



INVESTIGATION OF MAIZE DISEASES IN DINAJPUR AND NILPHAMARI DISTRICTS OF BANGLADESH

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ABSTRACT

A field survey and laboratory analyses were conducted to assess the status of maize diseases in two major maize-growing districts of Bangladesh, namely Dinajpur and Nilphamari. Five farmers from each Upazila in both districts were interviewed using pretested questionnaires to collect data. Diseased specimens and farmer-saved seeds were also collected to identify the associated pathogens. Based on the observations during the survey and opinions expressed by the farmers, Turcicum leaf blight (2.93%) was identified as the major disease, followed by sheath blight (0.74%), cob blight (0.54%), stalk rot (0.28%), brown spot (0.24%), cob rot (0.22%), southern leaf blight (0.13%), Curvularia leaf spot (0.13%), Alternaria leaf blight (0.03%), bacterial wilt (0.01%),

ear rot (0.01%), rust (0.01%), maize streak (0.01%), and bushy stunt (0.01%). Microscopic observations and pathogenicity tests confirmed the involvement of *Bipolaris maydis* and *Curvularia lunata* in two of the observed diseases. Seed health tests revealed the presence of various seed-borne pathogens, including *Fusarium moniliforme* (39.88%), *Aspergillus niger* (18.50%), *Aspergillus flavus* (21.81%), *Penicillium* sp. (10.25%), *Erwinia* sp. (4.38%), *Curvularia lunata* (0.44%), *Exserohilum turcicum* (0.13%), *Bipolaris maydis* (7.25%), *Chaetomium* sp. (0.19%), and *Alternaria alternata* (0.12%). The findings of this study highlight the significance of maize diseases as a major threat to maize production in Bangladesh.

Keywords: Survey, Diseases, Maize, Dinajpur, Nilphamari

INTRODUCTION

Maize (*Zea mays* L.) belongs to the family Gramineae and is a photo-insensitive versatile crop cultivated worldwide as a staple food and feed (Rahman *et al.* 2016, Alam *et al.* 2014, Anderson *et al.* 2004). It is a rich source of starch (72%), protein (10%), and fat (4%) and is also used for the production of biofuel (Deepavali and Nilima 2013, Islam and Hoshain 2022). Because of the low production cost, diversified use, and flexibility in the genetic makeup, maize has been considered as the most promising cereal crop since the last decade. However, the total acreage and production of maize was 1182000 acres and 4262000 metric tons

and ranked third position among all the grain crops in Bangladesh (BBS 2022). Though the acreage and production of maize are increasing in Bangladesh, still the production is not able to meet the domestic demand. Hence, each year Bangladesh imports a huge amount of maize or maize produce from abroad.

The production of maize is constrained by several abiotic and biotic factors such as mycoplasma, nematode, fungi, and bacteria (Keno *et al.* 2018, El-Sappah *et al.* 2022, Jugenheimer 1976). Around 30 diseases of maize including seed-borne were reported in Bangladesh which could be a mammoth threat to domestic agriculture as well (Azad *et al.* 2020,

Khaiyam *et al.* 2017, Erasto *et al.* 2023, Fakir 2001, Sreenu *et al.* 2019). Since disease incidence and severity directly affect the potential yield of maize, hence, it is necessary to know the status of maize diseases and their causes in the major maize-growing areas (Nwanosike *et al.* 2015; Suleiman *et al.* 2024). To know the current disease status, so far, little or no work has been conducted in the major maize- growing region of Bangladesh. we aimed to conduct this study to determine the disease status of maize in the two major maize-growing areas in the northern part of Bangladesh, specially in the Dinajpur and Nilphamari Districts.

MATERIALS AND METHODS

Survey and collection of diseased samples

Maize fields were randomly selected in different Upazilla belongs to Dinajpur (Dinajpur sadar, Phulbari, Kaharole, Birganj, Bochaganj, Biral, Khansama, Parbatipur, Nawabganj, Ghoraghat, Birampur, Chirirbandar, Hakimpur) and Nilphamari (Nilphamari Sadar, Saidpur, Dimla, Domar, Jaldhaka, Kishoreganj) Districts. Data on various maize diseases were collected based on farmer's responses and field observation. In each Upazilla, 5 farmer's fields were selected randomly where a W-pattern was marked to collect data on disease incidence (%) and severity (%) from five different spots. A total of 95 farmer's fields were surveyed starting from the silking to the milking stages during the Kharif season. Diseases were identified following "Maize Disease: A Guide for Field Identification" (CIMMYT 2004). During the survey, maize-diseased samples were also collected from the farmer's fields and carried to the laboratory for further study.

Calculation of disease incidence (%)

Twenty plants from the selected spots were randomly selected and the number of infected plants was recorded and expressed as a percent plant population and calculated following the formula (Alemu *et al.* 2016).

$$\text{Disease incidence (\%)} = \frac{\text{Number of diseased plants observed}}{\text{Total number of plants examined}} \times 100$$

Observation of associated pathogens

Infected parts of the plant were collected and washed thoroughly under running tap water followed by surface sterilization with 10% Clorox for 10 minutes and three times repeated washing with sterilized distilled water. The sterilized plant parts were then cut into 1.0 cm pieces with the help of sterile scissors and placed in a Petri dish containing three layers of moistened blotter papers and incubated for 5-7 days at room temperature. Pathogens grown on leaf tissues were observed under a stereo microscope (Olympus SZX2-ILLT). A microscopic slide was also prepared to observe

various pathogenic structures including conidia, fruiting bodies, ooze, etc. under a compound microscope (Olympus CX21) following a Laboratory Guide of CIMMYT. The isolated fungi and bacteria were also cultured onto PDA and NA media, respectively and stored at 4 °C until further use.

Study of pathogenicity

Pathogenicity assays were conducted by using the locally available susceptible maize variety. The collected seeds were sown in sterilized earthen pots containing sterilized soil and compost (3:1). The raised seedlings (45 days old) were inoculated by spraying with *Bipolaris maydis* and *Curvularia lunata* (10^4 - 10^6 spores mL⁻¹) and covered by a polythene bags for forty-eight hours to maintain humidity. Then the pots were transferred to the shed and disease severity (%) was recorded from 12 randomly selected plants. Disease severity (%) was recorded at 15 to 20 days after inoculation following a 1 to 5 scale (Payak and Sharma 1983). The percent disease index (PDI) was calculated by the formula of Wheeler (1969):

$$\text{PDI} = \frac{\text{Sum of numerical rating}}{\text{No of leaves examined} \times \text{maximum grade value}} \times 100$$

Observation of seed-borne pathogens

Maize seeds were collected from local farmers, sterilized, and incubated following the blotter method (400 seeds) (Anon. 2014). In brief, three layers of moistened sterile blotting paper were placed in sterilized Petri dishes (90 mm) where 10 seeds were placed equidistantly and incubated at 28±2 °C with alternating cycles of 12 h near- ultraviolet light and darkness. Following incubation for 7 days, the seeds were observed under a stereomicroscope to observe the associated pathogenic structures (conidia, conidiophores, mycelium, and ooze) under a compound microscope.

Analysis of data

All the collected data on disease incidence, severity, and associated pathogens were subjected to analysis using SPSS version 22 (Statistical Package of Social Science) software.

RESULTS AND DISCUSSIONS

Recording of diseases at Nilphamari district

Nilphamari district comprises six Upazillas where except Saidpur, Turicum leaf blight (~3.40%) was found as the major disease in all the Upazillas. Only Cob blight (2.40%) was found as the major disease at Saidpur Upazilla. The other major diseases like Sheath blight (~1.24%), Stalk rot (0.82%), Bushy stunt (0.02%), Rust (0.20%), Maize streak (0.04%), Ear rot (0.02%), etc. were reported in the surveyed area of Nilphamari District (Figure 1) and in addition to Nilphamari districts, Bogura, Dhaka, Chuadanga, and Lalmonirhat also produced a significant amount of

maize (BBS, 2022). Because of the increased cultivation of maize, diseases are also becoming a major concern. However, *Exserohilum turcicum*, *Puccinia sorghi*, and *Cercospora zea-maydis* along with the diseases namely turcicum leaf blight, gray leaf spot, and common leaf rust were also reported as the major threat to maize cultivation (Sintayehu 2018).

At Dinajpur District

All the Upazilla (except Parbatipur) under the District Dinajpur demonstrated Turcicum leaf blight as the major disease (~3.68%). Only Parbatipur showed Sheath blight (~2.00%) as the major maize disease. In the surveyed area, Cob blight (~0.80%), Stalk rot (~0.74%), Brown spot (~0.74%), Curvularia leaf spot (~0.60%), southern leaf blight (~0.60%), Alternaria leaf spot (~0.20%) (~0.02%), Bushy stunt (~0.02%), Maize streak (~0.02%) were also recorded (Figure 2).

Similar to the present findings, a survey of 20 maize-growing districts in Bangladesh reported 21 diseases of maize including leaf spot, stalk rot, cob rot, root rot, sheath blight, cob sheath blight, sheath rot, cob sheath rot, bacterial leaf blight, leaf blight, maize dwarf mosaic virus, leaf virus, corn stunt, and maize streak virus (Khaiyam, *et al.* 2017).

Recorded diseases in the surveyed area

According to farmers' opinions and observation, a total of fourteen (14) diseases were recorded in all the studied areas where Turcicum leaf blight (2.93%) was found as the major disease followed by Sheath blight (0.74%) and Cob blight (0.54%) (Figure 3). The minor diseases were recorded as Stalk rot (0.28%), Brown spot (0.24%), Cob rot (0.22%), Southern leaf blight (0.13%), Curvularia leaf spot (0.13%), Alternaria leaf

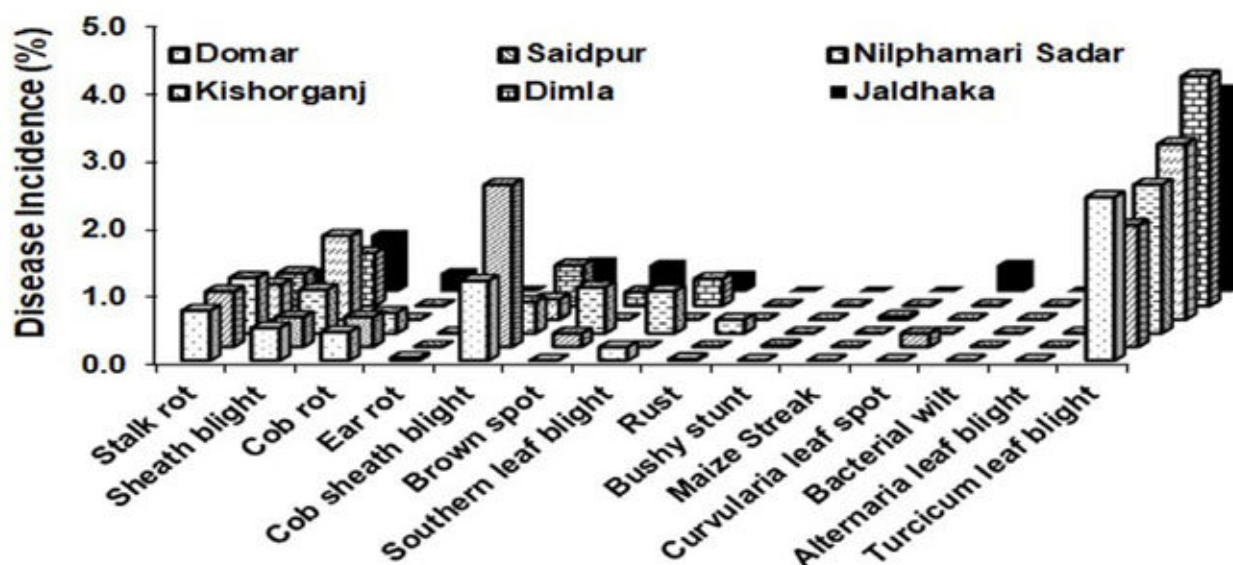


Figure 1. Disease incidence (%) of different maize diseases in different locations of Nilphamari District. Cob rot (~0.20%), Ear rot (~0.04%), Rust (~0.02%)

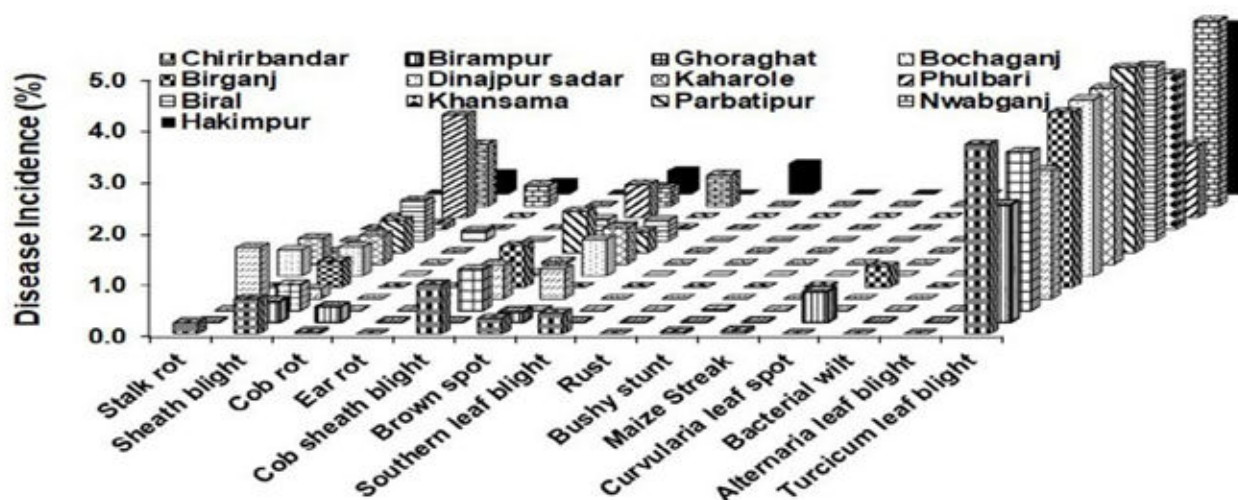


Figure 2. Disease incidence (%) of different maize diseases in different locations of Dinajpur District.

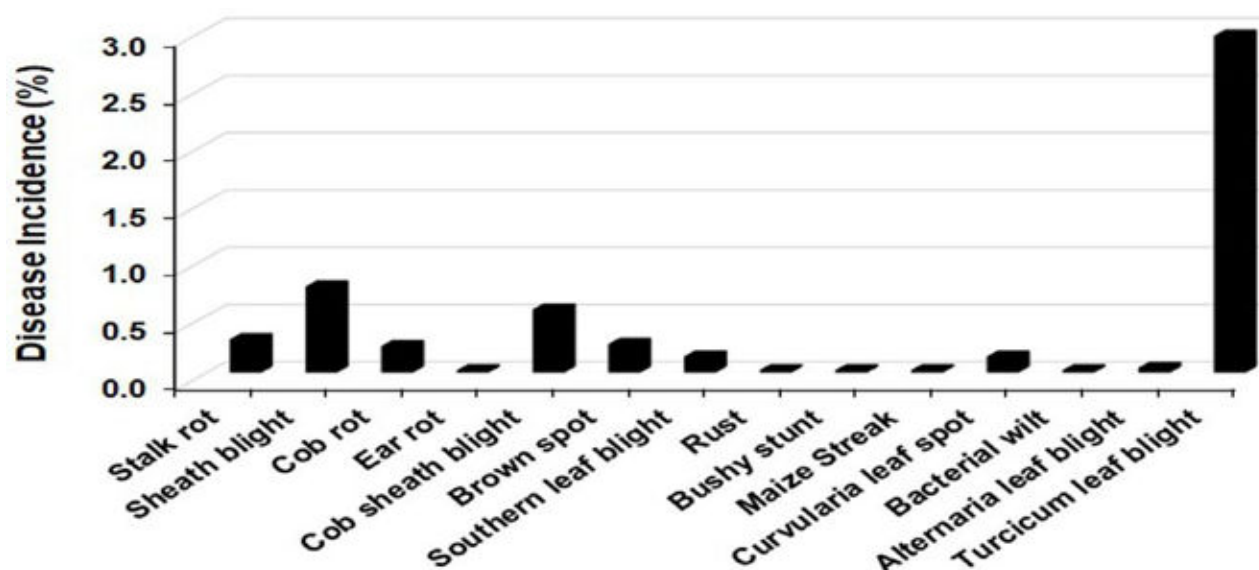


Figure 3. Prevalence of different diseases of maize in the studied area.

blight (0.03%), Bacterial wilt (0.01%), Ear rot (0.01%), Rust (0.01%), Maize streak (0.01%) and Bushy stunt (0.01%) (Figure 3). Among all the surveyed Upazilla in the two districts, Chiriribondor Upazilla was found as the major Turicum leaf blight (3.68%) infected area whereas, Turicum leaf blight less prevailed at Parbotipur Upazilla (1.40%) (Figure 3). Dinajpur and Nilphamari are the two major maize-producing regions where, a total of 14 diseases were recorded including Turicum leaf blight, Sheath blight, Cob blight, Stalk rot, Brown spot, Cob rot, Southern leaf blight, Curvularia leaf spot, Alternaria leaf blight, Bacterial wilt, Ear rot, Rust, Maize streak, and Bushy stunt. A very primitive work (Talukdar 1974) reported nine diseases of maize including Leaf blight, Cob blight, Kernel mold, smut, Leaf spot, Brown spot, Bacterial streak, soft rot, and Mosaic. Later, around 30 diseases have been reported in Bangladesh (Alam *et al.* 2014, Azad *et al.* 2020, Khaiyam *et al.* 2017, Ali and Alam 2003, Yasmin 2007).

Morphological identification of the pathogen associated with the identified diseases

Among the identified diseases, the morphological and microscopic study revealed the association of *Exserohilum turcicum*, *Bipolaris maydis*, *Alternaria alternata*, and *Curvularia lunata* with the major diseases of maize viz. Turicum leaf blight, Southern leaf blight, Alternaria leaf blight, and Curvularia leaf spot, respectively. Pathogenicity test of *Bipolaris maydis* and *Curvularia lunata* were confirmed by the development of characteristic symptoms where, *Bipolaris maydis* and *Curvularia lunata* demonstrated 42.11 and 26.32% disease severity, respectively (Figure 4). The weather conditions including temperature and precipitation, agro-ecological zones, etc. might play the determining role in the variation in the occurrence of different diseases of maize in different areas (Keno *et*

al. 2018, El-Sappah *et al.* 2022, Mehrotra and Aggarwal 2006). Among the weather factors, temperature plays the most significant role in the infection and the rate of penetration of the pathogen into the host. The latent periods of pathogens tend to decrease with increasing temperature (Keno *et al.* 2018, El-Sappah *et al.* 2022, Mehrotra and Aggarwal 2006).

Pathogens associated with farmers' saved seeds

A total of ten (10) seed-borne pathogens namely *Fusarium moniliforme* (39.88%), *Aspergillus flavus* (21.81%), *Aspergillus niger* (18.50%), *Penicillium* sp. (10.25%), *Bipolaris maydis* (7.25%), *Erwinia* spp. (4.38%), *Curvularia lunata* (0.44%), *Chaetomium* sp. (0.19%), *Exserohilum turcicum* (0.13%), and *Alternaria alternata* (0.12%) were associated with the seeds after 7 days of incubation (Table 1; Figure 5). The examination of farmer's saved seeds showed a variety of associated pathogens including *Fusarium moniliforme*, *Aspergillus niger*, *Aspergillus flavus*, *Penicillium* sp., *Erwinia* sp., *Curvularia lunata*, *Exserohilum turcicum*, *Alternaria alternata*, *Bipolaris maydis*, *Chaetomium* sp. etc. Seed-borne fungi viz. *Aspergillus niger*, *Aspergillus flavus*, *Fusarium verticillioides*, *Rhizopus* spp., *Penicillium* spp., *Curvularia* spp. *Alternaria alternata*, *Curvularia lunata*, *Bipolaris maydis*, *Colletotrichum graminicola*, and *Xanthomonas* sp. were detected in maize seeds (Erasto *et al.* 2023, Fakir 2001, Sreenu *et al.* 2019, Alam *et al.* 2003). Artificial inoculation of *Bipolaris maydis* and *Curvularia lunata* developed characteristic symptoms of leaf blight (Chowdhury *et al.* 2015). Disease incidences usually favored by climate change like rising temperatures, and precarious rainfall patterns have a significant impact on grain yield and quality through lower kernel weight, reduced photosynthesis efficiency, and water stress of maize (Islam and Hoshain 2022; Ahmed *et al.* 2021).

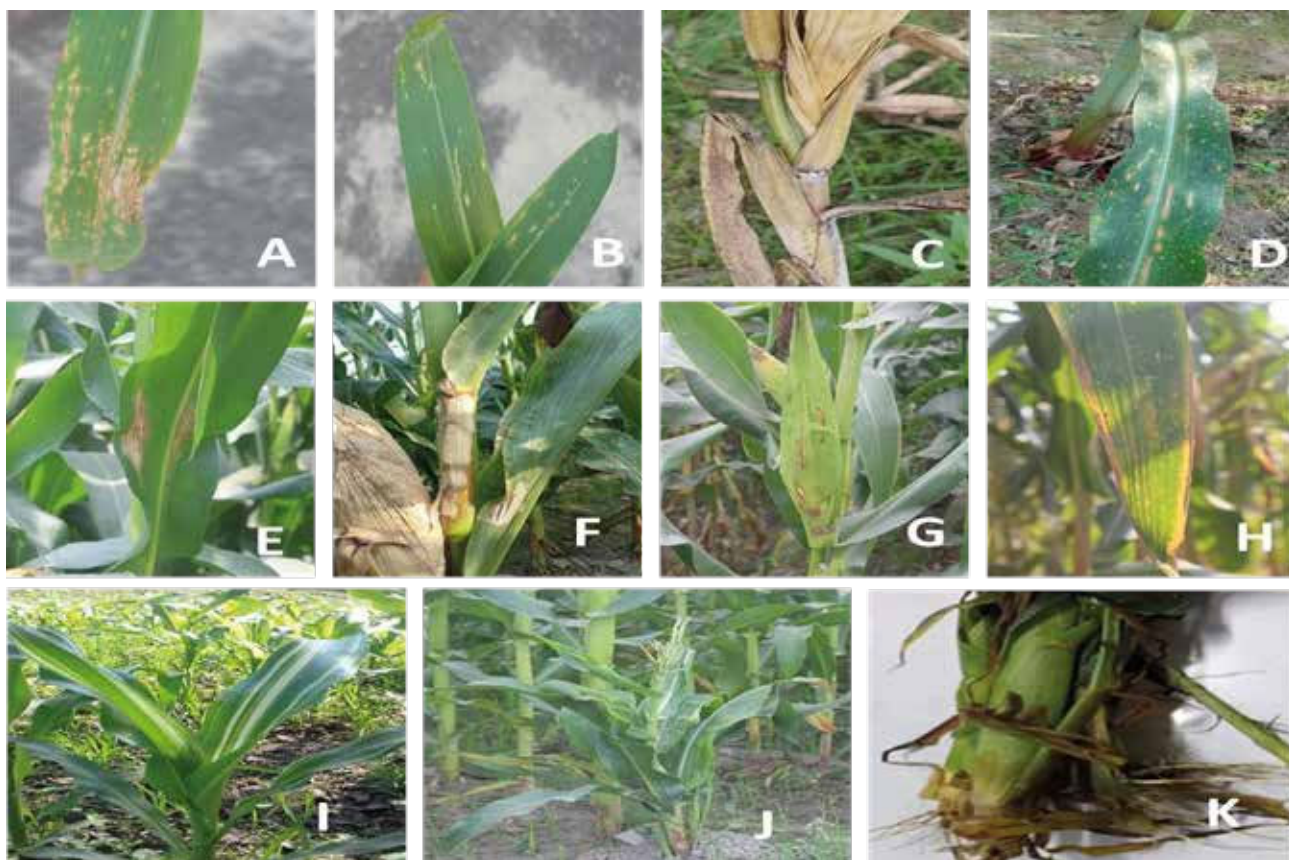


Figure 4. Field symptoms **A)** Southern leaf blight **B)** Curvularia leaf spot **C)** Maize stalk rot **D)** Alternaria leaf blight **E)** Turcicum leaf blight **F)** Sheath blight **G)** Cob sheath blight **H)** Rust on maize leaf **I)** Stripe virus of maize **J)** Bushy stunt of virus **K)** Bacterial wilt

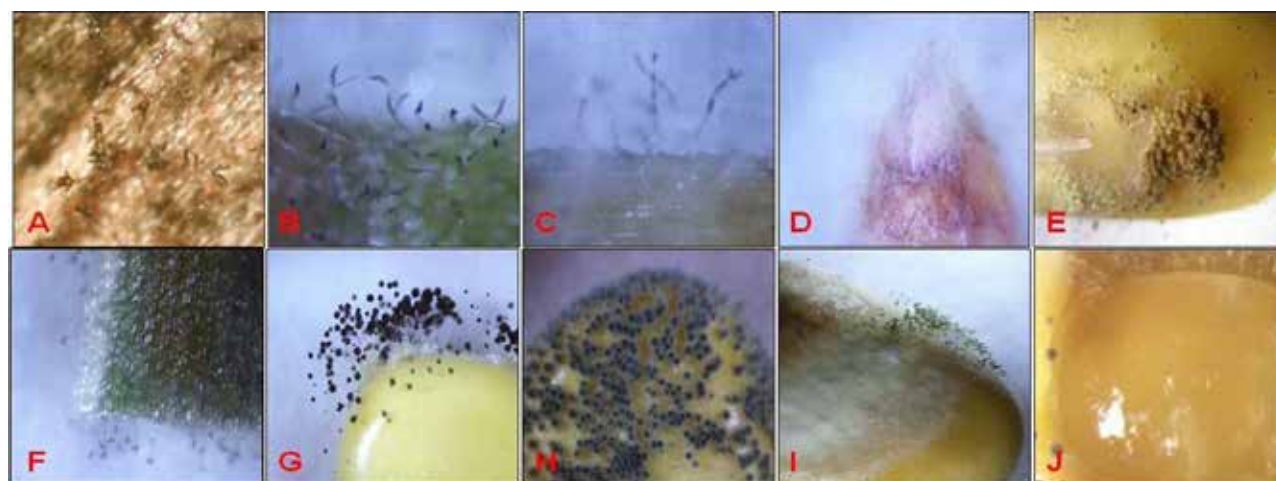


Figure 5. Pathogens associated with farmer's saved seeds: **A)** *Exosporohilum turcicum*; **B)** *Bipolaris maydis*; **C)** *Alternaria alternata*; **D)** *Fusarium moniliforme*; **E)** *Aspergillus flavus*; **F)** *Curvularia lunata*; **G)** *Aspergillus niger*; **H)** *Chaetomium* sp.; **I)** *Penicillium* sp.; and **J)** *Erwinia* sp.

Table 1. Pathogens associated with farmer's saved seeds

Name of the pathogens	Infected seeds (%)
<i>Aspergillus niger</i>	18.50 c
<i>Aspergillus flavus</i>	21.81 b
<i>Fusarium moniliforme</i>	39.88 a
<i>Penicillium</i> sp.	10.25 d
<i>Erwinia</i> sp.	4.38 f
<i>Bipolaris maydis</i>	7.25 e
<i>Curvularia lunata</i>	0.44 g
<i>Chaetomium</i> sp.	0.19 g
<i>Exosporium turcicum</i>	0.13 g
<i>Alternaria alternata</i>	0.12 g

CONCLUSION

A total of 14 diseases were identified in the field including Turcicum leaf blight, Sheath blight, Cob blight, Stalk rot, southern leaf blight, Alternaria leaf blight, Brown spot, cob rot, bacterial wilt, Ear rot, Rust, Curvularia leaf spot, Maize leaf streak, and Bushy stunt. Turcicum leaf blight was recorded as the major disease in the survey regions followed by Sheath blight and Stalk rot. Isolation and artificial inoculation of *Bipolaris maydis* and *Curvularia lunata* proved Koch's postulate by producing characteristic leaf blight symptoms. Health tests of farmer's saved seeds revealed the association of various pathogens including *Fusarium moniliforme*, *Aspergillus niger*, *Aspergillus flavus*, *Penicillium* spp., *Exosporium*, *Curvularia lunata*, *Alternaria alternata*, *Erwinia* sp., etc. The findings of the present investigation demonstrated the priority of the management of Turcicum leaf blight, Sheath blight, and Stalk rot diseases in the surveyed region of Bangladesh.

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