

Studies on Seed Borne Fungi Infecting to *Capsicum* (Chilli) Cultivated in Kannad Region of Aurangabad District

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Abstract: Survey was conducted in Kannad taluka of Aurangabad District from Maharashtra state. Naturally infected pepper seeds were extracted from *Capsicum* fruits collected from cultivated fields during the month of August, September and October 2018 from different localities in this area. During diagnosis it was found that total of three classes of fungi were isolated and identified from seed samples of *Capsicum*. *Colletotrichum capsici*, *Aspergillus niger* and *Aspergillus flavus* were the most frequently isolated fungi with 14%, 13% and 16% occurrence respectively. *C. capsici/A.niger* interaction had the highest occurrence of 2% followed by *A.flavus/A.niger* interaction with 1.20% Fungi association had occurrence are being reported on pepper seeds for the detection of the seed borne fungi. It's important in the management of pepper diseases and can be a useful guide to strategic disease control.

Keywords: *Colletotrichum capsici*, *Aspergillus flavus*, *Aspergillus niger*, Fungi, Agar, Pepper

1. Introduction

Chilli (*Capsicum annum L.* It is grown as annual crop in t) also called red pepper is an important vegetable cum spice crop grown in almost all tropical and subtropical countries of the world. It belongs to the family Solanaceae is native of tropical America (Brazil) where is found growing wild perennially. It is grown as annual crop in the temperate region. Chilli contains numerous chemical including Ascorbic acid, Oleracein, Voltaic oils, Fatty acids, Capsaicinoids, Carotenoids, Vitamins, Proteins, Fibres and Mineral elements. *Capsicum* fruits acts as a source of natural bactericidal agent to be used in food and medical science. Many chilli constituents are important for nutritional value, flavor, aroma, texture and colour. Chillies are low in sodium and cholesterol free, rich in vitamin A, C, and E, a good source of Potassium and Folic acid.

Among four revenue of Maharashtra, Marathwada region has occupied third position in Chilli production..

Capsicum suffers heavy losses due to Fungi, Bacteria, Viruses, and Nematodes many of which are carried through seed. The fungi associated include *Fusarium oxysporum*, *Colletotrichum capsici*, *Aspergillus fumigates*, *Alternaria alternata* and *Penicillium citrinum*. The fungal disease has been observed in three phases i.e. (a) seedling blight or damping off, prevalent in the nursery, (b) leaf spotting and die back which is found at different stages of growth and (c) fruit rot in which the ripe fruits are infected. The fungal disease is both seed borne and air borne and affects seed germination and vigor to a greater extent (Ahmed 1982, Asalmol *et al.*, 2001). For good quality of capsicum it is imperative that seeds must be tested before they are sown in the nursery.

2. Material and Methods

Seed borne fungi associated with chilli were carried out using Agar plate method and rolled paper towel method.

2.1 Materials

The details of the material used during the course of investigation were chilli seed, blotter paper, towel, paper, butter paper, culture media, chemicals, fungicides and bio agents formulations, equipments, glassware, etc.

2.2 Seeds

Untreated seed of chilli Garima 12 were collected from fields of *Capsicum* cultivated in Kannad Taluka.

2.3 Culture media

2.4 Potato dextrose agar

PDA (Potato Dextrose Agar) was used for isolation, of the pathogens and other fungi.

2.5 Mounting medium

Lacto phenol and cotton blue were used as the working media for studying the characteristics of the fungi.

2.6 Chemicals

- 1) Cleaning solution-contains Potassium dichromate 80g. Distilled water 300ml and concentrated Sulphuric acid 400ml was used to clean the glassware's like Petri dishes, flasks and culture tubes.
- 2) Mercuric chloride solution (1:1000) was used for surface sterilization of seeds

2.7 Glassware's

Standard Borosil wabe glassware like petridish, conical flask, glass micro slide, glass cover slip, funnel, pipettes culture tubes and beakers were used during the study.

2.8 Methods

2.9 Collection of seed samples

The seed samples of chilli GARIMA-12 were serially collected from the survey of these areas.

2.10. Cleaning and sterilization of glass wares :

- 1) The glass wares were cleaned by dipping them in cleaning solution of 15 minutes and finally rinsed with running tap water for 30 minutes. Petri dishes were sterilized in a hot air oven at 180 degree for two hours.
- 2) Media were sterilized in an autoclave at a pressure of 15 lbs for 25 minutes. Surface sterilization of seed was done by dipping there in 11000 mercuric chloride solution for 2 minutes follows by three washing with sterilized water.
- 3) The inoculating needle and other metallic instruments were sterilized by dipping them in rectified spirit and heating red hot over the flame of sprit lamp.

2.11. Identifications of fungi detected on seed.

2.12. Agar plate Method

- 1) The seed borne fungi associated with capsicum were isolated by the standard method i.e. agar plate method.
- 2) In this method, potato dextrose agar medium (PDA) was poured aseptically in the sterilized glass Petri dishes of 10 cm diameter @ about 15 to 20ml per Petri dish. After 24 hours of pouring the medium, seeds per petri dish were plated aseptically
- 3) Seed prior to planting were surface sterilized in 0.1 percent mercuric chloride solution for 30 to 45 seconds followed by 3 to 4 times and washing in sterilized water and absorption of excess moisture by keeping the seed on sterilized blotting paper in aseptic condition.
- 4) These Petri dishes were further incubated at $25-27 \pm 10C$ for seven days under 12 hours alternating cycles of NUV light and darkness. These seeds were examined for fungi after five and eight days of incubation.

Attempts were made to detect seed borne micro flora by employing Agar plate method using sample to chilli Garima 12 variety.. Findings on these aspects are being reported.

2.13 Fungal Identification

The seeds infected with fungi were used for isolation of mycoflora. A bit of actively growing mycelium from seed was transferred on PDA slants for purification, which were incubated at $26 \pm 20C$ for 48 hours the isolates thus obtained were maintained separately. Identification of the fungi was done by examining the culture under the microscope through the characters of mycelium, spores and fruiting bodies (Barnett and Hunter 1977).

3. Results and Discussion

A grading of seed samples

Seed samples of two chilli lines Garima -12 were taken from farmer field on Chilli varieties were graded into two categories as

- (i) Normal seeds
- (ii) Abnormal seeds

Predominance of seed -borne fungi/bacteria (%) associated with chilli Garima 12 was isolated by different methods of detection has been reported in table.All five fungi namely *Aspergillus flavus*, *Aspergillus niger*, *Colletotrichum capsici*, *Pencillium citrinum* and *Fusarium annum*. were found to be associated with seeds externally as well as Internally.

Isolated of seed borne fungi

The seed fungi associated with Garima-12 by the standard Agar plate method

| Sr. No. | Fungal pathogen | Percent of Pathogen detected by agar plate method | |
|---------|------------------------------|---|----------|
| | | Normal | Abnormal |
| 1 | <i>Aspergillus flavus</i> | 15 | 16 |
| 2 | <i>Aspergillus niger</i> | 11 | 13 |
| 3 | <i>Colletotrichm capsici</i> | 11 | 14 |
| 4 | <i>Penicillium citrinum</i> | 4.5 | 6 |
| 5 | <i>Fusarium annum</i> | 7.0 | 9 |

Aspergillus flavus (15%) was the most predominant fungus in normal seeds as well as in abnormal seeds (16%) this was followed by *Colletotrichum capsici* (11%) in normal seed and (14%) I abnormal seeds, *Aspergillus niger* (11%) in normal seeds and (13%) in abnormal seeds. Other fungi, which were detected by this method, were *Pencillium citrinum* and *Fusarium annum*.

The result revealed that five species of microflora associated with seed namely, *Aspergillus flavus*, *Aspergillus niger*, *Colletotrichum capsici*, *Pencillium citrinum* and *Fusarium annum* (Seed borne), All the fungi have been detected by all the four methods on Garima 12. It was also fond in result that variety Garima -12 has broad fungi in lesser number showed maximum percentage of germination. Out of a total of five fungi species detected are responsible for infecting diseases in chilli either as seed borne or seedling diseases as per the details below: 1. *Aspergillus flavus*, 2. *Aspergillus niger* 3. *Collectotrichum capsici*, 4. *Penicillium citrinum*, 5. *Fusarium annum*.

4. Conclusion

Based on results obtained from the present study, the following conclusions were drawn among the diagraphic methods.

- 1) Among the methods applied for detecting seed bone and seedling micro flora and percent fungi associated with Garima 12 varieties of chilli, agar plate method was found superior over standard blotter method, rolled paper towel method and nursery tray method.
- 2) A total of nine fungi namely, *Aspergillus flavus*, *Aspergillus niger*, *Colletotrichum capsici*, *Pencillium* and *Fusarium annuum* seed born fungi were isolated.
- 3) The cultivar of Capsicum crop will need to give attention for control of seed borne fungi by applying various diseases management practices.

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