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STUDY OF MYCOTOXIC EFFECT AND CONTROL OF *ASPERGILLUS NIGER* INFECTION IN LEGUMINOSAE (FABACEAE) WITH SPECIAL REFERENCE TO *ARACHIS HYPOGAEA*

Angira Thakar[1], Mrunali Patel*[2], Ankita Badlawala[3], Priti Patel[4]

[1]*Department of Biotechnology, Veer Narmad South Gujarat University, Surat, Gujarat, India*

[2]*Ph.D Research Scholar, Department of Biotechnology, Mehsana Urban Institute of Sciences, Ganpat University, Mehsana, Gujarat, India*

[3]*Department of Biotechnology, Veer Narmad South Gujarat University, Surat, Gujarat, India*

[4]*Department of Biotechnology, Mehsana Urban Institute of Sciences, Ganpat University, Mehsana, Gujarat, India*

**Corresponding author: mrunalipatel1296@gmail.com*

ABSTRACT

The strain of fungus namely *Aspergillus niger* was isolated from infected groundnut crop field, collection from Rajasthan Agricultural Research Institute, Durgapura, Jaipur. A number of fungicides were assayed for antifungal efficacy. The fungicides like Carbendazim, Mancozeb, kemoxyl and Antracol were used and they showed different antifungal activity against isolated infectious crop. The treatment of spray and drenching of different fungicide were observed and infected Leguminosae plant was recovered by the effect of fungicide. The best antifungal activity was observed in Antracol fungicide sample. The fungal resistance to Carbendazim has been reported in minor amount, which make it a very useful in preventing fungal infection in Leguminosae plants.

Keywords: Infectious plants, Fungicides, Resistance, Sensitivity, Collar rot disease.

1. INTRODUCTION

A fungus is any member of the group of eukaryotic organisms; *i.e.,* their cells contain membrane-bound organelles and clearly defined nuclei. The fungi are major disease causing agents on plants and may lose up to 90% agricultural yield [1]. *Aspergillus niger* can cause rotting of numerous fruits, vegetables and other food products, thus causing substantial economic loss. Collar rot caused by *Aspergillus niger* is one of the most important disease of groundnut which is more extensive in the kharif than the rabi and summer seasons and causes more damage in sandy loam and medium black soil [2]. A fungicide is a specific type of pesticides that controls fungal disease by specifically inhibiting or killing the fungus causing the diseases.

Carbendazim is a widely used, broad-spectrum benzimidazole fungicide and a metabolite of benomyl. The fungicide is used to control plant diseases in cereals and fruits, including citrus, bananas, strawberries, pineapples, and pomes.

Mancozeb is a dithiocarbamate non-systemic agricultural fungicide with multi-site, contact-protective activity. It is a combination of two other dithiocarbamates: mameb and zineb.

Antracol contains propineb, with broad spectrum activity against various diseases of rice, chilli, grapes, potato and other vegetables and fruits.

2. METHOD

A disease plant sample was obtained from Rajasthan Agricultural Research Institute, Durgapura, Jaipur. On careful examination of the plant it was predicted that the plant was infected with *Aspergillus niger*.

2.1. Survey, collection and Isolation from Jaipurdistrict

Several diseased peanut plants which showed evident symptoms of collar rot were collected from different fields in Rajasthan Agricultural Research Institute (RARI), Durgapura, Jaipur, Rajasthan.

Affected peanut tissue were cut into pieces of approximately 0.5 to 1 cm and washed under running tap water. After washing under laminar air flow cabinet, the infected tissues were surface sterilized in 70% Ethanol for 2 minutes. It was washed three times in sterilized distilled water. It was dried and placed on surface of Potato Dextrose Agar (PDA). The plate was incubated at 25°C for 7 days.

Fig. 1: Groundnut crop field

2.2. Morphological identification

Morphological identification is the most common method used to identify fungus and is based on similarity of the observable macroscopic and microscopic characteristics [3].

For the morphological identification the Lactophenol Cotton Blue (LPCB) assay was used. LPCB assay was done from isolated colony which was obtained from diseased plant. It was used for focusing and identifying mycelia of fungi as a standard staining procedure. One drop of LPCB was added on microscopicslide and observed over light microscope at 40x. It was later on identified that the infection was collar rot disease [4]. To check activity of different fungicides, the plant was cultured by plant tissue culture method as the study was not possible on land of origin [5].

Fig. 2: Isolation of fungus

2.3. Germination of Groundnut to check pathogenicity test of *Aspergillus niger*

Plastic glasses which contained autoclaved soil and vermi compost approximately were taken. Groundnut seeds were added in the sterilized soil. The plastic glasses were irrigated with water as needed and maintained under favourable conditions. Then it was allowed to grow as matured plant for 15 days.

After maturation of plants, *Aspergillus niger* was added in soil for the infection of plants to cause collar rot diseased. For the infection of plants, I cultured *Aspergillus niger* in peptone water and then injected 5mL of it in plant soil using syringe. Then the plants were allowed to get infected for 8 to 10 days. After the symptoms of collar rot disease were seen, study of effect and control of different fungicides against the *A. niger* infection was done [6].

Fig. 3: Germination of groundnut

2.4. Efficiency of fungicides against *Aspergillus niger* (*in vitro*)

In vitro study of fungi toxicity of different fungicides and plant against *Aspergillus niger* was conducted. Mancozeb, Carbendazim, Antracol, Kemoxyl fungicides were used. Using plant tissue culture method, infected plant was taken as an explant. The leaf and root part of the explant was cut and washed with tween 80 for 15 to 20 minute and washed with running tap water. The infected plant were surface sterilized in 70% ethanol for 2 minute and again washed three time in distilled water for 5 minute respectively. The fungus was grown on Potato Dextrose Agar (PDA) in which the desired quantity of fungicides was incorporated to obtain four different concentration (0.01gm, 0.02gm, 0.03gm,

0.04gm per 20ml of PDA media) under the laminar air flow. Desired quantity of fungicides were mixed thoroughly in PDA just before pouring in sterilized petri plates and allowed to solidify. All the plates were kept in incubator for incubation at 25°C for 7 days and allowed to spread the infection against the fungicides [7].

Infected plants were taken after collar rot disease was caused. Then to cure the disease, fungicides were added (Mancozeb, Carbendazim, Antacol, Kemoxyl) respectively using two different ways: Spray and Drenching.

A 0.01gm of fungicide powder (Mancozeb, Carbendazim, Antracol, Kemoxyle) was dissolved in 10 ml of distilledwater. Each fungicides was sprayed on the infected parts of plants through the spraybottle.

A 0.01 gm of fungicide powder was (Mancozeb, Carbendazim, Antracol, Kemoxyle) dissolved in 10 ml of distilled water. The fungicides were injected into the soils of infected plants.

3. RESULTS

3.1. Morphological identification

Aspergillus niger was isolated from the infected groundnut plant. Morphological characteristic used for identification of *Aspergillus* species.

Fig. 4: microscopic image of *Aspergillus niger*

3.2. Infection of Aspergillus nigerto germinated plant

Collar rot was the most serious disease of groundnut in early stage of growth and they further indicated that the emerged seedling were affected at collar region causing yellowing of lower leaves, slow death due to blighting effect on the shoot, finally leading to death of crown portion.

Fig. 5: Infected plants with *Aspergillus niger*

3.3. Efficiency of fungicides against *Aspergillus niger*

3.3.1. Effect of Mancozeb

After the treatment of spray and drenching of Mancozeb fungicide, it is observed that the collar rot disease can not recovered by this fungicide. Within a few days of infection, plant can stoped the growth and finally plant death can caused by the infection of *Aspergillus niger*.

Fig. 6: Effect of Mancozeb against collar rot disease

3.3.2. Effect of Carbendazim

After the treatment of spray and drenching of Carbendazim fungicide, i observed that the fungicide showed more effect against *Aspergillus niger* by the drenching method. Spray method was not as much effective as a treatment of collar rot disease. So, this

fungicide can be used but it takes a long time for it and the chances are less.

Fig. 7: Effect of Carbendazim against collar rot disease

3.3.3. Effect of Antracol

After the treatment of spray and drenching of Antracol fungicide decrease the *Aspergillus niger* infection and gave the best results against the collar rot disease by the both method spray and drenching also.

Fig. 8: Effect of Antracol against collar rot disease

On the basis of the plating method I observed the growth of *Aspergillus niger* against the different fungicides which were dissolved in agar with a different concentration (0.01mg, 0.02mg, 0.03mg and 0.04mg/ 20ml of media) and I can compared with the treatment of spray and drenching method. I can get the best results at the maximum concentration in which I observed the minimum growth of the *Aspergillus niger*.

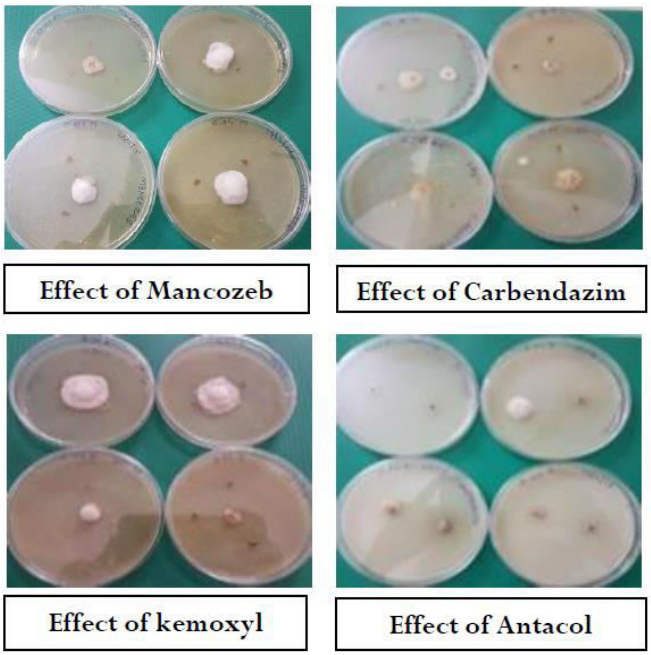

Fig. 9: Effect of Mancozeb, Carbendazi, kemoxyl and Antracol

4. CONCLUSION

Collar rot diseases of groundnut pose a threat to sustainable groundnut cultivation. The pathogenicity of *Aspergillus niger* was tested by inoculation techniques. So, further studies specially on management were carried out with *Aspergillus niger*. Efficacy of fungicides *viz.*, Carbendazim, Mancozeb, Antracol and Kemoxyl were tested by using different method. From the results obtained, it was conclude that the fungicide Antracol showed best results against collar rot disease in groundnut. While Carbendazim showed lower results in comparison to Antracol. While the other two fungicides namely Mancozeb and Kemoxyl did not show enough antifungal activity.

Conflict of interest

None declared

5. REFERENCES

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