

SHORT COMMUNICATION

***Fusarium* species isolated from forest soil samples**

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ABSTRACT

A total of 46 isolates of *Fusarium* were isolated from six forest soil samples in Muka Head, Teluk Bahang, Pulau Pinang. Two *Fusarium* species were identified from the soil samples namely, *F. solani* (93.5%) and *F. oxysporum* (6.5%). The present study showed that the diversity and occurrences of *Fusarium* species in forest soil was low compared to cultivated soils.

Keywords: forest soil, *F. solani*, *F. oxysporum*

INTRODUCTION

The genus *Fusarium* is one of the most common soil fungi, commonly associated with organic matter, plant roots and debris (Burgess, 1981; Summerell *et al.*, 1993). The fungus is widely distributed worldwide and have been isolated from a variety of soils (Kommendahl *et al.*, 1975; El-Gindy and Saad, 1990; Burgess and Summerell, 1992; Mandeel and Abbas, 1994; Latiffah *et al.*, 2009; Latiffah *et al.*, 2010a and b). However, a lot of studies have been done on the occurrence of *Fusarium* in cultivated soils compared to uncultivated soils. Thus, the present study was conducted to acquire data on the occurrence of *Fusarium* species from forest soil samples collected in a forest area at Muka Head, Teluk Bahang, Pulau Pinang.

MATERIALS AND METHODS

Soil samples

A total of six samples of forest soils were collected from an area (approximately 4 m x 4 m) in Muka Head, Teluk Bahang, Pulau Pinang. Approximately 1 kg of soil was taken from a depth of about 0 - 5 cm. Each soil sample was kept in a plastic bag and brought back to the lab for further analysis.

All the samples were air-dried for 2 - 3 days. After drying, the soils were ground using mortar and pestle, and sieved with 0.5 mm sieve to generate soil particles of a consistent size. The soils were used for soil analysis and isolation of *Fusarium* isolates.

Soil analysis

The forest soil samples were analysed for their texture, pH and moisture. Feel method was used to determine the soil

texture which was based on the procedure by Thein (1979). The formation of the ribbon is related to the clay contents and it is used to categorize soils as loams, clay loams and clays. The soil texture compositions are defined based on the USDA Texture Triangle.

Soil pH was measured by weighing 30 g of the soil samples and put in a 100 ml beaker. 75 ml of distilled water were added and mixed well. The mixture was then incubated at room temperature (27 ± 1 °C) for 24 h to ensure all substances in the soil sample were diluted. The pH reading was taken and recorded after 24 h of incubation. The average of pH reading was then calculated to ensure the accuracy of the results (Head, 1980).

For calculation of soil moisture, 10 g of each soil sample was put in a Petri dish and weighted. Then, the soil sample was incubated in an oven at 105 °C for 48 h. After 48 h of incubation, the soil sample was taken out and cools down at room temperature. The soil sample was weighted again and the moisture content of the soil sample was calculated (Head, 1980). The percentage of moisture in the soil samples was calculated based on the following formula:

$$\text{Soil moisture (\%)} = \frac{(\text{Weight of petri dish} + \text{soil before incubation}) \text{g} - (\text{Weight of petri dish} + \text{soil after incubation}) \text{g}}{(\text{Weight of soil sample}) \text{g}}$$

Isolation and Identification of *Fusarium* species

Three methods were used for isolation of *Fusarium* isolates from the forest soil samples namely, soil dilution, debris isolation and direct isolation techniques. The methods were based on the method described in The

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Table 1: Texture, pH, moisture and Fusarium species isolated from all soil samples

Soil sample	Texture	pH	Moisture (%)	Fusarium species
S1	Loamy sand	5.24	0.37	<i>F. oxysporum</i> <i>F. solani</i>
S2	Loamy sand	4.65	0.81	<i>F. solani</i>
S3	Loamy sand	5.72	0.17	<i>F. solani</i>
S4	Loamy sand	5.27	0.27	<i>F. solani</i>
S5	Loamy sand	5.32	0.31	<i>F. solani</i>
S6	Loamy sand	5.10	0.35	<i>F. solani</i>

Fusarium Laboratory Manual (Leslie and Summerell, 2006) for isolation of *Fusarium* species from soils. The morphological characteristics used for identification, and the media used for isolation and identification were also adapted from the manual.

For identification purposes, description of species was according to the description in the classification manual of Nelson *et al.* (1983) and The *Fusarium* Laboratory Manual (Leslie and Summerell, 2006).

RESULTS

The *Fusarium* species successfully isolated, the soil texture, pH and moisture level are shown in Table 1. The texture for all soil samples was loamy sand. Loamy sand forms a ball, but will not form a ribbon (Brady and Weil, 2008). Loamy sand contains 70 – 90 % sand, 0 – 30 % silt and 0 – 15 % clay that feels loose and gritty (Biondo and Lee, 1997).

The pH value of the forest soil samples ranging from 4.65 to 5.72 which was acidic. All the soil samples showed low moisture content with less than 1 % which was 0.167 to 0.368.

A total of 46 isolates of *Fusarium* were isolated from the six soil samples. Based on the morphological characteristics, 43 isolates (93.5%) were identified as *F. solani* while only three isolates were identified as *F. oxysporum* (6.5%). *F. solani* isolates were recovered from all the six soil samples and *F. oxysporum* isolates were only obtained from soil sample S1. Both isolates of *F. solani* and *F. oxysporum* can be differentiated by the arrangement of the conidiogenous cells and the shape of the macroconidia in which isolates of *F. solani* produced long monopodialides and, stout and robust macroconidia. Isolates of *F. oxysporum* produced short monopodialides and, straight and slightly curved macroconidia.

Most of the *Fusarium* isolates were recovered using direct isolation technique in which 34 isolates of *F. solani* and three isolates of *F. oxysporum* were successfully isolated. Eight isolates of *F. solani* were obtained from soil dilution plate technique and only one isolate of *F. solani* from debris isolation technique.

DISCUSSION

The results of the present study showed that only two *Fusarium* species, *F. solani* and *F. oxysporum* were isolated from six forest soil samples. The findings were

similar to the study by Latiffah *et al.* (2009) who reported that *F. solani* and *F. oxysporum* were recovered from a forested area in Bird Valley, Universiti Sains Malaysia. Both species are considered cosmopolitan species and have been reported to occur in a wide range of climatic regions such as tropical, arid, Mediterranean regions and the Arctic (Kommendahl *et al.*, 1975; Kommendahl *et al.*, 1988; Marasas *et al.*, 1988; Sangalang *et al.*, 1995).

Generally, the diversity and occurrences of *Fusarium* species in uncultivated soils such as forest soil was low compared to cultivated soils. In a study by Lim and Chew (1970), *Fusarium* species was not recovered from soil samples from three nature reserve in Singapore. Similar result was also obtained by Lim (1974) in which *Fusarium* species was not recovered from natural field vegetation and forest soils.

Fusarium solani was the most common species isolated from the six soil samples. The species was also the most common species isolated from various soil types in Singapore and Peninsula Malaysia (Latiffah *et al.*, 2007; Nik Mohd Izham, 2008; Latiffah *et al.*, 2009). *F. solani* has a worldwide distribution, can be both soil-borne and airborne in the tropics but only soil borne in the temperate region (Burgess, 1981). Besides forest soils, *F. solani* can also be found in native soils such as in subtropical, semi-arid and grassland soils (Burgess and Summerell, 1992) and in desert soils (El-Gindy and Saad, 1990).

The occurrence of *F. oxysporum* isolates was low and was recovered only from one soil sample. *F. oxysporum* is widely distributed in both temperate and tropical region (Burgess, 1981) and one of the most common species isolated from uncultivated and cultivated soils (Onyike and Nelson, 1993; Mandeel and Abbas, 1994; Latiffah *et al.*, 2007; Mandeel, 2006; Nik Mohd Izham, 2008; Latiffah *et al.*, 2010a; 2010b). Although *F. oxysporum* was reported to be active over a wide range of environmental conditions (Burgess, 1981), the low occurrences of *F. oxysporum*, in forest soils could be due to the forest vegetation because *F. oxysporum* is a well-known plant pathogen and is associated with plant debris in cultivated soils whereby the species can survive as hypha in the plant debris (Burgess, 1981).

The six soil samples have loamy sand texture which contains a mixture of clay and sand with moderately coarse texture. Forest soils are acidic and contain less nutrient due to rapid decomposition and leaching of nutrients in the forest ecosystem (Cossalter and Pye-Smith, 2003). Microbial communities including species of

Fusarium are involved in the decomposition and nutrient cycling in the ecosystem.

The present study could contribute to the knowledge on the ecology and distribution of *Fusarium* species in the tropical soils. Several factor such as nutrient availability, environmental conditions and ability for competition could contribute to the distribution of *Fusarium* species in soils (Rodriguez-Molina *et al.*, 2000). Moreover, different plant species have been suggested to influence the diversity of soil-borne population of *F. oxysporum* (Edel *et al.*, 1997). Further study particularly covering larger areas of sampling would provide more information on the occurrences and distribution of *Fusarium* species in the tropical soils.

REFERENCES

Biondo, R. J. and Lee, J. S. (1997). Introduction to Plant and Soil Science Technology. Interstate Publishers Inc. Danville, IL, USA.

Brady, N. C. and Weil, R. R. (2008). The Nature and Properties of Soils. 14th edn. Prentice Hall Inc. NJ, USA.

Burgess, L. W. (1981). General ecology of the Fusaria. In: *Fusarium: Diseases, Biology and Taxonomy*. Nelson, P. E., Toussoun, T. A., Cook, R. J. (eds.). University Park, The Pennsylvania State University Press, PA, USA. pp. 225-235.

Burgess, L. W. and Summerell, B. A. (1992). Myogeography of *Fusarium*: Survey of *Fusarium* species in subtropical and semi-arid grassland soils from Queensland, Australia. *Mycological Research* **96**, 780-784.

Cossalter, C. and Pye-Smith, C. (2003). Fast-Wood Forestry: Myths and Realities. CIFOR Publication, Bogor, Indonesia.

Edel, V., Steinberg N., Gautheron, N. and Alabouvette, C. (1997). Populations of nonpathogenic *Fusarium* species associated with roots of four plant species compared to soilborne populations. *Phytopathology* **87**, 693-697.

El Gindy, A. A. and Saad, R. R. (1990). Fungi in virgin and cultivated soil of Salhiah desert Egypt. *Zentralblatt fur Mikrobiologie* **145**, 547-551.

Head, K. H. (1980). Manual of Soil Laboratory Testing. John Wiley & Sons Halsted Press, USA and Canada. pp. 232-238.

Kommendahl, T., Windels, C. E. and Lang, D. S. (1975). Comparisons of *Fusarium* populations in grasslands of Minnesota and Iceland. *Mycologia* **67**, 38-44.

Kommendahl, T., Abbas, H. K., Barnes, P. M. and Mirocha, C. J. (1988). Prevalance and toxicogenicity of *Fusarium* species form soils of Norway near the Arctic Circle. *Mycologia* **80**, 790-794.

Latiffah, Z., Mohd Zariman, M. and Baharuddin, S. (2007). Diversity of *Fusarium* species in cultivated soils in Penang. *Malaysian Journal of Microbiology* **3**(1), 27-30.

Latiffah, Z., Padzilah, M. I., Baharuddin, S. and Maziah, Z. (2009). *Fusarium* species in forest soil of Bird Valley. *Malaysian Journal of Microbiology* **5**(2), 132-133.

Latiffah, Z., Nurul Izzati, H. and Baharuddin, S. (2010a). *Fusarium* species isolated from peat soil of Pondok Tanjung and Sungai Beriah, Perak. *Malaysian Journal of Microbiology* **6**(1), 132-133.

Latiffah, Z., Mah, K. F., Heng, M. H., Maziah, Z. and Baharuddin, S. (2010b). *Fusarium* species isolated from mangrove soil in Kampung Pantai Aceh, Balik Pulau, Pulau Pinang. *Tropical Life Sciences Research* **21**(1), 23-32.

Leslie, J. F. and Summerell, B. A. (2006). The *Fusarium* Laboratory Manual. 1st edn. Blackwell Publishing, Ames, Iowa, USA.

Lim, G. (1974). Distribution of *Fusarium* in some British soils. *Mycopathologic et Mycologia allicata* **52**(3-4), 231-237.

Lim, G. and Chew, C. H. (1970). *Fusarium* in Singapore soils. *Plant and Soil* **33**, 673-677.

Mandeel, Q. A. and Abbas, J. A. (1994). Survey of *Fusarium* species in a arid environment of Bahrain. *Mycopathologia* **127**, 167-173.

Mandeel, Q. A. (2006). Biodiversity of the genus *Fusarium* in saline soil habitats. *Journal of Basic Microbiology* **46**(6), 480-494.

Marasas, W. F. O., Burgess, L. W., Anelich, R. Y., Lamprecht, S. C. and van Schalkwyk, D. J. (1988). Survey of *Fusarium* species associated with plant debris in South African soils. *South African Journal of Botany* **54**, 63-71.

Nelson, P. E., Toussoun, T. A. and Cook, R. J. (1983). *Fusarium* species. An Illustrated Manual for Identification. The Pennsylvania State University Press, University Park and London.

Nik Mohd Izham, N. M. (2008). Morphological characteristics, distribution and mycotoxin profiles of *Fusarium* species from soils in Peninsula Malaysia. M. Sc. thesis, Universiti Sains Malaysia.

Onyike, N. B. N. and Nelson, P. E. (1993). The distribution of *Fusarium* species in soils planted to millet and sorghum in Lesotho, Nigeria and Zimbabwe. *Mycopathologia* **121**, 105-114.

Rodriguez-Molina, M. C., Tello-Marquina, J. C., Torres-Vila, L. M. and Bielza-Lina, P. (2000). Micro-scale systematic sampling of soil: heterogeneity in populations of *Fusarium oxysporum*, *F. solani*, *F. roseum* and *F. moniliforme*. *Journal of Phytopathology* **148**, 609-614.

Sangalang, A. E., Burgess, L. W., Backhouse, D., Duff, J. and Wurst, H. (1995). Mycogeography of *Fusarium* species in soils from tropical, arid and Mediterranean regions of Australia. *Mycological Research* **99**(5), 523-528.

Summerell, B. A., Rugg, C. A. and Burgess, L. W. (1993). Mycogeography of *Fusarium*: Survey of *Fusarium* species associated with forest and woodland communities in north Queensland, Australia. *Mycological Research* **97**, 1015-1019.

Thein, S. J. (1979). A flow diagram for teaching texture by feel analysis. *Journal of Agronomic Education* **8**, 54-55.