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Isolation and Identification of Fungi Associated with fruit Rot of Cucumber (Cucumis sativus L.)

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ABSTRACT

Cucumber fruit (*Cucumis sativus* L.) is believed to have originated from Southern Asia. The isolate and identify fungi responsible for fruit rot of Cucumber in the study area. The present investigation was carried to study the different fungal spp. present in various Cucumber fruit. Fungi, which caused Cucumber fruit rot in latur district area, were isolated from various samples of different Cucumber fruit rot in flied and Market, Isolated from diseased Cucumber fruit rot on PDA media. The isolated fungi were identified as *Macrophomina phaseolina*, *Fusarium oxysporum* and *Phytophthora colocasiae* on the basis of their microscopic characteristics. Finally, the pathogenicity tests showed that all the fungi isolated were pathogenic to all the Cucumber fruit rot.

Keywords: Cucumber fruit rot, Fungi and Potato Dextrose Agar (PDA) media and pathogenicity tests.

INTRODUCTION

Cucumber (*Cucumis sativus L.*) belongs to Curcumas genus in *Cucurbitaceae* family and is an economically important fruit vegetable. There are three wild or semi- wild varieties of Cucumber: *C. sativus* L. var. hardwickii, *C. sativus* L, var. sikkimensis, *C. sativus* L, var, xishuang bannanesis. Cucumber is indigenous to India and likely originated from the foothills of the Himalayan Mountain (Sebastian et al, 2010 and Lv J, et al, 2012). Cucumber was cultivated ~ 3000 years ago in India, and it seems to spread rapidly to Western Asia, and then to Southern Europe (Lv J, et al 2012). Cucumber was introduced respectively to North China through the Silk Route and to South China from Burma and India-China border and subsequently spread to East Asia (Lv J, et al 2012).

Isolation and identification of rot fungi on post-harvest of pepper (Capsicum annuum L.), were studied to determine the presence of fungi, isolate the fungi associated with deteriorated peppers, identify the fungal species and their relative abundance in this fruit. Some infested and healthy samples of Capsicum annuum were collected from Anyigba market three times in sterile polyethene bag and transported to the laboratory. The collected infested samples were sterilized, cultured on Potato Dextrose Agar for isolation. The cultures were incubated at room temperature and later subcultured to have purer culture. Fresh and healthy pepper (Capsicum annuum L.) fruits were also inoculated to test for pathogenicity according to Kock's postulate, then the fungal were subcultured for microscopic examination. The percentage of fungal occurrence was calculated. Rhizopus stolonifer, Aspergillus niger, Fusarium solani, Alternaria alternate, Geotrichum candidum and Mucor pussilus were isolated. Aspergillus niger and Rhizopus stolonifer were the most common and the most abundant fungi associated with the deterioration of Capsicum annuum from the market. The fresh

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International agencies that monitor world food resource have acknowledged that one of the most feasible options for meeting future food needs is reductions of post- harvest losses in our cultivated crops (Kelman, 1984). Sequel to that, since microorganism (pathogens) are mostly the casual agents of deterioration of this important fruit and vegetables i.e. sweet oranges, cucumber and lettuce, after harvest all over the nation, thus the need to isolation and identify the specific pathogens that causes this problem is highly imperative for proper selection of suitable protection and control strategy for sustainable production of fruit and vegetables in order to ensure food availability and its security globally. Thus the present study was aimed to evaluate the fungal spp. In laboratory conditions identify and isolate pathogencity tests the fungal associated with their Cucumber fruit rot.

MATERIAL METHODS

1. Study area

Cucumber fruit rot were selected for this study. The diseased samples were collected from various part of Latur district. The study was carried out in such as Latur, Ahmadpur, Chakur, Walandi, Deoni, Murud, Kassar Sirasi, Nalegaon, Udgir, Nilanga.

2. Collection of samples

Diseased part of fruits rot samples were collected in selected region of Latur district, from January to March 2024 in sterilized condition and stored in clean polythene bag for the isolation of pathogenic fungi carried to the Department of Botany, Kai. Rasika Mahavidyalaya Deoni.

3. Surface sterilization of sample

The collected samples were washed with running tap water to remove solid and dust particles. Fruits rots samples was cut in to small fragments (0.5-1cm). Then the samples are surface sterilized using 0.5% Sodium hypochlorite solution for 2 minutes which is followed by 90% ethyl alcohol for 2 minutes. Then the samples are washed using distilled water 1-2 times and kept aside and dried using sterile blotting paper. (Dobranic et. al.,1995).

4. Pathogenicity test

The isolated fungi were tested for their ability to induce rot in healthy cucumber fruits. Healthy samples of cucumber fruits were washed and surface sterilized with 75% ethanol and then washed distilled water. With the aid of a sterile Cork borer, a 5 mm diameter cylindrical hole was made into the

healthy cucumber fruits and the plugs pulled out. In each hole, a 3 mm diameter mycelia disc of pure culture of each of the fungal isolates were introduced into the hole. The plug was carefully replaced and the wounded area sealed with petroleum jelly. The inoculated fruits were incubated at room temperature (28 ± 2ºC) for 10 days. Inoculated cucumber fruits were observed for rot development at two, four, six, eight and ten days after inoculation. The degree of pathogenicity of each fungus on the inoculated samples was determined by measuring the length and width of rotten parts at two days intervals for ten days (Jidda and Muhammad 2017). The measurement was taken by dividing the cucumber into two equal parts.

5. Isolation of pathogenic fungi

For the isolation of pathogenic fungi Potato dextrose agar media (PDA) supplemented with tetracycline or streptomycin to avoid the bacterial contamination was used and samples were inoculated into the petri plates containing medium. To each Petri plates about fruit rot part fragments are placed while one at the centre and one are arranged at the periphery of the plate, and then petriplates were sealed and incubated it for 1 – 8 days at room temperature (25±20). Obtained fungi were identified and isolation on the basis of colony characteristics and microscopic examinations by using relevant literature (Watanabe. 2010. Nagamani. et.al, 2005, Gilman. 2001 Barnet .1972)

RESULTS AND DISCUSSION

This work was mainly concentrated on the isolation and indication of pathogenic fungi causing fruit rot diseases in Cucumber Latur district from January to March 2024. Before the development of the fruit the fungal pathogens located in fruit rot parts of the plant. These pathogenic fungi were isolated from fruit parts of Cucumber plant. We could able to isolate 3 pathogenic fungal species from diseased part of Cucumber fruit plant responsible for 3 diseases. Among them 3 spp. Isolated from fruit. Had the highest frequency of distribution *Macrophomina phaseolina*, *Phytophthora colocasiae* and *Fusarium oxysporum* isolated in all samples and is responsible for fruit, highest disease incidence of fruit was recorded in the study area.

(Yaji et al., 2016) similarly reported the presence of *Aspergillus* spp. in all their surveyed locations and concluded that fruits sold in all these locations were contaminated. Isolations from diseased cucumber fruits recorded A. niger (percentage occurrence =18.4) as the most occurring fungi among *Aspergillus* spp., followed by A. flavus (14.8%) and *A. ustus* (11.0%). The least encountered fungi were *A. oryzae* and *A. wentii*. Rotten cucumber fruits were similarly reported (Al-Sadi et al., 2011; Chiejina and Onaebi, 2013; Yaji et al., 2016) to be host to *Aspergillus* species. (Jidda and Benjamin 2016) found *Aspergillus* species as the dominant fungal isolates in tomato, orange, onions and other spoiled fruits in Nigeria. All the *Aspergillus* spp. found in the present study was pathogenic to fresh cucumber fruit samples. A. ustus was the most pathogenic isolate with the highest rot diameter (47mm) that led to 25% loss in fresh fruit weight. Loss in fresh fruit weight was due to loss in water content resulting from rots induced by the inoculated pathogens. The findings agreed with the reports of (Akinmusire 2011) that Aspergillus spp. were pathogenic to all their tested fruits. Aspergillus spp. were however, found by (Al-Sadi et al., 2011) to be weakly pathogenic to immature cucumber fruits. Among the yeasts, *Z. bailii* (Isolation frequency =22.0%) were the most frequently isolated fungi from the diseased cucumber fruits followed by *S. cerevisiae* (15.3%). Fruit deterioration was also high among the yeasts (*Saccharomyces spp. and Z. bailii*) inoculated samples. These yeasts were also highly pathogenic to fresh cucumber fruits with rot diameter reaching up to 43mm which resulted in more than 24% loss in fresh fruit weight. *Z. bailii* has

been found to be the major cause of food spoilage in many different products including fruit juices, sauces, carbonated soft drinks, and ketchup. These yeasts have a reputation of being able to grow on a very high concentration of sugars (*Pitt and Hocking,* 2009) and have caused significant economic losses to the food industry (*Anonymous,* 2016).

CONCLUSION

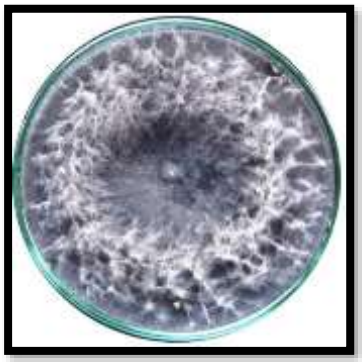
It's evident from the study that, there are various fungi

The present study has shown that associated with diseased cucumber samples but actually caused the spoilage of cucumber. The isolation and identification of the fungal pathogens *Macrophomina phaseolina*, *Fusarium oxysporum* and *Phytophthora colocasiae* hawed that *Macrophomina phaseolina* and *Phytophthora colocasiae* had the highest frequency of distribution. The pathogenicity tests haws that *Fusarium oxysporum* the most pathogenic with rapid lesion development. Based on the results obtained from frequency distribution and pathogenicity, it could be concluded that *Macrophomina phaseolina* and *Phytophthora colocasiae* even with the highest frequency, is not the most pathogenic. Identifying fungi that cause spoilage to cucumber fruits will help to adopt appropriate preventive and control measures that will help minimize fungi infection on cucumber fruits

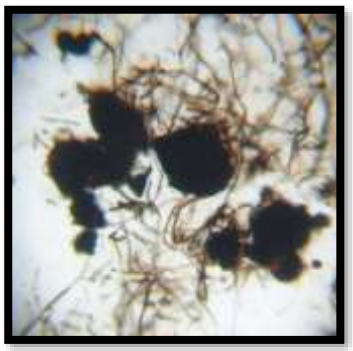
PHOTO PLATE-I



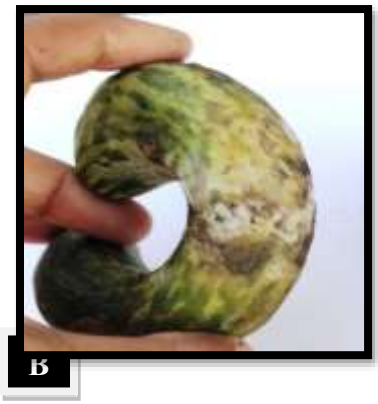
Diseased fruit



Colony plate



Macrophomina phaseolina



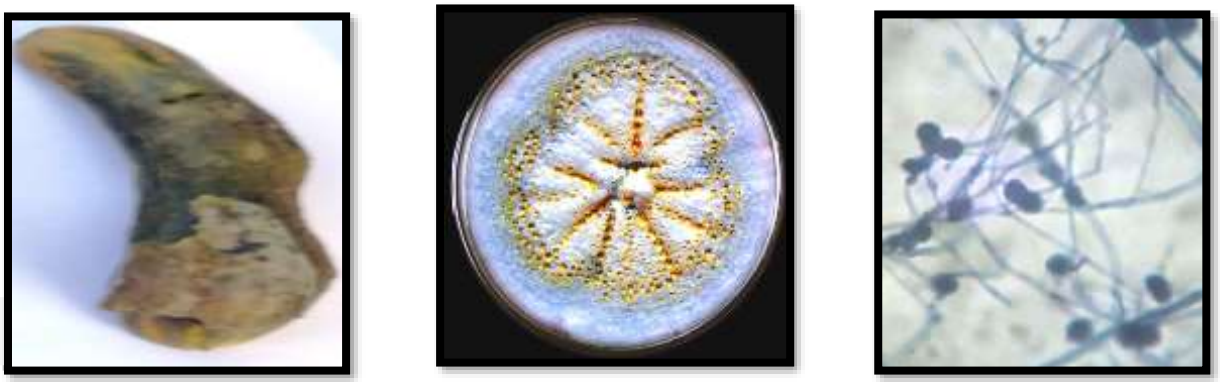
Diseased fruit



Colony plate



Fusarium proliferatum



Diseased fruit **Colony plate** ***Phytophthora colocasiae***

Fig. Fungal diseases of Cucumber fruit, Macrophomina phaseolina, Fusarium proliferatum Phytophthora colocasiae

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