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Antagonistic Activity Of Trichoderma Harzianum Against Grain Mold Associated Alternaria Alternata From Sorghum

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Abstract

Grain mold is a one of major fungal disease, which affects the quality and quantity of grain Sorghum. It is a combine association of different group of fungi, which affect the panicle from flowering stage to harvesting stage at favorable conditions. *Alternariaalternata, A. Tennuissima, Aspergillusflavus, A. niger, A. ochraceus, Curvularialunata, Penicilliumchrysogenum, P. citrinum, Rhizopusnigricans*and *R.stolonifer* are dominating member of this complex. In the present study attempt was carried out on biological control of one of the pathogen from these complex *Alternariaalternata* by using *Trichodermaharzianum*. *Trichodermaharzianum* shows antagonistic activity against *Alternariaalternata*. *Trichodermaharzianum* inhibit the growth of *Alternariaalternata*species. Mycelium of *Trichodermaharzianum*enters into the mycelial structure of *Alternariaalternata* and absorbs all the nutrient and sap present in the pathogens body. Which ultimately lead towards the death of mycelial structure of *Alternariaalternata*.

Keywords:*Trichodermaharzianum*, *Alternariaalternata*, Grain mold, Sorghum

Introduction:-

Grain mold is a serious problem in sorghum as concern with yield. Grain mold is a complex fungal disease which hamper the growth and development of sorghum panicles, ultimately reduces the yield.More than 40 genera were associated with grain mold disease, which create complexity in the occurrence and intensity of pathogenicity. Grain mold is a serious constraint in optimal sorghum yield. Early maturing and high yielding hybrid varieties grown in rainy season are more vulnerable for it. Grain molding and grain weathering are two successive events occurring in sorghum caryopsis. Grain molding was occurred at the time of anthesis and grain weathering succeeds after the saprophytic fungi were grown on mature grains.

Such damage reduces the quality of grains and fungi secrete certain secondary metabolites in sorghum like mycotoxins. Such sorghum was not good for consumption (Thakur et al, 2007). Fungal members like *Alternariaalternata, Fusariummoniliformae, Curvularialunata, Penicilliumchrysogenum, Aspergillusniger*and *Aspergillusflavus*were more dominating members reported on the infected heads. Time of their infection to the head is varied from each other which start from the anthesis, physiological maturity and end at the grain maturity. Weather factors like relative humidity plays very important role in the development of grain mold (Das et al, 2020).For the control of infection and spreading the pathogens on different heads, various methods were implemented. Biological control measures were also use for the control of fungal pathogens like; use of leaf extracts for spraying purposes and use of *Trichoderma* to minimize the fungal invagination.

Trichodermaharzianum and *Trichodermaviride* controls the *Alternariaalternata* being a mycoparasite on it. This antibiosis maybe occurred due to some diffusible antifungal substances. The mycelial hyphae of *Trichoderma* were overgrown on the hyphae of *Alternariaalternata* which reduces its growth and limits the further spread (PandeAdarsh, 2010).*Trichodermaharzianum* inhibits the growth of *Alternariaalternata* by dual culture methods. It is promising ecofriendly good bio-controlled agents to control the fungal invagination. It is a cost effective and good for farmers to use frequently (Chaitanya et al, 2018).The growth of fungal pathogen *Alternariaalternata* was severely hampered by the biocontrol agent *Trichodermaharzianum*. It inhibits the growth of *Alternariaalternata* by 67.83% (Yassin et al, 2022).

Material and methods:-

1) Collection of infected samples

For the collection of infected samples the method described by Neergaard (1973) has been adopted. Accordingly random samples of different varieties of panicles were collected from various fields of Sangrampur tehsil of Buldana district, Maharashtra. A composite sample of each variety was

prepared by mixing the individual samples together, preserved in cloth bags in laboratory conditions at room temperature during the studies.

2) Isolation of *Alternaria*:-

The fungi from infected panicles was isolated by using standard Agar plate methods (APM) as recommended by International Seed Testing Association (ISTA 1996), Neergaard (1973).

a) Agar plate method (APM)

In this method, pre-sterilized corning glass petriplates of 10cm diameter were poured with 15ml of autoclaved potato dextrose agar (PDA) medium. On cooling the medium, ten seeds per petriplates of the test sample were placed at equal distance aseptically.

In order to isolate only internal mycoflora, seeds were pre-treated with 0.1% solution of mercuric chloride for two minutes and subsequently thoroughly washed thrice with sterile distilled water and placed on agar plates. And seeds without pre-treatment were also placed on agar plates for external seed mycoflora.

b) Identification of fungi

The fungi occurring on each and every seed in the plates were identified preliminary on the basis of sporulation characters like sexual or asexual spores with the help of stereoscopic binocular microscope. The identification of fungi was made by microscopic observation of the fungal growth. The identification was made with the help of standard literature and monographs. Pure cultures of *Alternariaalternata* were prepared and maintained on potato dextrose agar (PDA) slants.

3) Composition of media used in isolation

Potato Dextrose Agar (PDA)

Peeled potato – 100gm, Dextrose 20g, Agar 20 gm and distilled water 1000ml, pH 5.6.100 gram of potato were taken and peeled; boiled until get soft and squeeze through muslin cloth. Then dextrose was added in it and final volume of solution was made up to 1000ml. In this solution agar was added, pH was adjusted to 5.6.

4) Use of antagonistic microorganisms:-

Antagonistic potential of *Trichodermaharzianum* against *Alternaria* was studied by dual culture method. An agar disc 5mm containing mycelium of *Trichodermaharzianum* was inoculated at the center of PDA poured petriplates and culture discs of the *Alternariaalternata* were placed at the center of the plate. Petriplates were incubated for a week at 25± 1^{o}C plates without antagonists served as control. Two replicates were kept for each treatment and observation on colony diameter (mm) and formation of inhibition zone were recorded.

Experimental result:-

Antagonistic potential of *Trichodermaharzianum* were tested against *Alternariaalternata*by dual culture method and incubated for one week. After incubation period observations were recorded. From the observation it is observed that *Trichodermaharzianum* hampers the growth of *Alternariaalternata*. In *Trichodermaharzianum* treated plate the growth of *Alternariaalternata* were recorded as 2.7 mm as compare to the control plate were *Trichoderma* were not added the growth were recorded as 5.5 mm. It means*Trichodermaharzianum* retards the growth of *Alternariaalternata*. Total percent of inhibition were recorded in the form of zone of inhibition. *Trichodermaharzianum* shows 51% of inhibition against *Alternariaalternata*. Similar type of results was shown by (Chaitanya et al, 2018). They reveal that *Trichodermaharzianum*inhibits the growth of *Alternariaalternata*. It shows 60.71% inhibition against the *Alternariaalternata*.*Alternariaalternata* strains were more susceptible to the concern antagonistic strains of *Trichodermaharzianum*. They show 67.83 % inhibition against the *Alternariaalternata* (Yassin et al, 2022).

Table: Antagonistic activity of *Trichodermaharzianum* against *Alternariaalternata*

Name of Fungi	(C)	(T)	% Inhibition
Alternariaalternata	5.5 mm	2.7 mm	51.0 %

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