

Plant_Pathology_Journal_2017. pdf *by*

Submission date: 10-Apr-2018 12:08PM (UTC+0800)

Submission ID: 944117014

File name: Plant_Pathology_Journal_2017.pdf (509.68K)

Word count: 4238

Character count: 24126



Plant Pathology Journal

ISSN 1812-5387

science
alert

ANSI*net*
an open access publisher
<http://ansinet.com>



Research Article

Morphological Characteristics and Mating Populations of *Fusarium* Species in *Gibberella fujikuroi* Species Complex (Gfsc) Associated with Stalk Rot Disease of Maize in Indonesia, Malaysia and Thailand¹Darnetty and ²Baharuddin Salleh¹Department of Plant Pests and Diseases, Faculty of Agriculture, Andalas University, 25163 Limau Manis, Padang, Indonesia²School of Biological Sciences, Universiti Sains Malaysia, 11800 Minden, Pulau Pinang, Malaysia

Abstract

Background: *Fusarium* stalk rot disease of maize is universally important because it is the most widespread destructive disease throughout the maize plantations all over the world including Southeast Asia. So far, the studies on the disease have not been carried out intensively in tropical countries including Indonesia, Malaysia and Thailand. **Objective:** The study was designed to determine the species and mating populations (MPs) of *Fusarium* in Gfsc associated with stalk rot disease in Indonesia, Malaysia and Thailand. **Materials and Methods:** A total of 106 strains of *Fusarium* in Gfsc were isolated from maize plants showing typical stalk rot symptoms and cultured on Potato Dextrose Agar (PDA) and carnation leaf pieces agar (CL A) for morphological identification. For MRs, the strains of *Fusarium* were crossed with 9 standard tester strains on Carrot Agar (CA). **Results:** Four species of *Fusarium* were morphologically identified as *E. verticillioides* (76%), *E. proliferatum* (38%), *E. subglutinans* (18%) and *E. konzium* (3%). Three mating populations were identified as MP-A, *Gibberella moniliformis* (71.7%), MP-D, *G. intermedia* (18.87%) and MP-E, *G. subglutinans* (2.83%) and 7 strains were not detected. All strains identified as MP-A, MP-D and MP-E were the strains morphologically identified as *F. verticillioides*, *F. proliferatum* and *F. subglutinans*, respectively. The MP-A (*F. verticillioides*) was the most dominant species associated with stalk rot disease of maize in this region. **Conclusion:** The results of biological identification and mating populations were corresponded to the results of morphological identification. This is the first report on the presence of MP-A, MP-D and MP-E on stalk rot-infected maize in Indonesia and Thailand, MP-A and MP-E in Malaysia. Additionally, the occurrence of *F. konzium* on stalk rot-infected maize plants are new records.

Key words: MP-A, MP-D, MP-E, *Gibberella moniliformis*, *G. intermedia*, *G. subglutinans*

Received: October 09, 2016

Accepted: November 10, 2016

Published: December 15, 2016

Citation: Darnetty and Baharuddin Salleh, 2017. Morphological characteristics and mating populations of *Fusarium* species in *Gibberella fujikuroi* species complex (Gfsc) associated with stalk rot disease of maize in Indonesia, Malaysia and Thailand. Plant Pathol. J., 16: 33-40.**Corresponding Author:** Darnetty, Department of Plant Pests and Diseases, Faculty of Agriculture, Andalas University, 25163 Limau Manis, Padang, Indonesia**Copyright:** © 2017 Darnetty and Baharuddin Salleh. This is an open access article distributed under the terms of the creative commons attribution license, which permits unrestricted use, distribution and reproduction in any medium, provided the original author and source are credited.**Competing interest:** The authors have declared that no competing interest exists.**Data Availability:** All relevant data are within the paper and its supporting information files.

INTRODUCTION

Fusarium stalk rot disease of maize is universally important because it is the most wide spread destructive disease throughout the maize plantations all over the world, including Southeast Asia^{1,2}. The disease not only reduces the quantity and quality of maize yield but also affects animal and human health because of mycotoxin production by *Fusarium*³⁻⁵. In developed countries such as the United States, researches on the stalk rot disease have been conducted intensively by various agricultural agencies. Unfortunately, the study on the disease has not been carried out intensively in the tropical countries, including Indonesia, Malaysia and Thailand. The distribution and prevalence of *Fusarium* species varies depending on geographical regions, environmental conditions and parts of the plant studied³.

The study on morphological characteristics of *Fusarium* species is very important for initial classification. For many species of *Fusarium*, morphological characteristics are the only ones that are well described and widely available⁶. Although, morphology of *Fusarium* species is highly variable, the identification for most of the species is easier if the cultures are grown on a consistent and appropriate media, culturing procedures and incubation conditions⁷.

Fusarium stalk rot is caused by *F. fujikuroi* species complex, formerly known as *F. moniliforme*^{3,6,7}. Some species in Gfsc are difficult to distinguish by using morphological characteristics e.g., between *F. fujikuroi* and *F. proliferatum* and between *F. sacchari* and *F. subglutinans*. Those species can be confidently differentiated by using biological species concept or phylogenetic concept⁶. The biological concept has been already widely used for identification of *Fusarium* in the Gfsc. Eleven mating populations or biological species, denoted by the letters A-K have been identified within the Gfsc⁸⁻¹³. These fungi are important pathogens of various crops in many world regions⁷.

The objectives of the study were to determine the species and mating populations (MPs) of *Fusarium* in the Gfsc isolated from maize plants showing typical stalk rot symptoms in Indonesia, Malaysia and Thailand.

MATERIALS AND METHODS

Fusarium strains: A total of 106 strains of *Fusarium* species that belong to Gfsc were isolated from maize plants showing typical stalk rot symptoms in Indonesia, Malaysia and Thailand by using the semi-selective medium (peptone pentachloronitrobenzene agar, PPA)¹⁴. The pure cultures were obtained by single-spore method.

Morphological characteristics: The *Fusarium* strains were cultured on Potato Dextrose Agar (PDA)⁷ to observe the macroscopic characteristics i.e., colony appearance, pigmentations and growth rates. Colony appearance and pigmentations were observed after 7 days incubation while growth rates after 72 h incubation. To observe microscopic characteristics, the *Fusarium* strains were transferred onto CLA¹⁵ and incubated for 7-15 days under the standard growth conditions. Microscopic characteristics observed were macroconidia, microconidia, chlamydozoospores and the mode of microconidial formations by *in situ* observation. The morphological characteristics were observed under a light microscope and photographed.

Mating Population (MP): Before the crosses were carried out mating type alleles (*MAT-1* and *MAT-2*) of the strains were identified based on PCR amplification by using the primers *MAT-1* (Gfmat1a (5'-GTTTCATCAAAGGGCAAGCG-3') and Gfmat 1b (5'-TAAGCGCCCTCTTAACGCCTTC-3') and *MAT-2* Gfmat 2c (5'-GCTTCATTATTCGATCAAG-3') and Gmat 2d (5'-CTACGTTGAGAGCTGTACAG-3'). By identifying the mating type of the strains, the number of crosses can be reduced by half, because the unidentified strains need to be crossed only with the tester of each species that is of the opposite mating type.

Crosses were made on Carrot Agar (CA)¹⁶ following protocol of Klittich and Leslie¹⁷. The standard mating population testers strains (MP-A to MP-I) were obtained from stock of the *Fusarium* Culture Collection Section, School of Biological Sciences, Universiti Sains Malaysia. On the same day the strains serving as male parents were grown on complete medium (CM)¹⁸ and testers serving as female parents on CA for 7 days. After 1 week of incubation, 1 mL of spore suspension of strains were spread on CA containing tester by using a bent glass rod and incubated for 3-6 weeks at 26±1°C with cool-white and near-UV fluorescent tubes (approximately 1,900 lux). Then the formations of perithecia were inspected for all crosses. Crosses were scored as fertile if the mature perithecia were observed.

RESULTS AND DISCUSSION

Characteristics of *Fusarium* species: Characteristics of *F. verticillioides* and *F. proliferatum* almost similar, they formed white floccose mycelium which may become grayish violet or grayish magenta with age. Pigmentation in agar varied, ranging from no pigmentation or grayish orange to violet grey, dark violet or dark magenta (almost black). Macroconidia were long, slender, falcate to almost straight, foot-shape basal cell, slightly curved apical cell, usually

3-5 septate for *F. verticillioides* and 3-4 septate for *F. proliferatum*. Both of them produced single cell clavate microconidia in false heads and in long chains. Although they were similar, but they were different in production of microconidia in which *F. verticillioides* produced microconidia from monophialides and *F. proliferatum* from monophialides and polyphialides. Besides that, *F. verticillioides* produced swollen cells while *F. proliferatum* did not produce swollen cells. Different from *F. verticillioides* and *F. proliferatum*, *F. subglutinans* formed oval, elliptical microconidia only in false heads. The specific character of *F. konzum* was the

production of microconidia on small false heads, but not on chains. The characteristics of *Fusarium* species mentioned in accordance with the proposed by Burgess *et al.*⁷ and Leslie and Summerell⁶.

Based on morphological characteristics, 106 of *Fusarium* strains obtained from maize showing typical stalk rot were identified as *Gibberella fujikuroi* species complex that consisted of 4 species i.e., *F. verticillioides* (79 strains, 74.53%), *F. proliferatum* (21 strains, 19.81%), *F. subglutinans* (4 strains, 3.77%) and *F. konzum* (2 strains, 1.89%) as shown in Table 1. The three species of *Fusarium* i.e., *F. verticillioides*,

Table 1: Species, mating type and mating population of *Fusarium* strains

Strains	Locations	<i>Fusarium</i> species	Mating type	Mating population
P20920	Pematang 3 Ringgit, Penang, Malaysia	<i>F. verticillioides</i>	MAT-1	A
P20930	Pematang 3 Ringgit, Penang, Malaysia	<i>F. verticillioides</i>	ND	A
P20940	Pematang 3 Ringgit, Penang, Malaysia	<i>F. verticillioides</i>	MAT-1	-
K24040	Gurun Kedah, Kedah, Malaysia	<i>F. verticillioides</i>	MAT-1	A
K24050	Gurun Kedah, Kedah, Malaysia	<i>F. verticillioides</i>	MAT-1	A
K24060	Gurun Kedah, Kedah, Malaysia	<i>F. verticillioides</i>	MAT-1	A
K24070	Gurun Kedah, Kedah, Malaysia	<i>F. verticillioides</i>	MAT-1	A
K24080	Gurun Kedah, Kedah, Malaysia	<i>F. verticillioides</i>	MAT-2	A
K24190	Gurun Kedah, Kedah, Malaysia	<i>F. verticillioides</i>	MAT-1	A
HLN01520	Buri, Thailand	<i>F. verticillioides</i>	MAT-1	A
HLN01550	Tak Fa, Thailand	<i>F. verticillioides</i>	MAT-1	A
HLN01590	Ban Wang Chao, Thailand	<i>F. verticillioides</i>	MAT-1	A
HLN01630	Mae Phrik, Thailand	<i>F. verticillioides</i>	MAT-1	A
HLN01650	Tron, Thailand	<i>F. verticillioides</i>	MAT-1	A
HLN01660	Tron, Thailand	<i>F. verticillioides</i>	MAT-2	A
HLN01680	Nam Nao, Thailand	<i>F. verticillioides</i>	MAT-1	A
HLN01700	Thailand	<i>F. verticillioides</i>	MAT-1	A
HLN01740	Ban Nong Bua Koke, Thailand	<i>F. verticillioides</i>	MAT-1	A
HLN01750	Ban Nong Bua Koke, Thailand	<i>F. verticillioides</i>	MAT-1	A
HLN01760	Ban Nong Bua Koke, Thailand	<i>F. verticillioides</i>	MAT-1	A
HLN01780	Saphan Bari, Thailand	<i>F. verticillioides</i>	MAT-1	A
HLN01790	Saphan Bari, Thailand	<i>F. verticillioides</i>	MAT-1	A
HLN01820	Prince of Songkhla University, Hat Yai, Thailand	<i>F. verticillioides</i>	MAT-1	A
HLN01830	Prince of Songkhla University, Hat Yai, Thailand	<i>F. verticillioides</i>	MAT-1	A
OLN01910	Sungai, Baringin, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN01940	Sungai, Baringin, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	-
OLN02120	Sitiung, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02150	50 Kota, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02160	Sitiung, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02170	Sitiung, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	A
OLN02180	Sitiung, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02300	Medan, North Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02310	Medan, North Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02320	Medan, North Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02340	Medan, North Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	A
OLN02350	Medan, North Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02370	Baso, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02390	Baso, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02440	Lunang, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	A
OLN02450	Lunang, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02460	Lunang, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02470	Lunang, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02480	Lunang, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02490	Lunang, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN02600	Panampuang, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	A

Table 1: Continue

Strains	Locations	<i>Fusarium</i> species	Mating type	Mating population
OLN0261O	Panampuang, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0262O	Panampuang, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0270O	Alahan Panjang, Solok, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0274O	Pariaman, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0275O	Pariaman, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0276O	Pariaman, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0288O	Sungai, Baringin, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	A
OLN0325O	Kaju Bajak, Padang, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0326O	Kaju Bajak, Padang, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0327O	Kaju Bajak, Padang, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0329O	Kaju Bajak, Padang, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	A
OLN0330O	Kaju Bajak, Padang, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0331O	Kaju Bajak, Padang, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	A
OLN0332O	Kaju Bajak, Padang, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0333O	Kaju Bajak, Padang, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	A
OLN0334O	Kaju Bajak, Padang, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	A
OLN0335O	Kaju Bajak, Padang, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0336O	Kelok Kuranji, Padang, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	A
OLN0337O	Kelok Kuranji, Padang, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	A
OLN0341O	Sungai Buluh, Padang-Pariaman, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0344O	Sungai Buluh, Padang-Pariaman, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	A
OLN0345O	Sungai Buluh, Padang-Pariaman, West Sumatra, Indonesia	<i>F. verticillioides</i>	ND	-
OLN0346O	Talang Julo, Padang-Pariaman, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0349O	Sungai Baringin, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0380O	Banda Aceh, Aceh, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0386O	Bukittinggi, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0486O	Talang Julo Padang-Pariaman, West Sumatra Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0488O	Koto Tuo, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0493O	Labuang, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
HLN0494O	Tak Fa, Thailand	<i>F. verticillioides</i>	MAT-1	A
OLN0495O	Labuang, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	ND	-
OLN0497O	Sungai, Baringin, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0499O	Sungai, Baringin, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-1	A
OLN0500O	Sungai, Baringin, Agam, West Sumatra, Indonesia	<i>F. verticillioides</i>	MAT-2	A
HLN0151O	Buri, Thailand	<i>F. proliferatum</i>	MAT-2	D
HLN0153O	Takhli, Thailand	<i>F. proliferatum</i>	MAT-1	D
HLN0156O	Ban Wang Chao, Thailand	<i>F. proliferatum</i>	MAT-1	D
HLN0160O	Tak, Thailand	<i>F. proliferatum</i>	ND	D
HLN0172O	Khon Kaen, Thailand	<i>F. proliferatum</i>	MAT-1	D
HLN0181O	Tak Fa, Thailand	<i>F. proliferatum</i>	MAT-1	D
OLN0238O	Baso, Agam, West Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-2	D
OLN0240O	Baso, Agam, West Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-2	D
OLN0241O	Baso, Agam, West Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-2	D
OLN0243O	Baso, Agam, West Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-2	D
OLN0273O	Alahan Panjang, Solok, West Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-1	D
OLN0291O	Kelok Kuranji, Padang, West Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-1	D
OLN0279O	Parapat, North Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-1	D
OLN0342O	Kelok Kuranji, Padang, West Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-1	D
OLN0348O	Talang Julo, Padang Pariaman, West Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-1	D
OLN0350O	Talang Julo, Padang-Pariaman, West Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-1	D
OLN0381O	Banda Aceh, Aceh, Indonesia	<i>F. proliferatum</i>	MAT-1	D
OLN0452O	Tomok, Medan, North Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-1	D
OLN0454O	Tomok, Medan, North Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-1	D
OLN0467O	Tomok, Medan, North Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-1	D
OLN0491O	Labuang, Agam, West Sumatra, Indonesia	<i>F. proliferatum</i>	MAT-1	D
S4895O	Ranau, Sabah, Malaysia	<i>F. subglutinans</i>	MAT-2	E
OLN0140O	Bukittinggi, West Sumatra, Indonesia	<i>F. subglutinans</i>	MAT-1	E
HLN0154O	Tak Fa, Thailand,	<i>F. subglutinans</i>	MAT-1	-
OLN0340O	Kelok Kuranji, Padang, West Sumatra, Indonesia	<i>F. subglutinans</i>	MAT-1	E
OLN0343O	Sungai Buluh, Padang Pariaman, West Sumatra, Indonesia	<i>F. konzum</i>	MAT-2	-
OLN0492O	Labuang, Agam, West Sumatra, Indonesia	<i>F. konzum</i>	MAT-1	-

ND: Not detected, -: Infertile, *Fusarium* strains were recovered from maize plants showing typical talk rot symptom in Indonesia, Malaysia and Thailand

F. proliferatum and *F. subglutinans* have been reported as causal agents of stalk rot disease of maize^{6,12,19,20}. *Fusarium verticillioides* and *F. proliferatum* were well distributed in Indonesia, Malaysia and Thailand and also the predominant species. This is in accordance with what is stated by Leslie¹⁰ that these species grow well in hotter regions. Although *F. subglutinans* was reported as a main causal agent of stalk rot disease of maize, it was recovered in small numbers. It occurred because these species grow well in cooler regions^{16,6} meanwhile Indonesia, Malaysia and Thailand are hotter regions. The occurrence of *F. konzum* on stalk rot-infected maize plants are new records.

Mating Populations (MPs): By using the primers *MAT-1* and *MAT-2*, the mating type of the strains were identified as shown in Table 1. The PCR reactions specifically amplified either an ~800 bp fragment from *MAT-2* isolates or an ~200 bp fragment from *MAT-1* (Fig. 1, 2). Not all strains tested were

detected their mating types. Out of the 106 strains tested, only 102 (96.23%) were identified as either *MAT-1* or *MAT-2*. Four strains (3.77%) were not detected their mating types. The number of strains of *F. verticillioides*, *F. proliferatum*, *F. subglutinans* and *F. konzum* identified their mating types were 76 (96.2%), 20 (95.24%), 4 (100%) and 0 (0%), respectively. The mating type (*MAT-1*) was predominant over the mating type (*MAT-2*) in 79:23 ratios. The ratio of *MAT-1* and *MAT-2* of *F. verticillioides*, *F. proliferatum*, *F. subglutinans* and *F. konzum* were 61:15, 15:5, 3:1 and 1:1, respectively (Table 1). This is in conformity with those obtained by Kovacevic *et al.*²¹, Mansuetus *et al.*²² and Sabet *et al.*²³ who stated that the ratios of *MAT-1* and *MAT-2* of *F. verticillioides* were 14:6, 97:18 and 59:17, respectively.

Only 99 (93.4%) strains of *Fusarium* were crossed-fertile with standard mating population tester's strains. Seventy six strains (71.7%) were identified as MP-A (*G. moniliformis*), 20 strains (18.87%) as MP-D (*G. intermedia*) and 3 strains

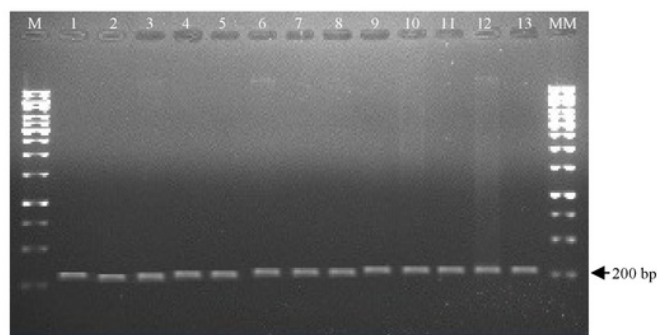


Fig. 1: *MAT-1* of some strains of *Fusarium* species in Gfsc. M: Marker, 1: K2404O, 2: K2405O, 3: K2406O, 4: K2407O, 5: K2408O, 6: K2419O, 7: HLN0152O, 8: HLN0155O, 9: HLN0159O, 10: HLN0163O, 11: HLN0163O, 12: HLN0165O and 13: HLN0166O

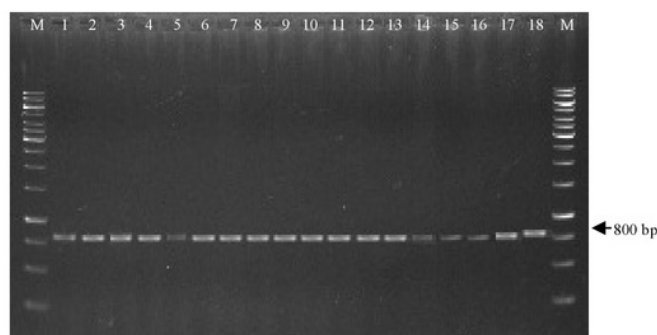


Fig. 2: *MAT-2* of some strains of *Fusarium* species in Gfsc. M: Marker, 1: K2408O, 2: OLN0184O, 3: OLN0199O, 4: OLN0203O, 5: OLN0205O, 6: D2424O, 7: OLN0219O, 8: OLN0240O, 9: OLN0260O and OLN0280O, 10: S4867O, 11: S4895O, 12: OLN0302O, 13: OLN0329O, 14: OLN0331O, 15: OLN0332O, 16: OLN0336O, 17: OLN0337O and 18: OLN0344O

(2.83%) as MP-E (*G. subglutinans*). Out of 79 species of *F. verticillioides*, 21 species of *F. proliferatum*, 4 species of *F. subglutinans* and 2 species of *F. konzum* morphologically identified, 76 were identified as MP-A, 20 as MP-D, 3 as MP-E and 2 not detected respectively. The results of biological identification, mating populations were corresponded to the results of morphological identification. Some species of *Fusarium* i.e., *F. verticillioides* (3 strains), *F. proliferatum* (1 strains), *F. subglutinans* (1 strain) and *F. konzum* (2 strains)

did not produce perithecia after crossing with mating population testers because these strains were probably sterile. For these species, it is possible to use the other mating population testers or by doing molecular approaches. The MP-A, MP-D and MP-E are shown in Fig. 3-5, respectively. The MP-A (*G. moniliformis*) was the most dominant species associated with stalk rot disease of maize in this region. According to Moretti *et al.*⁵, the most frequently found on maize were *F. moniliforme* (MP-A), *F. proliferatum*

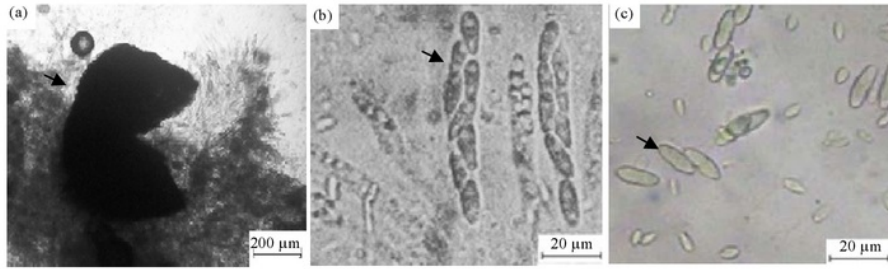


Fig. 3(a-c): Perithecia and ascospores of *Gibberella moniliformis* (MP-A) produced by crossing OLN03800 strain with MP-A tester, (a) Perithecia on carrot agar *in situ* observation, (b) Ascospores in ascus and (c) Ascospores

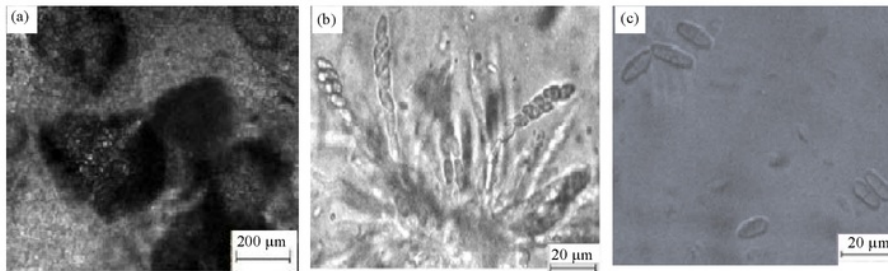


Fig. 4(a-c): Perithecia and ascospores of *Gibberella intermedia* (MP-D) produced by crossing OLN02790 strain with MP-D tester, (a) Perithecia on carrot agar *in situ* observation, (b) Ascospores in ascus and (c) Ascospores

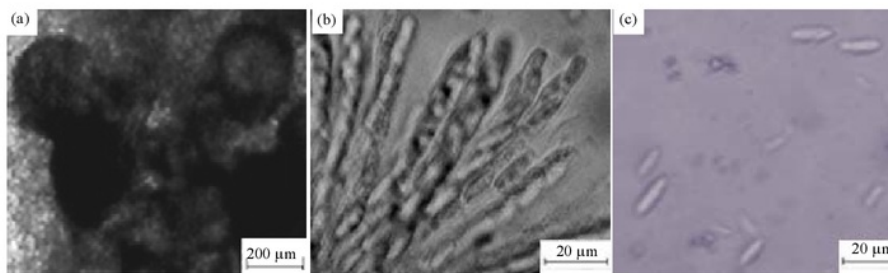


Fig. 5(a-c): Perithecia and ascospores of *Gibberella subglutinans* (MP-E) produced by crossing OLN01400 strain with MP-E tester, (a) Perithecia on carrot agar *in situ* observation, (b) Ascospores in ascus and (c) Ascospores

(MP-D) and *F. subglutinans* (MP-E), which were also differentiated by their toxigenic capability. This is the first report on the presence of MP-A, MP-D and MP-E on stalk rot-infected maize in Indonesia and Thailand and MP-A and MP-E in Malaysia.

CONCLUSION

Four species of *Fusarium* were morphologically identified as *F. verticillioides* (79 strains, 74.53%), *F. proliferatum* (21 strains, 19.81%), *F. subglutinans* (4 strains, 3.77%) and *F. konzum* (2 strains, 1.89%). Only 102 strains (96.23%) were identified as either *MAT-1* or *MAT-2*. The mating type (*MAT-1*) was predominant over the mating type (*MAT-2*) in 79:23 ratios. In crosses with 9 standard testers, three mating populations were identified as MP-A, *G. moniliformis* (76 strains, 71.7%), MP-D, *G. intermedia* (20 strains, 18.87%) and MP-E, *G. subglutinans* (3 strains, 2.83%). The MP-A (*F. verticillioides*) was the most dominant species associated with stalk rot disease of maize in this region. The results of biological identification, mating populations were corresponded to the results of morphological identification. This is the first study on the presence of MP-A, MP-D and MP-E on stalk rot-infected maize in Indonesia and Thailand, MP-A and MP-E in Malaysia. Additionally, the occurrence of *F. konzum* on stalk rot-infected maize plants are new records.

REFERENCES

1. Afolabi, C.G., P.S. Ojiambo, E.J.A. Ekpo, A. Menkir and R. Bandyopadhyay, 2008. Novel sources of resistance to *Fusarium* stalk rot of maize in tropical Africa. *Plant Dis.*, 92: 772-780.
2. White, D.G., 1999. Compendium of Maize Disease. 3rd Edn., APS Press, USA., ISBN: 9780890542347, Pages: 78.
3. Bottalico, A., 1998. *Fusarium* diseases of cereals: Species complex and related mycotoxin profiles, in Europe. *J. Plant Pathol.*, 80: 85-103.
4. Logrieco, A., G. Mule, A. Moretti and A. Bottalico, 2002. Toxigenic *Fusarium* species and mycotoxins associated with maize ear rot in Europe. *Eur. J. Plant Pathol.*, 108: 597-609.
5. Moretti, A., G.A. Bennett, A. Logrieco, A. Bottalico and M.N. Beremand, 1995. Fertility of *Fusarium moniliforme* from maize and sorghum related to fumonisin production in Italy. *Mycopathologia*, 131: 25-29.
6. Leslie, J.F. and B.A. Summerell, 2006. The *Fusarium* Laboratory Manual. Blackwell Publishing, Ames, Iowa, USA.
7. Burgess, L.W., B.A. Summerell, S. Bullock, K.P. Gott and D. Bckhouse, 1994. Laboratory Manual for *Fusarium* Research. 3rd Edn., Royal Botanic Gardens, Sydney, Australia.
8. Britz, H., E.T., Steenkamp, T.A. Coutinho, B.D. Wingfield, W.F. Marasas and M.J. Wingfield, 2002. Two new species of *Fusarium* section *Liseola* associated with mango malformation. *Mycologia*, 94: 722-730.
9. Klittich, C.J.R. and J.F. Leslie, 1992. Identification of a second mating population within the *Fusarium moniliforme* anamorph of *Gibberella fujikuroi*. *Mycologia*, 84: 541-547.
10. Leslie, J.F., 1991. Mating populations in *Gibberella fujikuroi* (*Fusarium* section *Liseola*). *Phytopathology*, 81: 1058-1060.
11. Moretti, A.N., 2009. Taxonomy of *Fusarium* genus: A continuous fight between lumpers and splitters. *Proc. Nat. Sci. Matica Srpska Novi Sad*, 117: 7-13.
12. Visentin, I., D. Valentino, F. Cardinale and G. Tamiatti, 2010. DNA-Base Tools for the Detection of *Fusarium* spp. Pathogenic on Maize. In: Molecular Identification of Fungi, Gherbawy, Y. and K. Voigt (Eds.). Springer Science and Business Media, USA., pp: 106-129.
13. Zeller, K.A., B.A. Summerell, S. Bullock and J.F. Leslie, 2003. *Gibberella konza* (*Fusarium konzum*) sp. nov. from prairie grasses, a new species in the *Gibberella fujikuroi* species complex. *Mycologia*, 95: 943-954.
14. Nelson, P.E., T.A. Toussoun and W.F.O. Marasas, 1983. *Fusarium* Species: An Illustrated Manual for Identification. 1st Edn., Pennsylvania State University Press, University Park, University Park, PA., USA., ISBN-13: 978-0271003498, Pages: 226.
15. Fisher, N.L., L.W. Burgess, T.A. Toussoun and P.E. Nelson, 1982. Carnation leaves as a substrate and for preserving cultures of *Fusarium* species. *Phytopathology*, 72: 151-153.
16. Goertz, A., S. Zuehlke, M. Spiteller, U. Steiner and H.W. Dehne *et al.*, 2010. *Fusarium* species and mycotoxin profiles on commercial maize hybrids in Germany. *Eur. J. Plant Pathol.*, 128: 101-111.
17. Klittich, C. and J.F. Leslie, 1988. Nitrate reduction mutants of *Fusarium moniliforme* (*Gibberella fujikuroi*). *Genetics*, 118: 417-423.
18. Correll, J., C.J.R. Klittich and J.F. Leslie, 1987. Nitrate non-utilizing mutants of *Fusarium oxysporum* and their use in vegetative compatibility tests. *Phytopathology*, 77: 1640-1646.
19. Levic, J., S. Stankovic, V. Krnjaja, A. Bokarov-Stancic and D. Ivanovic, 2012. Distribution frequency and incidence of seed-borne pathogens of some cereals and industrial crops in serbia crops in Serbia. *Pestic. Phytomed. (Belgrade)*, 27: 33-40.

20. Steenkamp, E.T., B.D. Wingfield, T.A. Coutinho, K.A. Zeller, M.J. Wingfield, W.F. Marasas and J.F. Leslie, 2000. PCR-based identification of MAT-1 and MAT-2 in the *Gibberella fujikuroi* species complex. *Applied Environ. Microbiol.*, 66: 4378-4382.
21. Kovacevic, T., J. Levic, S. Stankovic and J. Vukojevic, 2013. Mating populations of *Gibberella fujikuroi* (Sawada) S. Ito species complex isolating from maize, sorghum and wheat in Serbia. *Genetika*, 45: 749-760.
22. Mansuetus, A.S., G.N. Odvody, R.A. Frederiksen and J.F. Leslie, 1997. Biological species in the *Gibberella fujikuroi* species complex (*Fusarium* section *Liseola*) recovered from sorghum in Tanzania. *Mycol. Res.*, 101: 815-820.
23. Sabet, K.K., A.M.A. Ashour, E.M. El-Assiuty and E.M. El-Shabrawy, 2006. Mating populations and effective population number in *Gibberella fujikuroi* species complex of rotted maize ears under Egyptian conditions. *Egypt. J. Phytopathol.*, 34: 29-41.

ORIGINALITY REPORT

7 %	%	7 %	%
SIMILARITY INDEX	INTERNET SOURCES	PUBLICATIONS	STUDENT PAPERS

PRIMARY SOURCES

1 Nur Ain Izzati, M. Z., Azmi, A. R., Baharuddin, S.. "Secondary metabolite profiles and mating populations of *Fusarium* species in section *Liseola* associated with bakanae disease of rice", *Malaysian Journal of Microbiology*, 2008 **4%**
Publication

2 Ednar G. Wulff. "*Fusarium* spp. associated with rice Bakanae: ecology, genetic diversity, pathogenicity and toxigenicity", *Environmental Microbiology*, 03/2010 **3%**
Publication

Exclude quotes On
Exclude bibliography On

Exclude matches < 3%