al. in 2005 exploring new genetic sources of resistance to GSB. The watermelon breeding program at North Carolina State University developed efficient screening methods for testing watermelon germplasm, including systems for mass production of inoculum and disease assessment scales.

To address the challenges posed by GSB in watermelon cultivation and enhance production while boosting the income of watermelon growers, various strategies from previous literature were considered. These included implementing pathogen-free seeds and selecting resistant varieties. Given the importance of managing GSB, it is essential to observe the seed health status of the watermelon commercial varieties and to evaluate them against wilt and stem blight disease.

Materials and Methods

Evaluation seed health status of commercial varieties of watermelon against stem blight disease

Watermelon seeds of imported commercial hybrid variety (Table 1) were collected from the seed market, while BARI inbred lines were acquired from HRC, BARI, and kept at 4°C. The seeds underwent sterilization using a 2% NaOCl solution for 5 minutes, followed by rinsing with sterilized distilled water three to four times. After sterilization, the seeds were air-dried and subjected to the standard blotter method to assess mycoflora incidence and germination rates.

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Varieties / line	Туре	Colour
Black supper	Hybrid	Black
Black diamond	Hybrid	Black
Black Diamond	Hybrid	Black
Sweet dragon	Hybrid	Green with stripe
Tropical dragon	Hybrid	Green with stripe
Big family	Hybrid	Green with stripe
World queen	Hybrid	Green with stripe
Jumbo jaguar	Hybrid	Black
Black giant	Hybrid	Black
Thailand 2	Hybrid	Black
Black bull	Hybrid	Black
Dragon beauty	Hybrid	Green with stripe
Sweet green	Hybrid	Green with stripe
Sugar emperor	Hybrid	Green with stripe
BARI line 01-08x07	Inbreed	Green with stripe
BARI line 02-07x08	Inbreed	Green with stripe
BARI line 03-144x21	Inbreed	Green with stripe
BARI line 04-21x144	Inbreed	Green with stripe

 Table 1.
 Commercial hybrid varieties/inbreed lines including type and colour of watermelon seeds used in the study

Screening of watermelon commercial varieties for evaluating against D. bryoniae by detached leaves method

The study was carried out in the Plant Pathology Division laboratory at BARI following the procedure of Alam et al. 2014, 2015 and 2020. Two isolates D. bryoniae, namely S002 and S005 were employed in this investigation. Healthy, fully grown water melon leaves were used and subjected to surface sterilization using a 2% NaOCl solution for 90 seconds, followed by rinsing in sterilized water three times. After air drying on paper towels, the leaves were laid out, and wounds were created with a sterile needle before inoculation with a 4 mm mycelial plug from 7-day-old Didymella bryoniae (S002 and S005) cultures grown on PDA plates. The inoculated leaves were then placed on wet blotting paper in a 30 cm sterilized petri dish within a humid chamber. This chamber, measuring 170x240x80 mm, contained 5 ml of distilled water and was sealed with a tight lid. The setup was incubated at 25±2 °C with a 16-hour photoperiod. Disease symptoms were evaluated three days post-inoculation by measuring the length and width of lesions extending beyond the mycelial plugs. Control leaves were inoculated with sterilized water after wounding. To prevent cross-contamination, different isolates of D. bryoniae were incubated separately in their own chambers, with three replicate chambers per isolate, each containing three leaves.

Evaluation of commercial watermelon varieties against gummy stem blight under artificially inoculated condition (Pot culture experiment)

This experiment was conducted in the pot house of Plant Pathology Division, BARI, Gazipur, Bangladesh. In this experiment, S002 of *D. bryoniae* was used for evaluation of commercial verities. S002 were grown in water-soaked mixture of ground corn, wheat bran and grass pea seed coat (1:10: 5, w/w) for 10 days. Thirty-five mycelial blocks (5 mm in diameter) of PDA cultures (7-day-old) were used to inoculate 1 kg of mixture and incubated for 10 days at room temperature with a 12-h photoperiod. To confirm their pathogenicity, 15-day-old seedlings (10 seedlings/isolate, repeated three times) of watermelon were transplanted into 20-cm-diameter pots filled with soil (mixed with 10 g of inoculum/kg of soil).

On appearance of symptoms, the disease incidence was recorded by using following formula. Disease rating was made on weekly basis to find out the percentage of disease incidences among these cultivars by following the scale of Thompson and Jenkins (1985) with slight modification (Table 2).

Evaluation of commercial varieties of watermelon against stem blight diseases under natural field condition

A disease screening nursery was established to identify source of resistance against stem blight diseases of water melon in the Plant Pathology Division, BARI, Gazipur and RHRS (Regional Horticultural Research Station), BARI, Patuakhali. Collected eighteen watermelon varieties/lines were evaluated against wilt and stem blight disease under natural condition in field (Table 1). All the recommended horticultural practices were followed to maintain the crop in good condition.

Scale	Disease incidence (%)	Grading
0	0 %	Immune
1	up to 10 %	Resistant/Moderately resistant
2	11 – 25 %	Moderately Resistant
3	26-45 %	Moderately Susceptible
4	46 – 70 %	Susceptible
5	71-100%	Highly Susceptible

 Table 2. Disease rating scale (Thompson and Jenkins, 1985)

Disease incidence was recorded using the following formula:

% Disease incidence =
$$\frac{\text{Total number of infected plants}}{\text{Total of observed plants}} \times 100$$

Disease severity for each location was calculated following the equation:

% Disease severity = $\frac{\Sigma \text{ infection frequencies } \times \text{ number of leaves of each class}}{\text{Total of observed leaves } \times \text{ highest value of the evaluation scale}} \times 100$

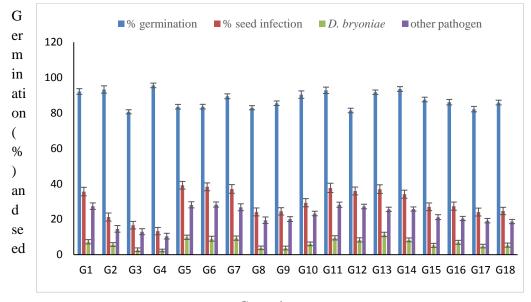
Both the categorized data and calculated disease severity % were presented.

Results and Discussion

Seed health of commercial varieties and BARI advanced lines of watermelon against *Didymella bryoniae*

To know the incidence of mycoflora, germination and seedling vigor, respectively seed samples of popular variety of watermelon listed in Table 1 was collected from seed market and Horticultural Research Center (HRC), BARI. Fungi such as Didymella bryoniae, Fusarium oxysporum f.sp. nevium (Fon) and other seed borne fungi (F. oxysporum f.sp. nevium (Fon) and Penicillium sp.) was isolated from the incubated seeds of watermelon and their cultures were maintained on Potato Dextrose Agar plates for the purpose of identification. The seed health status of fourteen commercial varieties and four BARI inbreed lines was assessed, as detailed in Fig. 1. The primary seed-borne fungal pathogens identified were D. bryoniae, F. oxysporum f.sp. nevium (Fon) and Penicillium sp. Among the evaluated varieties/genotypes, Sweet Dragon watermelon varieties exhibited the highest germination rate of 97.28%, while Dragon Beauty displayed the lowest germination rate. Regarding seed infection, the World Queen variety had the highest incidence of 47.82% (all seed borne fungi), whereas Sweet Dragon had the lowest (18.3%) (all seed borne fungi). Sweet Green had the highest Didymella infection, 14.3%, while Sweet Dragon had the lowest (3.2%). Penicillium infection was most pronounced in Black Giant (9.2%) and least in Black Diamond (2.3%).

The current investigation clearly demonstrates that *D. bryoniae* acts as seedborne pathogen, capable of transferring inoculum from the seed to the plant. The study revealed that *D. bryoniae* predominantly resides in the seed coat, a finding supported by similar observations in cucumber and pumpkin by Lee *et al.* (1984). Additionally, research conducted elsewhere indicates that in watermelon *D. bryoniae* can infiltrate the epidermis, cotyledons, and embryos (Rankin, 1954). Agarwal and Sinclair (1997) noted that seed-borne pathogens can be transmitted through infection of the embryo, endosperm, or contamination of the seed coat. During seed development and maturation, there is a significant potential for *D. bryoniae* spores to disperse, germinate, and systematically invade the seeds (Rankin, 1954).



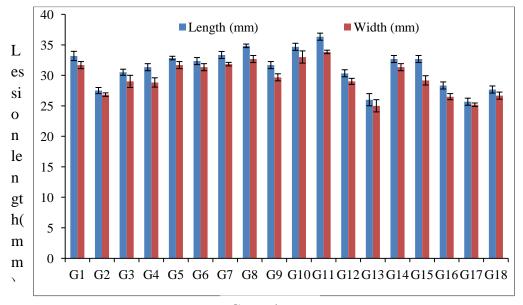
Germplasm

Fig. 1. Seed born pathogens of commercial varieties and BARI advanced lines of watermelon showing %germination, seed infection due to *D. bryonae* and other pathogen. G1=Black Super, G2=Black Diamond (Metal seed), G3=Black Diamond (Alamgir Seed),G4=Sweet Dragon, G5=Tropical Dragon, G6=Big Family, G7=World Queen,G8=Jumbo Jaguar, G9=Black Giant, G10=Thailand 2, G11=Black Bull, G12=Dragon Beauty, G13=Sweet Green, G14=Sugar Emperor, G15=BARI line 01-08x07, G16=BARI line 02-07x08, G17=BARI line 03-144x21, G18=BARI line 04-21x144

Evaluation of watermelon commercial varieties against D. bryoniae by detached leaves method in laboratory

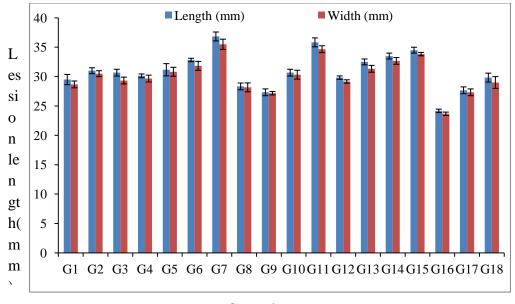
By using isolates S002 and S005 of *D. bryoniae* all varieties of watermelon developed lesion in the inoculated leaf. Susceptibility to *D. bryoniae* isolates was varied among the varieties (Fig. 2, 3 and 4). In case of S002, Black bull verity developed the highest lesion (length 36.33mm and width 33.83mm) and inbreed line BARI line 03-144x21 showed the lowest lesion development (length 25.67mm and width 25.17mm). In case of S005, world queen developed the highest (length 36.83mm and width 35.50mm) black lesion and BARI line 02-07x 08 developed the lowest lesion (length

24.17 mm and width 23.67mm (Fig. 2 and 3). Detached-leaf tests offer several benefits compared to alternative methods like multi-race or multi-pathogen testing. These advantages include avoiding issues related to systemic acquired resistance, the capacity to assess and preserve susceptible plants crucial in genetic studies, and the opportunity for enhanced replication by utilizing more leaves when evaluating individual plants of distinct genotypes. Amand and Wehner (1995) briefly noted the susceptibility of cucumber detached leaves to *D. bryoniae*. Detached-leaf tests, while convenient and exhibiting lower coefficients of variation compared to greenhouse tests, did not yield results correlated with field outcomes. Unlike greenhouse screening where supplemental nutrients were provided, detached-leaf tests didn't include them to avoid encouraging secondary organism growth. The practice of rinsing detached leaves before inoculation aimed to decrease surface organisms but might have inadvertently removed guttation exudates, potentially contributing to the disparity between field and detached-leaf infection (Amand and Wehner, 1995).



Germplasm

Fig. 2. Leaf lesion developed 72 hours after inoculation of *D. bryoniae* S002. G1=Black Super, G2=Black Diamond (Metal seed), G3=Black Diamond (Alamgir Seed),G4=Sweet Dragon, G5=Tropical Dragon, G6=Big Family, G7=World Queen,G8=Jumbo Jaguar, G9=Black Giant, G10=Thailand 2, G11=Black Bull, G12=Dragon Beauty, G13=Sweet Green, G14=Sugar Emperor, G15=BARI line 01-08x07, G16=BARI line 02-07x08, G17=BARI line 03-144x21, G18=BARI line 04-21x144



Germplasm

Fig. 3. Leaf lesion developed 72 hours after inoculation of *D. bryoniae* S002. Showing the length and width of lesion. G1=Black Super, G2=Black Diamond (Metal seed), G3=Black Diamond (Alamgir Seed), G4=Sweet Dragon, G5=Tropical Dragon, G6=Big Family, G7=World Queen,G8=Jumbo Jaguar, G9=Black Giant, G10=Thailand 2, G11=Black Bull, G12=Dragon Beauty, G13=Sweet Green, G14=Sugar Emperor, G15=BARI line 01-08x07, G16=BARI line 02-07x08, G17=BARI line 03-144x21, G18=BARI line 04-21x144

Evaluation of imported varieties and BARI inbred lines against gummy stem blight under artificially inoculation condition in pot

Under artificial inoculation condition, the incidence of stem blight in watermelon varied from 20.64% to 56.05% (Table 3). The highest incidence, 56.05%, was observed in the Black Giant (G_9) variety, which statistically similar to Jambo Jaguar (G_8), Dragon Beauty(G₁₂), Big Family (G₆), Tropical Dragon (G₅) Black Bull (G₁₁), and World Queen (G_7) varieties, all those varieties were categorized as susceptible (S). Conversely, the lowest incidence, 20.64%, was recorded in BARI line 04-21X144 (G18), which was statistically similar to BARI line 03-144X21 (24.04%), BARI line 01-08X07 (24.71%) and Black Diamond (Metal Seed) (G_2) and classified as moderately resistant (MR). The severity of stem blight followed a similar trend, with BARI line 04-21X144 (G18) exhibiting the lowest severity of 10.68%, statistically close to BARI line 03-144X21 (12.38%) and BARI line 01-08X07 (14.49%) all the imported varieties showed moderately susceptible to susceptible against stem blight disease (Table 3). Plants grown in the greenhouse and inoculated at dawn showed severe infection, whereas those inoculated in the field exhibited fewer visible symptoms of GSB. The timing of inoculation had a significant impact, with plants inoculated in the greenhouse being more susceptible compared to those inoculated in the field.





- Fig. 4. Symptom developed on upper and lower surface of different commercial varieties against isolate S002 and S005 of *D. Bryoniae* in detached leaves assay. A, upper surface of S002 inoculated leaves of G_1 - G_{18} and control; B, lower surface of S002 inoculated leaves of G_1 - G_{18} and control; C, upper surface of S005 inoculated leaves of G_1 - G_{18} and control; D, lower surface of S005 inoculated leaves of G_1 - G_{18} and control; D, lower surface of S005 inoculated leaves of G_1 - G_{18} and control; D, lower surface of S005 inoculated leaves of G_1 - G_{18} and control.
- **Table 3.** Incidence, severity and disease reduction of stem blight disease of watermelon in 18 varieties/lines at Pot house, PPD, BARI Gazipur.

Variety/Germplasm	% Stem blight incidence	% Stem blight severity	Disease reaction
G1=Black Super	37.57bc	25.66ab	MS
G2=Black Diamond (Metal Seed)	24.89de	18.30de	MS
G3=Black Diamond (Alamgir Seed)	27.20d	16.71de	MS
G4=Sweet Dragon	42.79b	22.78bc	MS
G5=Tropical Dragon	53.03a	25.81ab	S

Variety/Germplasm	% Stem blight incidence	% Stem blight severity	Disease reaction
G6=Big Family	55.01a	27.76a	S
G7=World Queen	52.23a	26.18ab	S
G8=Jumbo Jaguar	55.12a	27.38a	S
G9=Black Giant	56.05a	29.45a	S
G10=Thailand 2	46.92b	27.79a	S
G11=Black Bull	54.37a	27.85a	S
G12=Dragon Beauty	55.08a	27.15a	S
G13=Sweet Green	39.18bc	20.08cd	MS
G14=Sugar Emperor	32.40c	17.67de	MS
G15= BARI line 01-08x07	24.71de	14.49ef	MR
G16= BARI line 02-07x08	26.16d	17.10de	MR
G17= BARI line 03-144x21	24.04de	12.38f	MR
G18= BARI line 04-21x144	20.64e	10.68f	MR
CV (%)	9.96	11.82	

Evaluation commercial varieties of watermelon against stem blight disease under natural field condition

A field experiment was carried out at two locations: RHRS in Patuakhali and the Plant Pathology research field of BARI in Gazipur, to assess various commercial watermelon varieties and BARI lines against stem blight disease under natural field condition. The findings from this experiment were documented and presented through Tables 4 to 6.

First male flower opening days and numbers

In the field at RHRS in Patuakhali, it was observed that the earliest male flower opening occurred at 40 days after seed sowing in genotypes BARI line 03-144x21, Sugar Emperor, and World Queen, while the Big Family genotype exhibited male flower opening by 49 days later. The range of male flower opening among the 18 genotypes varied from 40 to 49 days (Table 4). Similarly, at BARI in Gazipur, the earliest male flower opening was recorded at 39 days in BARI line 03-144x21 followed by the varieties Black Diamond and Black Giant where male flower opened at 40 days and the Big Family variety displaying later male flower opening at 48 days (Table 6). The range of male flower opening among the 18 genotypes ranged from 40 to 48 days.

At RHRS in Patuakhali, on the first flowering, the highest number 9.67 of male flowers was recorded in the Big Family genotype while the lowest number 5.00 was observed in the genotypes Jumbo Jaguar, Black Ball, BARI line 01-08x07, and BARI line 02-07x08 (Table 6). Similarly, at BARI in Gazipur, the highest number of male flowers

on the first day of observation was noted in the Big Family genotype at 8.67, while the lowest number of 4.33 was observed in the genotypes Black bull, BARI line 01-08x07 and BARI line 02-07x08 (Table 6).

First female flower opening days and numbers

At RHRS in Patuakhali, the World Queen genotype showed the earliest female flower opening on the 42nd day of seed sowing, while the Big Family genotype exhibited later openings on the day 49. On the first day of female flower emergence, Black Diamond (Alamgir Seed), Tropical Dragon, and BARI line 03-144x21 displayed higher number of flowers at 17.33 whereas Black Diamond, Sweet Dragon, Sweet Green, and BARI line 02-07x08 had a lower count of 13.33 (Table 4). At BARI in Gazipur, the World Queen genotype exhibited the earliest opening of female flowers on the 41st day, while the Big Family genotype showed later openings on day 48. On the initial day of female flower emergence, Tropical Dragon displayed a higher count of 17.00, while Black Diamond, Sweet Dragon, and Sweet Green varieties had a lower count of 12.00 (Table 6).

Number of fruit per plant (NFPP)

In the field at RHRS in Patuakhali, the highest number of fruits per plant was observed in BARI line 01-08x07 at 3.95 followed by Black Bull, Black Super, World Queen, Sugar Emperor and BARI line 04-21x144 where the fruit number per plant was 3.10, 3.06, 3.05, 3.03 and 2.97, respectively. and the lowest number was recorded in the Big Family variety of 1.89 (Table 4).

Individual fruit weight (IFW)

The Thailand-2 genotype had the highest individual fruit weight (6.63 kg), while the lowest weight of 3.03 kg was observed in the BARI line 01-08x07 (Table 4).

Fruit length (cm) and fruit breadth (cm)

Thailand 2, Sweet Green and exhibited higher fruit length at 31.20 and 30.74 cm, respectively followed by Black Giant with fruit length 29.65 cm. while BARI line 04-21x144 displayed the shortest length at 17.15 cm (Table 4). Thailand 2 and Super Emperor showed the widest fruit breadth of 25.22 and 25.58 cm, respectively, whereas BARI line 01-08x07, BARI line 02-08x07, BARI line 03-144x21 and BARI line 04-21x144 had the narrowest breadth (Table 4).

Fruit yield (t/ha)

At RHRS in Patuakhali, Fruit yield ranged from 21.16 to 65.13 t/ha. The variety Sugar Emperor exhibited the highest fruit yield 65.13 t/ha followed by Black Bull, Dragon Beauty and Sweet Green where the yield was 53.45, 50.82 and 50.60 t/ha,

respectively. The lowest yield was recorded from Big Family variety of 21.16 t/ha (Table 4).

Field Evaluation of different cultivar/ varieties/lines against the stem blight disease of watermelon

Two field experiments were conducted at RHRS, Patuakhali and Plant Pathology Division, BARI, Gazipur. At RHRS, Patuakhali, results revealed that, various varieties, or lines exhibited significant differences in both the incidence and severity of stem blight diseases (Table 5). Stem blight disease incidence ranged from 13.5% to 47.5%. Black Giant (G9) showed the highest disease incidence at 47.5%, which was statistically comparable to Jambo Jaguar (46.4%), Big family (46.2%), Black Bull (44.9%), and Tropical Dragon (44.2%). Conversely, the lowest disease incidence was noted in BARI line 04-21X144 at 13.5%, which was statistically similar to BARI line 03-144X21 (15.0%), Black Diamond (16%), and BARI line 01-08X07 (16.3%). Stem blight severity in field observations ranged from 10.3% to 26.6%. The highest disease severity was observed in Jambo Jaguar (G8), statistically a kin to Black Giant (G9), Big family (G6), Dragon Beauty (G16), Tropical Dragon (G5) watermelon varieties (Table 5).

At Plant Pathology Division, BARI, Gazipur, stem blight incidence ranged from 10.4% to 45.9% ((Table 6). Black Giant (G9) variety exhibited the highest disease incidence of 45.9%, which was statistically similar to Jambo Jaguar (45.2%), Big Family (45.3%), Black Bull (43.7%), and Tropical Dragon (43.2%). The lowest incidence was observed in BARI line 04-21X144 at 10.4%, followed by BARI line 03-144X21 (12.9%) and Black Diamond (14.9%). At Plant Pathology Division, BARI, stem blight disease severity ranged from 9.2% to 25.6%. Jambo Jaguar (T8) had the highest disease severity followed by Black Giant (G9), Big Family (G6), Dragon Beauty (G12), Tropical Dragon (G5), among other watermelon varieties. The lowest disease severity was recorded in BARI line 04-21X144, similar to BARI line 03-144X21 (9.9%) and BARI line 01-08X07 (11.69%) (Table 6). Based on these findings, it was concluded that BARI lines demonstrated moderate resistance against stem blight disease under both artificial conditions and inoculum pressure. However, hybrid varieties from abroad displayed susceptibility to the disease.

The assessment of GSB resistance shows variability ranging from 11-70% (2 to 4 rating units) among different plants and replications (Wehner and Amand, 1993). They observed variability in GSB outbreaks may stem due to genetic or environmental factors (Wehner and Amand, 1993), which can influence pathogen aggressiveness, leading to variations across years and between field and greenhouse tests (Gusmini and Wehner, 2002; Stewart *et al.*, 2015). Recent findings indicate that genetically distinct fungal species serve as causal agents of GSB, suggesting that resistance variability across different years and environments, such as greenhouses and fields, may arise from interactions between the environment and fungal species (Stewart *et al.*, 2015). The

inconsistency in observed ratios may be due to interactions among multiple genetic loci, as well as interactions with environmental factors (Kumar, 2009). In the controlled environment of the greenhouse, conditions were standardized for both plant and pathogen development. However, in the field, where a large number of cultivars were evaluated, the need for extensive space led to increased environmental variability across different sites and years.

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Variety/Germplas m	1stMF O	NO1stM F	1stFF O	NO1stF F	NFPP	IFW(k g)	FL(C M)	FB (CM)	YT H
G1=Black Super	44	7.67 ^{bc}	46	15.3 ^{bc}	3.06 ^b	4.35 ^{ef}	27.4 ^{bc}	19.17 ^c	
G2=Black Diamond(Metal Seed)	41	5.67 ^{d-e}	44	13.0 ^d	2.45 ^{ef}	4.08 ^f	28.3 ^{bc}	21.95 de	•
G3=Black Diamond (Alamgir Seed)	42	6.00 ^{c-e}	44	17.3 ^a	2.74 ^{b-} d	4.94 ^{b-e}	26.8 ^{cd}	21.91 de	43.8 0
G4=Sweet Dragon	43	8.00 ^{ab}	46	13.0 ^d	2.93 ^{bc}	4.74 ^{c-e}	19.2 ^d	20.95 ^c	50.4 9
G5=Tropical Dragon	47	7.67 ^{bc}	48	17.3 ^a	2.79 ^{b-} d	4.64 ^{d-f}	27.8 ^{bc}	22.36 ^c	40.2 1
G6=Big Family	49	9.67 ^a	49	14.0 ^{bc}	2.13 ^{ef}	4.74 ^{c-e}	26.1 ^d	20.75 de	21.1 6
G7=World Queen	40	7.00 ^{b-d}	42	16.3 ^a	3.05 ^b	4.86 ^{b-e}	28.3 ^{bc}	20.55 de	40.9 1
G8=Jumbo Jaguar	44	5.00 ^e	46	13.3 ^d	2.60 ^{b-} e	4.91 ^{b-e}	30.4 ^a	20.68 de	49.2 8
G9=Black Giant	41	6.67 ^{b-e}	44	16.3ª	2.37 ^{df}	4.59 ^{d-f}	29.7 ^{ab}	21.96 de	32.6 9
G10=Thailand 2	45	6.00 ^{c-e}	47	16.3 ^a	2.10 ^{ef}	6.63 ^a	31.2 ^a	25.22ª	43.7 0
G11=Black Bull	44	5.00 ^e	45	14.0 ^{bc}	3.10 ^b	4.99 ^{b-e}	27.3 ^{cd}	22.55 bc	53.4 5
G12=Dragon Beauty	40	6.00 ^{c-e}	43	17.0 ^a	2.76 ^{b-} d	5.07 ^{b-d}	28.0 ^{bc}	22.25 ^c	50.8 2
G13=Sweet Green	45	6.67 ^{b-e}	46	13.0 ^d	2.80 ^b	5.31 ^{b-d}	30.7 ^a	23.69	50.6 0

 Table 4.
 Performance of water melon 18 varieties/lines at RHRC research field, Patuakhali.

Variety/Germplas	1stMF	NO1stM	1stFF	NO1stF	NEDD	IFW(k	FL(C	FB	YT
m	0	F	0	F	NFPP	g)	M)	(CM)	Η
G14=Sugar Emperor	40	5.67 ^{de}	46	17.0 ^a	3.03 ^b	5.49 ^b	28.2 ^{bc}	25.58 ^a	65.1 3
G15=BARI line 01-08x07	42	5.00 ^e	45	14.0 ^{bc}	3.95 ^a	3.03 ^g	19.4 ^e	16.19 ^e	37.3 9
G16=BARI line 02-07x08	44	5.00 ^e	45	13.0 ^a	2.07 ^f	3.20 ^g	18.9 ^e	18.35 ^e	22.4 0
G17=BARI line 03-144x21	40	6.67 ^{b-e}	43	17.3 ^a	2.83 ^{b-} d	4.48 ^g	19.1 ^e		38.1 2
G18=BARI line 04-21x144	45	7.00 ^{b-d}	46	15.33 ^{ab}	2.97 ^b	4.02 ^{d-f}	17.2 ^f	16.81 ^e	34.5 4
CV (%)	-	5.32	-	6.26	3.37	3.46	7.54	8.37	7.77

1stMFO= First male flower opening, NO1stMF= No of First Male Flower, 1stMFO= First female flower opening, NO1stFF= No of First Female Flower, IFW= Individual Fruit Weight, NFPP= No of fruit per plant, FL(cm.) = Fruit length (cm.), FB(cm.) = Fruit breath (cm.) and YTH= Yield (ton/ha)

Table 5. Incidence and severity of stem blight disease of water melon in 18 varieties/lines at RHRC research field, Patuakhali.

Variety/Germplasm	Stem blight incidence (%)	Stem blight severity (%)	Disease reaction
G1=Black Super	27.8e	20.39cd	MS
G2=Black Diamond(Metal Seed)	16fg	16.38fg	MR
G3=Black Diamond (Alamgir Seed)	18.2f	14.32fg	MR
G4=Sweet Dragon	34.0d	19.95de	MS
G5=Tropical Dragon	44.2ab	24.07ab	MS
G6=Big Family	46.2ab	24.93ab	S
G7=World Queen	43.3bc	22.70bc	MS
G8=Jumbo Jaguar	46.4ab	26.57a	S
G9=Black Giant	47.5a	26.41a	S
G10=Thailand 2	40.4c	21.24cd	MS
G11=Black Bull	44.9ab	23.04bc	MS
G12=Dragon Beauty	46.4ab	24.41ab	S
G13=Sweet Green	34.3d	17.34ef	MS
G14=Sugar Emperor	27.0e	16.55fg	MS

Variety/Germplasm	Stem blight incidence (%)	Stem blight severity (%)	Disease reaction
G15= BARI line 01-08x07	16.3fg	11.80hi	MR
G16= BARI line 02-07x08	18.3f	14.74fg	MR
G17= BARI line 03-144x21	15.0fg	10.91i	MR
G18= BARI line 04-21x144	13.5g	10.27i	MR
CV (%)	6.47	8.42	

MR= moderately resistant, S= Susceptible, MS= moderately susceptible

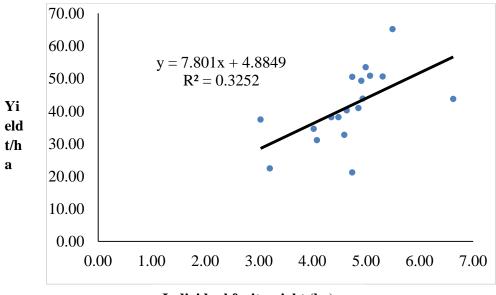
Table 6.	Incidence	and	severity	of	stem	blight	disease	of	water	melon	in	18
	varieties/li	nes at	BARI res	sear	ch field	d Gazipi	ur.					

varieties/ intes a				pur.			
Variety/Germplasm	1stMFO	NO1stMF	1stFFO	NO1stFF	% Stem blight incidence	% Stem blight severity	Disease reaction
G1=Black Super	43	7.00bc	45	13.7cd	27.1f	20.5с-е	MS
G2=Black Diamond(Metal Seed)	40	5.00d-e	43	12.0e	15.0hi	14.8gh	MR
G3=Black Diamond (Alamgir Seed)	41	5.33d-e	43	16.3ab	17.2h	13.3hi	MR
G4=Sweet Dragon	42	7.33bc	45	12.0e	33.1e	19.0ef	MS
G5=Tropical Dragon	46	7.00bc	47	17.0a	43.2а-с	23.2а-с	MS
G6=Big Family	48	8.67a	48	13.0de	45.3ab	23.5ab	MS
G7=World Queen	41	6.33b-e	41	15.7b	42.3c	21.8b-d	MS
G8=Jumbo Jaguar	45	5.00de	45	13.3de	45.2ab	25.6a	S
G9=Black Giant	40	6.00b-e	43	16.0ab	45.9a	24.8a	S
G10=Thailand 2	44	5.33de	46	15.7b	37.2d	20.0de	MS
G11=Black Bull	45	4.33e	44	13.0de	43.7а-с	23.8ab	MS
G12=Dragon Beauty	42	5.33de	42	16.0ab	45.3ab	24.9a	S
G13=Sweet Green	44	6.00b-e	45	12.0e	32.4e	16.9fg	MS
G14=Sugar Emperor	41	5.00de	45	16.0ab	22.8g	15.4gh	MR
G15=BARI line 01-08x07	41	4.33e	44	13.7cd	14.7hi	11.7ij	MR
G16=BARI line 02-07x08	43	4.33e	44	12.7de	16.6h	14.9gh	MR
G17=BARI line 03-144x21	39	6.00b-e	42	16.3ab	10.0j	MR	
G18=BARI line 04-21x144	44	6.33b-e	45	14.3c	10.4j	9.1j	MR
CV (%)	-	6.67	-	5.43	4.97	8.79	

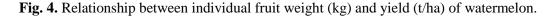
1stMFO= First male flower opening, NO1stMF= No of First Male Flower, 1stMFO= First female flower opening, NO1stFF= No of First Female Flower

Relationship between individual fruit weight (kg) and yield (tha-1) of watermelon

Fig. 4 indicates a linear correlation between individual watermelon fruit weight (kg) and their yield per hectare (measured in t). This suggests a positive relationship between fruits weight and their yield. A regression line was devised to represent this relationship as y = 7.801x + 4.8849, where 'y' stands for yield (tons/ha) and 'x' represents individual fruit weight (kg). The coefficient of determination, $R^2 = 0.3252$, implies that 32.52% of the variation in watermelon yield per hectare can be explained by differences in individual fruit weight.



Individual fruit weight (kg)



Water melon seeds may be contaminated with seed borne fungi like *Didymella bryoniae*, *Fusarium oxysporum* f.sp. *niveum* and other seed borne fungi. BARI lines 03-144x21, and 02-07x08 showed the lowest gummy stem blight disease symptom development in the laboratory detached leaf test and the lowest disease incidence and disease severity under an artificially inoculated condition in a pot house. All the commercial varieties and the advanced line have susceptibility to the gummy stem blight disease of water melon. From these studies, it can be concluded that the BARI lines, viz., BARI line 01-08X07, BARI line 02-07X08, BARI line 03-144X2 and BARI line 04-21X144, showed moderately resistance against stem blight disease of water melon and all the commercial varieties and advanced lines showed susceptibility of GBD of water melon.

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Conflicts of Interest

The authors declare no conflicts of interest regarding publication of this manuscript.

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