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Rhizospheric Fungus: Morphological Characterization Of Rhizosphere Flower On Onion Plant In Enrekang District

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Abstract

The control of moler disease caused by Fusarium oxysporum on shallot plants in Enrekang Regency still relies on the use of chemical fungicides. Plants that are symbiotic with microbes will produce phytohormones optimally so that it will trigger the tolerance level of plants to abiotic and biotic stresses including plant pathogens, so it is necessary to explore the rhizosphere fungi of onion plants in Enrekang Regency to identify morphologically. This study used a red onion soil rhizosphere soil sample taken at the shallot farming center in Enrekang Regency, isolation and testing were carried out in the plant disease laboratory of Hasanuddin University. The results achieved were 20 isolates with 5 genera of fungi namely Fusarium on isolates 1,2,4,12 and 13, Gliocladium on isolates number 15,

Keywords: *Identification, Macroscopic, Microscopic*

1. INTRODUCTION

Shallots are an agricultural commodity whose production stability is maintained in the midst of the COVID-19 pandemic and the drought season. FAO has warned of a potential food crisis as a result of the COVID-19 pandemic and drought. Various efforts have been made to maintain shallot production, one of which is preventing root rot disease or moler disease caused by the fungus Fusarium oxysporum f.sp. cepae (FOC), with attack intensity 72% - 83.33% on susceptible varieties(Prakoso, et., al., 2016). Moler disease in Enrekang district has been reported to be identified macroscopically and microscopically as Fusarium oxsysporum(Hikmahwati, et al, 2020)

Moler disease control in shallots uses chemical fungicides, the residues of which have an impact on the quality of food and feed, pollute the environment and cause health problems for the community. (Zulfikar, 2017) Zulfikar reported that the control of shallots in Enrekang Regency is focused on pesticides, where as many as 63 types of pesticides, there are 9 types that are not officially registered, 65% of farmers spraying at intervals of once every two days, 22% increasing the dose by 900% from the recommended dose.

Alternative use of pesticides is the use of environmentally friendly biocontrol agents. Biocontrol can be isolated from the rhizosphere around plant roots, including a group of fungi that are able to stimulate plant growth, then called Plant Growth Promoting Fungi (PGPF) and are able to act as antagonists that can suppress the growth of plant pathogens. Rhizospheric microbes collected from bamboo, shallot, grape, and guava plantations showed antagonistic effectiveness against the pathogens

Fusarium oxysporum and Rhizoctonia solani with an inhibition percentage of 52.96 - 92.59%(Xa & Nghia, 2020). Isolates of rhizosphere fungi in shallots were reported to have the ability to suppress the growth of pathogenic Foc in vitro with an inhibition percentage between 65.58 – 84.71% and could trigger the germination of shallot seeds by isolates of the genus Aspergillus and Paecilomyces.(Suganda., 2020) Through the dual culture test, three types of Trichoderma isolated from the tomato rhozosphere were able to suppress soil-borne pathogens by 65%. (Hammad et al., 2021). An et al., (2021)reported that Trichoderma asperellum which is a rhizosphere fungus on Malus sieversisi can suppress the pathogen Fusarium oxysporum (FOX68) by 66.7%. Trichodema sp is also reported to increase spinach growth(Susanti et al., 2021)

Rhizosphere fungi also produce the hormones Indole-3-acetic acid (IAA) and Gibberellins (GA3) which trigger plant resistance and plant growth. From the rhizosphere of aromatic rice Pulu mandoti in Tana Toraja there is a fungus that produces IAA ranging from 0.048-2.190 mg/(Abri et. al, 2015). The fungi isolated from cocoa plantations are the genera Gliocladium and Trichoderma(Tambingsila, 2016), Mycena sp, Lycoperdon sp, Auricularia sp, Schizophyllum sp, Coprinus sp, Tremella sp, Crepidotus sp, Tremetes sp and Pleurotus sp. and produces the hormone GA3(Rahim and Suherman, 2019). Aspergilus niger produced IAA acid on the 6th day of incubation at 25 °C and pH 6.0 and GA3 production on day 12 at 30 °C and pH 5.0(Bilkay et al., 2010).Turaeva et al., (2020)reported that the rhizosphere fungus Trichoderma harzianum produced IAA in the range of 0.16 mg/ml and 0.74 mg/ml and GA 0.318 mg/ml and 0.17 mg/ml, respectively. However, the rhizsofer fungus of shallots in Enrekang Regency has never been isolated and morphologically identified so that the diversity of species and their roles are unknown. Morphological identification can be done through macroscopic and microscopic observations.(Kurniati, et al (2020) conduct macroscopic observations to identify seed-borne fungi.

Rhizosphere fungi in general have the potential as antagonists and are able to produce auxin and gibberellin hormones in plants, so research is needed to isolate rhizosphere fungi in shallot plantations in Enrekang district and to know the morphological characteristics to determine the genus.

2. RESEARCH METHODS

The research implementation method is carried out in several stages (Figure 1)

Rhizosphere Fungal Sampling in the Field

Rhizosphere fungi were collected from the soil and plants around healthy plantations at the shallot production center in Enrekang Regency, South Sulawesi. The samples taken were soil, roots and all parts of healthy plants, wrapped in newsprint to be tested at the Plant Disease Laboratory, Faculty of Agriculture, Hasanuddin University.

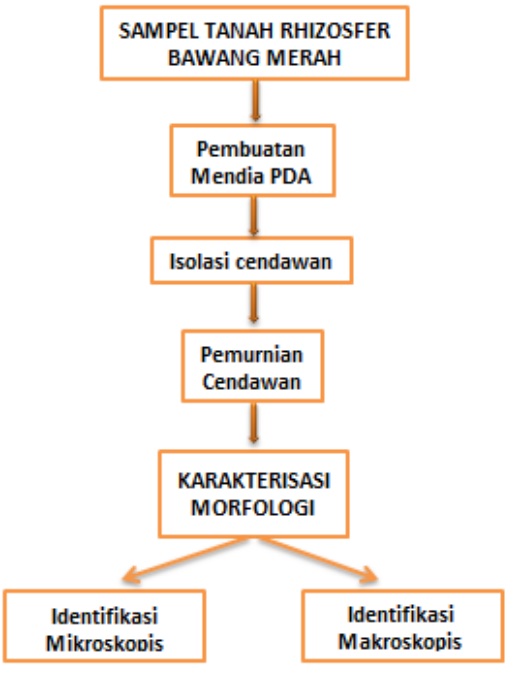

Figure 1: Flowchart of research implementation procedures

Preparation of Growing Medium

The growing medium used was Potato Dextrose Agar (PDA) which was made from a mixture of 200 g of potato, 20 g of granulated sugar, 16 g of agar flour, 1,000 ml of distilled water and the addition of Streptomycin sulfate as an antibiotic contaminant bacteria.

Isolation of rhizosphere fungi from onion plantations

Soil sample of 1 g of soil was diluted with sterile water (10-4) then 1 ml of the dilution was poured into PDA media. After 48 hours of incubation, the temperature was 28 0C, then purified.

Plant samples were washed and cut (4mm) then surface sterilized and grown on PDA to be incubated at room temperature for 5 days, then purified.

Fungus Identification

Identification is done by observing the macroscopic and microscopic characteristics of the fungus. The macroscopic characteristics observed were colony color and microscopic characteristics including hyphae, spores, sporangium, conidia and conidiophores as determinants of the type of fungus.

Documenting the isolates using a camera microscope and identification was carried out by referring to the Barnett and Hunter's (2006) book on fungal identification.(Hunters, 2006)

3. RESULTS AND DISCUSSION

The results of the isolation of the red onion rhizosphere fungus from Enrekang Padi Regency on day 7 of DAI as many as 20 purified isolates had varied morphological characteristics (Figure 1). The results of observations of morphological characterization of rhizosphere fungi showed that all isolates had diameters ranging from 2.5 to 9 cm. The color of the colonies obtained was white in isolates 1, 2, 13, 18 and 20, some were grayish white in isolate 12, white orange in isolate number 4 and there were also isolates that were dark green and white in isolates 5 and 9, green the leaves on isolate number 11 were green with white circles on isolates 15, 16, 17 and 19 and green to black in isolates 3,6,7,8,10 and 14, . This is in line with Gusmiyati and Larekeng (2020)(Gusmiaty and Siti Halimah Larekeng, 2020) who reported that the rhizosphere fungus of mahogany trees in the Unhas educational forest was macroscopically white, gray green to blackish green.

(a)

(b)

(c)

(d)

(e)

Figure 2. Macroscopic and Microscopic Morphological Observations on the rhizosphere Fungi of shallots from Enrekang Regency (a) Genus Fusarium, (b) Aspergillus, (c) Trichoderma, (d) Penicillium and (e) Glicladium

The results of morphological observations of 20 isolates of shallot rhizosphere fungi from Enrekang Regency microscopically using an electron microscope (Table 1) were carried out by observing the generative structures, namely spores and conidiophores. There were 18 isolates that were identified, the other two isolates of the fungus were not identified correctly because they did not find the spores.

Makroskopis	Nomor Isolat	Mikroskopis	No Isolat	Genus
Putih	1, 2, 18, 20	Bulat lonjong (Barnet dan Hunter, 1972)	1,2, 4,12, 13	Fusarium (Barnet dan Hunter, 1972)
Putih Orange	4, 13			
Putih Abu	12			
Hijau kehitaman dengan lingkaran putih Gusmiyati dan Larekeng (2020)	3, 7, 10, 14	Bulat dengan konidiofor bulat bagian ujung (Barnet dan Hunter, 1972)	3,6,7,8,1014	Aspergillus (Barnet dan Hunter, 1972)
Hijau daun	6			
Hijau kehitaman	8			
Hijau campur putih Gusmiyati dan Larekeng (2020)	5, 9	Bulat, konidiofor bercabang, ujung oval dan meruncing (Barnet dan Hunter, 1972)	5,9	Trichoderma (Barnet dan Hunter, 1972)
Hijau daun dengan lingkran putih	11	Bulat, konidiofor bercabang dengan tangkai (Barnet dan Hunter, 1972)	11	Penicillium (Barnet dan Hunter, 1972)
Hijau tua dengan lingkaran putih Gusmiyati dan Larekeng (2020)	15, 16, 17, 19	Bulat, konidiofor bercabang di bagian ujung (Barnet dan Hunter, 1972)	15, 16, 17, 19	Gliocladium (Barnet dan Hunter, 1972)

Table 2. Genus of macroscopic and microscopic observations of shallot rhizosphere fungi from Enrekang Regency

The results of the identification of 18 isolates of the fungus found 4 different forms of conidia and conidiophores. There are conidia that are oval in shape, namely in isolates number 1, 2, 4, 12, 13, small round conidia with branching conidiophores at the ends were found in isolates number 15, 16, 17, and 19, small round conidia with rounded conidiophores was found in isolates number 3, 6, 7, 8, 10, 14 and there were also small round conidia with stem and branched conidiophores at the ends found in isolate number 11 and small round conidia with crossed conidiophores with a tapered oval shape were found in isolates numbers 5 and 9. This is also in line with Gusmiyati and Larekeng (2020)(Gusmiaty and Siti Halimah Larekeng, 2020)who reported that the rhizosphere fungus in mahogany trees in the Unhas educational forest microscopically had a branching conidiophore shape like a brush that was densely arranged and conidia, some had many conidiophores and branched like pyramids, phialid and looked slender and long, especially at the apex of the branches. with conidia in the form of semi-spherical to short oval clumps, and some have spherical spores.

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Based on the results of macroscopic and microscopic observations (table 2), there were 5 genera that could be determined from 18 isolates, namely the genus Fusarium, Trichoderma, Aspergillus, Glicladium and Penicillium. This is in line with the key of determination(Barnet and Hunter, 1972). Rhizosphere fungi found in tobacco are Trichodrma, Pennicilium and Fusarium (Mubashar et al., 2011).

4. CONCLUSION

The results of microbial isolation from the rhizosphere of shallot plants from Enrekang Regency obtained 18 isolates of fungi that had varied morphological characteristics, both from colony color and texture. The results of microscopic observations, obtained 5 genera of microbes from the rhizosphere

onion plants namely Fusarium on isolates 1,2,4,12 and 13, Gliocladium on isolates 15, 16, 17 and 19, Trichoderma on isolates 5 and 9, Aspergillus on isolates 3,6,7,8, 10 and 14 and Penicillium on isolate number 11

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Fusarium oxysporum on shallot (Allium ascolonicum L) in Enrekang district. The author would like to thank for the opportunity and financial support provided.

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