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MYCOFLORAL STUDY OF CULTIVATED MAIZE SEED IN DISTRICT POONCH AZAD JAMMU AND KASHMIR

Mehr-un-Nisa, Mussadiq Hussain, Basharat Mehmood, Muhammad Tariq-Khan, Hafiz Matee-ur-Rehman
Department of Plant Pathology, Faculty of Agriculture, University of the Poonch, Rawalakot, Azad Jammu and Kashmir, Pakistan.

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ABSTRACT

The current study was carried out on the prevalence of mycoflora associated with maize seeds using blotter paper method. Maize seed samples were collected from six different locations of district Poonch, Azad Jammu and Kashmir. Objective of this study was to determine the fungi associated with maize seeds. A total of seven species of fungi were isolated. *Penicillium* spp., *Fusarium* spp., *Aspergillus niger, A. flavus*, *Fusarium* spp., *Fusarium oxysporum* and *Pythium* spp., were identified. Davigali had the infection percentage of 72 % in all locations. Occurrence frequency and type of fungi isolated varied with location. Prevalence of pathogenic fungi with maize seeds of district Poonch was found variable. Resistant varieties of maize, maintaining temperature, relative humidity and their treatment is suggested to reduce disease and increase yield.

Corresponding Author: Hafiz Matee-ur-Rehman
Email: hafizmatee1@gmail.com

INTRODUCTION

Maize (*Zea mays* L.) belongs to family Poaceae. It is an important cereal crop which serves as a source of staple food (Satish et al., 2010). For dietry components, this crop is considered as a main commodity of agriculture worldwide and according to nutritional value this is also the most important cereal (Abd El-Baky and Hussain, 2009). It is cultivated on an area of 1.016 million hectares with 3.037 million tons of production in Pakistan which is very low as compared to other developing countries (Ahmad et al., 2010). The crop is facing sever yield constraints including biotic and abiotic stresses. Among biotic stresses, fungal pathogen is an important one (Ismail, 2001). Some fungi are seed borne which effect quality and quantity of crop and decrease market value (Hussain et al., 2007; Klyszejko et al., 2005). Microorganisms especially fungi are the largest group which play a vital role in effecting the seeds quality. At the initial growth stage, seeds vigor is reduced by these pathogens and plants becomes weak. Fungi that cause seed-borne diseases are difficult to control because hyphae of the fungi are dormant due to establishment. Seed-borne diseases are mostly found in economically important crops (Hussain et al., 2007; Niaz and Dawar, 2009). Early detection of the pathogen is the most crucial step in disease diagnosis and also for management programs (Majumder et al., 2013). District Poonch falls in temperate region where prevalence of pathogenic and saprophytic fungi is very high. For the purpose of seed health, present

study was conducted to evaluate the presence of seed associated mycoflora in maize under natural storage conditions around the district Poonch.

MATERIALS AND METHODS

Seed samples were collected from six different locations of district Poonch, Azad Jammu and Kashmir (Figure 1). The seeds were collected from stores to ensure the natural presence of the associated mycoflora. Seed samples were carried to the Department of Plant Pathology, University of The Poonch, Rawalakot during June to October, 2018 and isolation of associated mycoflora was done.

Figure1. Sampling locations from district Poonch Azad Jammu and Kashmir.

Three layers of blotting paper were placed in the petri dishes and moistened with distilled water. Fifteen seeds were placed at equal distances per petri dish. The plates were incubated at 26±2˚C for a period of eight days. After eight days, the fungal growth was seen on seeds. The seeds with fungal growth were counted and shifted to separate plates containing Potato Dextrose Agar (PDA). Purification and identification of fungal cultures was done by using Illustrated Genera of Fungi (Barnett and Hunter, 1986).

The percentage frequency of occurrence was calculated by using following formula.

$$\text{Percentage frequency} = \frac{\text{No of contaminated seeds}}{\text{Total No. of seeds}} \times 100$$

RESULTS

Seven fungal species belonging to five genera were isolated from maize seeds collected from six different locations of district Poonch. *Aspergillus* was found in all the six sites, *Fusarium* in four sites, *Alternaria* in three sites and *Penicillium* and *Pythium* each in two sites. Seeds from Davigali got maximum mycoflora of 72%, Abbaspur and Paniola 60%, Hajjira 47% and Mandhole 46%. Lowest mycoflora percentage was found in Jandali which was 33% (Table1, figure 2).

Table 1. Infection percentage from maize seeds collected from six locations of District Poonch.

Locations	*A. Alternate*	*Penicillium* spp.	*A. niger*	*A. flavus*	*Fusarium* spp.	*F. oxysporum*	*Pythium* spp.	% frequency
Mendhole	0	0	2	3	1	1	0	46
Abbaspur	3	0	3	2	0	0	1	60
Hajira	1	3	0	3	0	2	0	47
Davigali	1	0	2	4	1	0	2	72
Jandali	0	2	0	3	0	0	0	33
Paniola	1	0	2	3	2	0	2	60

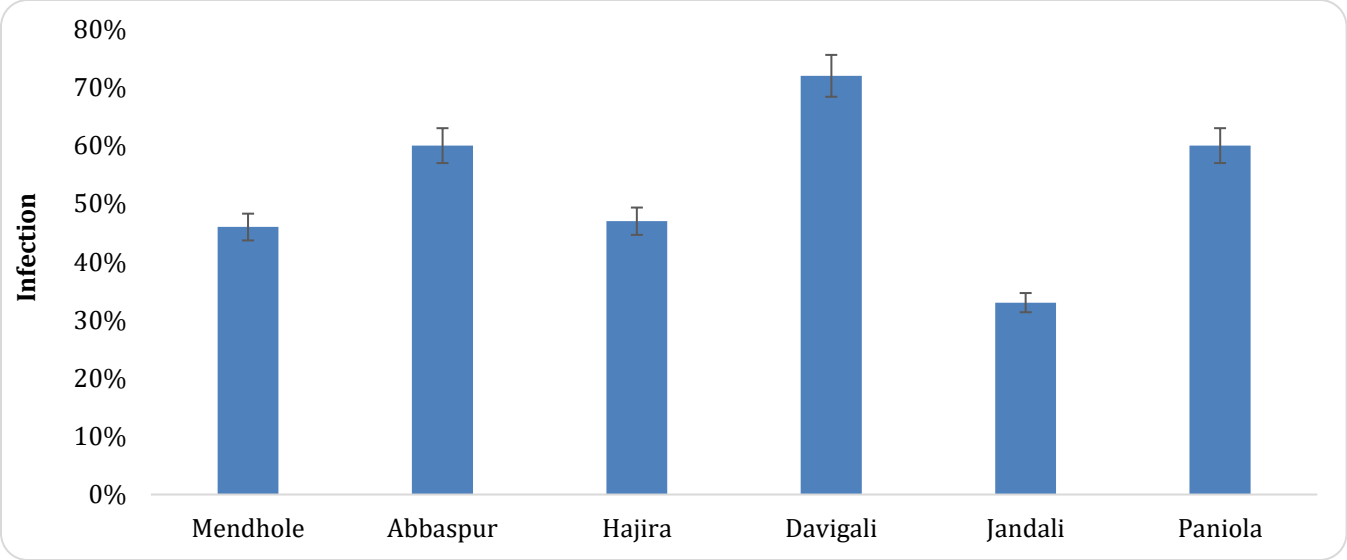


Figure 2. Infection percentage frequency at different locations of Azad Jammu and Kashmir.

Identification of fungi: The isolated fungi were recognized according to charactistics of colony and morphology of spores (Figure 3).

Alternaria alternate: The species *A. alternata* was recognized as it produced powdery and wooly zips of brown color conidia of unequal forms and size. The color of the culture was highly brown. Hyphae were branched, dark brown, thick and septate. Conidiophores were simple, erect and with septate conidia (Figure 3).

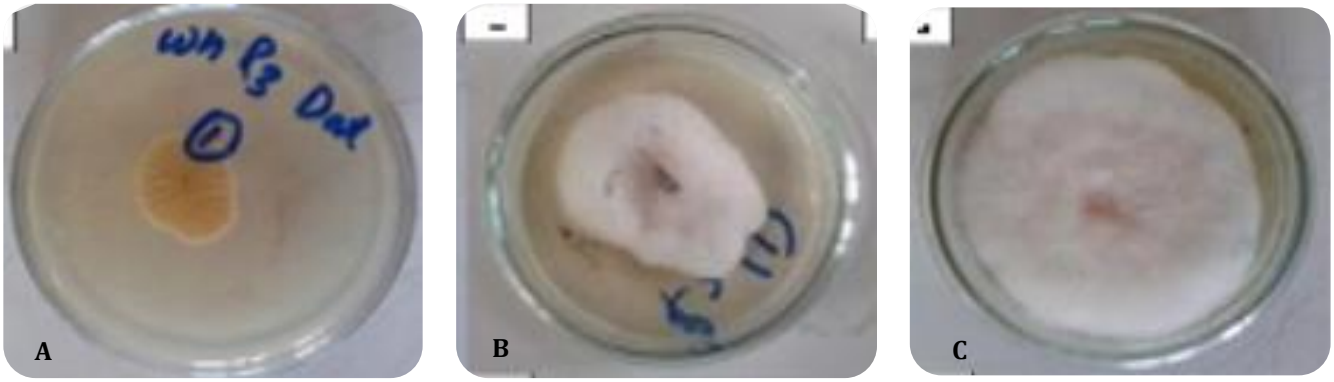
***Aspergillus niger*:** *Aspergillus* colony showed slow growth on seeds, comprising of mycelium that is hard to properly movable white to faintly yellow, which accepts abundant straight and frequently congested conidial structure. Conidiophores arose from protective layer and were hyaline flat or faintly brown close to the apex (Figure 3).

Aspergillus flavus: Colony of *A. flavus* on incubated seed was very light yellow to deep yellow green. The fungus colony created compact globose to radiate conidial heads in green shades. Conidiophores were also present and simple, transparent, colorless, smooth and unbranched. Conidia were charactistically spherical to subspherical.

***Penicillium* spp.:** The colonies of *Penicillium* spp. on PDA were initially white and with the passage of time became blue green. Hyphae were septate, hyaline with simple or branched conidiophores. The conidia were unicellular, round and visualized as chain without branching at the tips of the plialies (Figure 3).

***Fusarium* spp.:** It showed rapid growth on PDA. The texture of the colony was flat to wooly and became pink in color. Conidia were two or more celled, curved, smooth and thick walled (Figure 3).

***Pythium* spp.**: It produced colonies with cottony aerial mycelium on PDA. Sporangia were filamentous and consist of terminal complex of swollen hyphae.



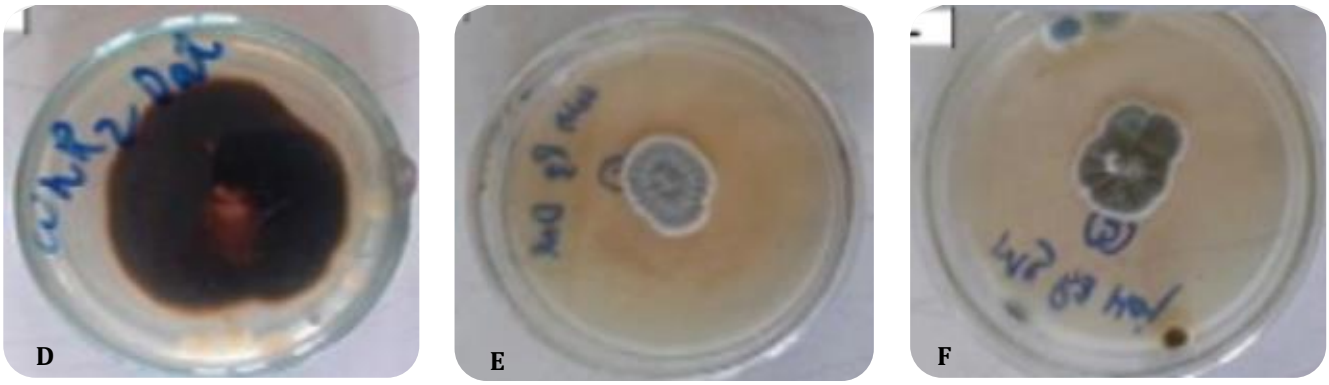


Figure 3. Showing fungal growths (A) *Penicillium* spp., (B) *Fusarium* spp., (C) *Fusarium* spp. (D) *Alternaria alternaria* (E) *Aspergillus* spp. and (F) *Aspergillus flavus* isolated from maize seeds.

DISCUSSION

Seven genera of fungi both saprophytic as well as pathogenic were obtained from samples of maize seeds. Fungi isolated were *A. alternata, Pythium* spp. *Penicillium* spp. *A. niger, A. flavus* and *Fusarium* spp. Percentage infection ranged from 15 to 72% in all the tested locations. Devigali showed 72% infection followed by Abbaspur and Paniola 60%, Hajira 47%, Mendhole 46% and Jandali showed 33% infection. These results are in close conformity with those of Sitara and Akhtar (2007) who isolated several genera and eleven species viz. *A. niger*, *A. flavus*, *A. wentii*, *Cheaturnium* species, *Fusarium oxysporum*, *F. nivale*, *F. semitectum*, *Nigrosporas* species, *Rhizpous* species, *Poma* species from maize by using Freezing Technique and Standard Blotter Method.

It is clear from the present studies that maize seeds have been found infected with different pathogenic fungi and therefore, must be treated with systemic fungicides to avoid primary infection and losses.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHORS' CONTRIBUTION

MUN, MH, BM and MTK designed the study, MUN and HMR conducted the experiments, collected and analyzed the data, MTK supervised the work and all the authors edited and approved the final manuscript.

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