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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 Thailand

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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 (CLA) for morphological identification. For MPs, the strains of *Fusarium* were crossed with 9 standard tester strains on Carrot Agar (CA). **Results:** Four species of *Fusarium* were morphologically identified as *F. verticillioides* (75%), *F. proliferatum* (20%), *F. subglutinans* (4%) and *F. konzum* (2%). 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. konzum* on stalk rot-infected maize plants are new records.

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

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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 studied[3].

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 available[6]. 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 conditions[7].

Fusarium stalk rot is caused by *F. fujikuroi* species complex, formerly known as *F. moniliforme*[2,3,6,7]. 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 concept[6]. 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 Gfsc[8-13]. These fungi are important pathogens of various crops in many world regions[7].

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 CLA[15] and incubated for 7-15 days under the standard growth conditions. Microscopic characteristics observed were macroconidia, microconidia, chlamydospores 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'-GTTCATCAAAGGGCAAGCG-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 Leslie[17]. 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 $26\pm1°C$ 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 Summerell[6].

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	*Fusarium* species	Mating type	Mating population
P2092O	Pematang 3 Ringgit, Penang, Malaysia	*F. verticillioides*	*MAT-1*	A
P2093O	Pematang 3 Ringgit, Penang, Malaysia	*F. verticillioides*	ND	A
P2094O	Pematang 3 Ringgit, Penang, Malaysia	*F. verticillioides*	*MAT-1*	-
K2404O	Gurun Kedah, Kedah, Malaysia	*F. verticillioides*	*MAT-1*	A
K2405O	Gurun Kedah, Kedah, Malaysia	*F. verticillioides*	*MAT-1*	A
K2406O	Gurun Kedah, Kedah, Malaysia	*F. verticillioides*	*MAT-1*	A
K2407O	Gurun Kedah, Kedah, Malaysia	*F. verticillioides*	*MAT-1*	A
K2408O	Gurun Kedah, Kedah, Malaysia	*F. verticillioides*	*MAT-2*	A
K2419O	Gurun Kedah, Kedah, Malaysia	*F. verticillioides*	*MAT-1*	A
HLN0152O	Buri, Thailand	*F. verticillioides*	*MAT-1*	A
HLN0155O	Tak Fa, Thailand	*F. verticillioides*	*MAT-1*	A
HLN0159O	Ban Wang Chao, Thailand	*F. verticillioides*	*MAT-1*	A
HLN0163O	Mae Phrik, Thailand	*F. verticillioides*	*MAT-1*	A
HLN0165O	Tron, Thailand	*F. verticillioides*	*MAT-1*	A
HLN0166O	Tron, Thailand	*F. verticillioides*	*MAT-2*	A
HLN0168O	Nam Nao, Thailand	*F. verticillioides*	*MAT-1*	A
HLN0170O	Thailand	*F. verticillioides*	*MAT-1*	A
HLN0174O	Ban Nong Bua Koke, Thailand	*F. verticillioides*	*MAT-1*	A
HLN0175O	Ban Nong Bua Koke, Thailand	*F. verticillioides*	*MAT-1*	A
HLN0176O	Ban Nong Bua Koke, Thailand	*F. verticillioides*	*MAT-1*	A
HLN0178O	Saphan Bari,Thailand	*F. verticillioides*	*MAT-1*	A
HLN0179O	Saphan Bari, Thailand	*F. verticillioides*	*MAT-1*	A
HLN0182O	Prince of Songkhla University, Hat Yai, Thailand	*F. verticillioides*	*MAT-1*	A
HLN0183O	Prince of Songkhla University, Hat Yai, Thailand	*F. verticillioides*	*MAT-1*	A
OLN0191O	Sungai, Baringin, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0194O	Sungai, Baringin, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	-
OLN0212O	Sitiung, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0215O	50 Kota, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0216O	Sitiung, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0217O	Sitiung, West Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	A
OLN0218O	Sitiung, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0230O	Medan, North Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0231O	Medan, North Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0232O	Medan, North Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0234O	Medan, North Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	A
OLN0235O	Medan, North Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0237O	Baso, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0239O	Baso, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0244O	Lunang, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	A
OLN0245O	Lunang, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0246O	Lunang, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0247O	Lunang, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0248O	Lunang, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0249O	Lunang, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0260O	Panampuang, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	A

Table 1: Continue

Strains	Locations	*Fusarium* species	Mating type	Mating population
OLN0261O	Panampuang, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0262O	Panampuang, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0270O	Alahan Panjang, Solok, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0274O	Pariaman, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0275O	Pariaman, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0276O	Pariaman, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0288O	Sungai, Baringin, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	A
OLN0325O	Kaju Bajak, Padang, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0326O	Kaju Bajak, Padang, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0327O	Kaju Bajak, Padang, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0329O	Kaju Bajak, Padang, West Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	A
OLN0330O	Kaju Bajak, Padang, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0331O	Kaju Bajak, Padang, West Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	A
OLN0332O	Kaju Bajak, Padang, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0333O	Kaju Bajak, Padang, West Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	A
OLN0334O	Kaju Bajak, Padang, West Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	A
OLN0335O	Kaju Bajak, Padang, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0336O	Kelok Kuranji, Padang,West Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	A
OLN0337O	Kelok Kuranji, Padang, West Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	A
OLN0341O	Sungai Buluh, Padang-Pariaman, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0344O	Sungai Buluh, Padang-Pariaman, West Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	A
OLN0345O	Sungai Buluh, Padang-Pariaman, West Sumatra, Indonesia	*F. verticillioides*	ND	-
OLN0346O	Talang Julo, Padang-Pariaman,West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0349O	Sungai Baringin, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0380O	Banda Aceh, Aceh, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0386O	Bukittinggi, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0486O	Talang Julo Padang-Pariaman, West Sumatra Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0488O	Koto Tuo, Agam, West Sumatra ,Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0493O	Labuang, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
HLN0494O	Tak Fa, Thailand	*F. verticillioides*	*MAT-1*	A
OLN0495O	Labuang, Agam, West Sumatra, Indonesia	*F. verticillioides*	ND	-
OLN0497O	Sungai, Baringin, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0499O	Sungai, Baringin, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-1*	A
OLN0500O	Sungai, Baringin, Agam, West Sumatra, Indonesia	*F. verticillioides*	*MAT-2*	A
HLN0151O	Buri, Thailand	*F. proliferatum*	*MAT-2*	D
HLN0153O	Takhli, Thailand	*F. proliferatum*	*MAT-1*	D
HLN0156O	Ban Wang Chao, Thailand	*F. proliferatum*	*MAT-1*	D
HLN0160O	Tak, Thailand	*F. proliferatum*	ND	D
HLN0172O	Khon Kaen, Thailand	*F. proliferatum*	*MAT-1*	D
HLN0181O	Tak Fa, Thailand	*F. proliferatum*	*MAT-1*	D
OLN0238O	Baso, Agam, West Sumatra, Indonesia	*F. proliferatum*	*MAT-2*	D
OLN0240O	Baso, Agam, West Sumatra, Indonesia	*F. proliferatum*	*MAT-2*	D
OLN0241O	Baso, Agam, West Sumatra, Indonesia	*F. proliferatum*	*MAT-2*	D
OLN0243O	Baso, Agam, West Sumatra, Indonesia	*F. proliferatum*	*MAT-2*	D
OLN0273O	Alahan Panjang, Solok, West Sumatra, Indonesia	*F. proliferatum*	*MAT-1*	D
OLN0291O	Kelok Kuranji, Padang, West Sumatra, Indonesia	*F. proliferatum*	*MAT-1*	D
OLN0279O	Parapat, North Sumatra, Indonesia	*F. proliferatum*	*MAT-1*	D
OLN0342O	Kelok Kuranji, Padang, West Sumatra, Indonesia	*F. proliferatum*	*MAT-1*	D
OLN0348O	Talang Julo, Padang Pariaman, West Sumatra, Indonesia	*F. proliferatum*	*MAT-1*	D
OLN0350O	Talang Julo, Padang-Pariaman, West Sumatra, Indonesia	*F. proliferatum*	*MAT-1*	D
OLN0381O	Banda Aceh, Aceh, Indonesia	*F. proliferatum*	*MAT-1*	D
OLN0452O	Tomok, Medan, North Sumatra, Indonesia	*F. proliferatum*	*MAT-1*	D
OLN0454O	Tomok, Medan, North Sumatra, Indonesia	*F. proliferatum*	*MAT-1*	D
OLN0467O	Tomok, Medan, North Sumatra, Indonesia	*F. proliferatum*	*MAT-1*	D
OLN0491O	Labuang, Agam, West Sumatra, Indonesia	*F. proliferatum*	*MAT-1*	D
S4895O	Ranau, Sabah, Malaysia	*F. subglutinans*	*MAT-2*	E
OLN0140O	Bukittinggi, West Sumatra, Indonesia	*F. subglutinans*	*MAT-1*	E
HLN0154O	Tak Fa, Thailand,	*F. subglutinans*	*MAT-1*	-
OLN0340O	Kelok Kuranji, Padang, West Sumatra, Indonesia	*F. subglutinans*	*MAT-1*	E
OLN0343O	Sungai Buluh, Padang Pariaman, West Sumatra, Indonesia	*F. konzum*	*MAT-2*	-
OLN0492O	Labuang, Agam, West Sumatra, Indonesia	*F. konzum*	*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 verticilliiodes* 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 Leslie[10] 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 (3.76%) 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

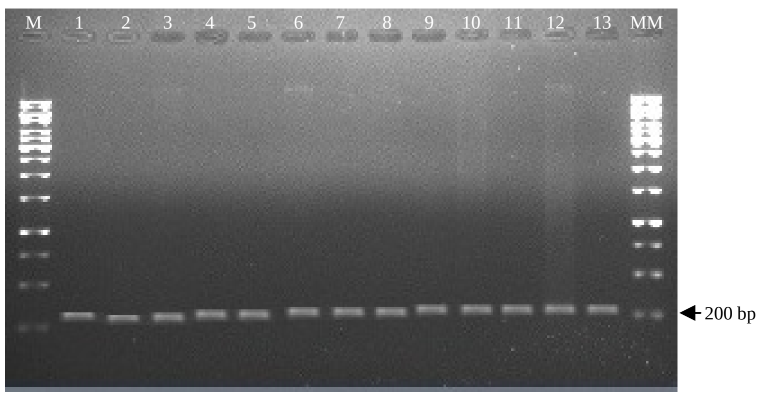

Fig. 1: *MAT-1* of some strains of *Fusarium* species in Gfsc. M: Marker, 1: K2404O, 2: K2405O, 3: K2406O, 4: K2407O, 5: K2408O, 6: K2419O, 7: HLN0152O, 8: HLN0155O, 9: HLN0159O, 10: HLN0163O, 11: HLN0163O, 12: HLN0165O and 13: HLN0166O

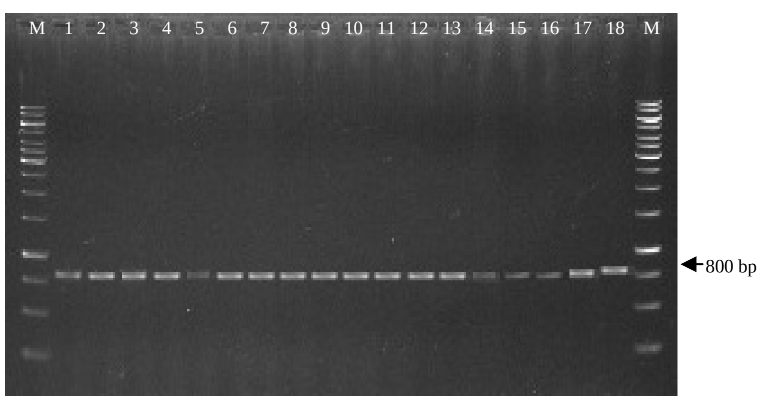

Fig. 2: *MAT-2* of some strains of *Fusarium* species in Gfsc. M: Marker, 1: K2408O, 2: OLN0184O, 3: OLN0199O, 4: OLN0203O, 5: OLN0205O, 6: D2424O, 7: OLN0219O, 8: OLN0240O, 9: OLN0260O and OLN0280O, 10: S4867O, 11: S4895O, 12: OLN0302O, 13: OLN0329O, 14: OLN0331O, 15: OLN0332O, 16: OLN0336O, 17: OLN0337O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

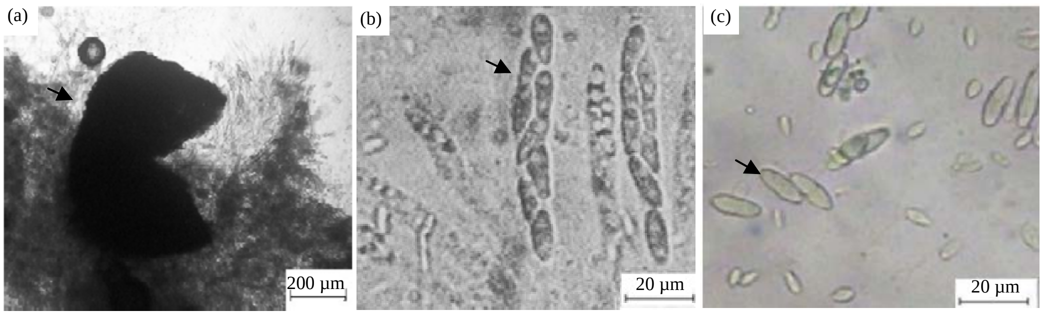

Fig. 3(a-c): Perithecia and ascospores of *Gibberella moniliformis* (MP-A) produced by crossing OLN0380O strain with MP-A tester, (a) Perithecium on carrot agar *in situ* observation, (b) Ascospores in ascus and (c) Ascospores

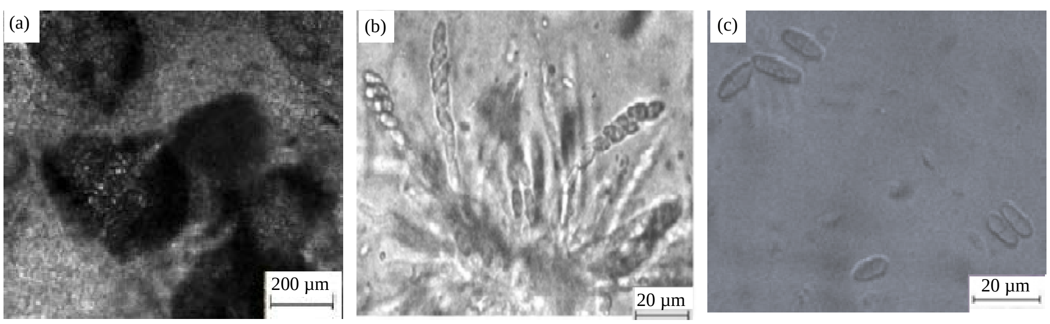

Fig. 4(a-c): Perithecia and ascospores of *Gibberella intermedia* (MP-D) produced by crossing OLN0279O strain with MP-D tester, (a) Perithecia on carrot agar *in situ* observation, (b) Ascospores in ascus and (c) Ascospores

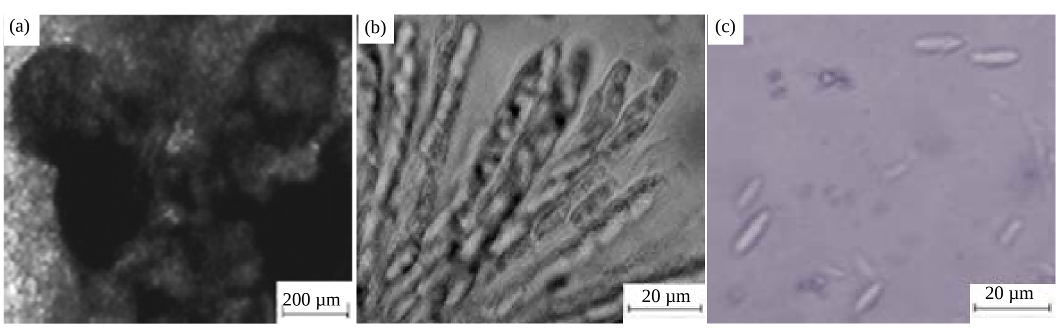

Fig. 5(a-c): Perithecia and ascospores of *Gibberella subglutinans* (MP-E) produced by crossing OLN0140O strain with MP-E tester, (a) Perithecia on carrot agar *in situ* observation, (b) Ascospores in ascus and (c) Ascospores

(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.

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