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Isolation and characterization of plant-growth promoting fungi associated with indigenous fermented amendment from root vicinity of *Phyllostachys aurea*

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Abstract Results revealed that ITS sequence analyses expressing five isolated fungi belonging to Ascomycota, with three distinct genera, *Aspergillus*, *Cladosporium* and *Parengyodontium* were phylogenetically determined which closed to the reference strains. Most of the isolates were categorized as dark septate endophytes in appearances with exception of white mycelium *Parengyodontium album* IMX 2. The result showed that *Parengyodontium album* IMX 2 had a maximum IAA productivity of 21.10 ± 1.04 μg/mL in the presence of 500 mg/L tryptophan. The fungal isolates of *Cladosporium sphaerospermum* IMX 1, *Parengyodontium album* IMX 2, *Cladosporium endophytica* IMX 3 and *Aspergillus pseudotamarii* IMX 8 exhibited variable efficiency for solubilizing tricalcium phosphate salts. Moreover, *Cladosporium sphaerospermum* IMX 1 significantly suppressed the growth of *Fusarium oxysporum* and *Botrytis cinerea* by 49.64% and 76.73% respectively as compared to other isolates. These results suggested that IMOs and their plant growth-promoting fungi potentially could be used as a beneficial resource in promoting sustainable agriculture, which may act as a possible substitute for chemical fertilizers.

Keywords: *Phyllostachys aurea*, IMO, Plant-growth promoting fungi, Dark septate endophytes

Introduction

Agrochemicals namely chemical fertilizers are generally applied to crops to supply essential nutrients in ensuring plant's growth and productivity. However, the skyrocketing prices, availability issues and even environmental hazards as a result of indiscriminate use of synthetic fertilizers are becoming an worrying issue as demand to feed the whole world population is rapidly increasing (Phua *et al.*, 2012; Nicolopoulou-Stamati *et al.*, 2016). Nowadays, agricultural practice systems are highly concerned to shift to new paradigm for

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getting the best quality of yield while mitigating adverse effect in a more sustainable way. Consequently, natural farming strategies that rely on eco-friendly management practice are taking a global-eye attention to replace the old-conventional way of agriculture. Aiming to obtain the similar quality of produce without having long term consequences, a purported strategy could be centered on the utilization of belowground naturally-occuring microbes for robust crop production (Santoyo *et al.*, 2017).

Using microbial bioinoculants or any organic amendment supplements by exploiting beneficial microbes including plant-growth promoting fungi are critical contributing factor in boosting agricultural productivity. In parallel to reduce the agrochemical dependency, these elements are recognized to provide cross-protection against phytopathogen and mineralizing soil organic matter without comprimising quality of yield. In the same regard, Indigenous Microorganisms (IMOs) that artifically fortified microbial amendments with the fungi as the key player in fermentation process has widely practiced all over the world especially in Japan and Asian countries (Trump, 2016). IMOs, invented by Cho Han Kyu, a Korean scientist, are the group of an innate microbial consortium that inhabits the soil and other surfaces of forest soil vicinity area (Nyein *et al.*, 2017). The uniqueness of harmonious interaction among residing microbes and their biosynthetic capabilities, have conferred a great benefit to plants, assuring the accesibility of essential nutrients (Kumar and Gopal, 2015; Bánfi *et al.*, 2021). By having readily-available materials to 'bait' the fungi inocula before fortification with molasses substrate, nutrient-packed IMOs are feasible to be applied under variable ecological conditions.

Nature is bestowed with an abundance of microbes in which most of them are beneficial to sustain our environment. Among them, presence of filamentous fungal species in rhizosphere soils are very promising source, representing an attractive inoculant for the agricultural improvement. As the price of agrochemical is costly, the use of filamentous fungi as a free-living organism incorporated with readily-available agrowastes provides a cheaper option for bioorganic fertilizer with equal performance. Furthermore, these fungi are capable producing lytic enzymes to expedite decomposition process organic mattter and agricultural residues rapidly (Song *et al.*, 2010; Tamayo-Velez and Osorio, 2017; Naher *et al.*, 2021). It worth noting that some of these filamentous fungi were commonly reported to have proven not only as the best candidate for plant nutrient acquisition, but also to enhance immunity-induced antagonistism against various phytopathogens (Naziya *et al.*, 2020). Even though interest in utilizing IMO-based technology has been recorded for the past couple of decades in early 1991s, few studies regarding isolation of microbes within IMOs especially focusing on plant-growth promoting fungi has

been documented (Abu-Bakar and Ibrahim, 2013; Anyanwu *et al.*, 2013). With the exception of recent findings by Jan *et al.* (2020) and Xa and Nghia (2020) whom reported that the majority of the isolates were belonging to *Aspergillus* and *Trichoderma*. To best to our knowledge, other diversity of fungal species remains to be explored. Hence, a good microbial agent is required to enhance nutrient uptake and phytohormone release towards plants. Therefore, it is critical to isolate and characterize the local plant-growth promoting fungal species from IMOs sample taken from bamboo vicinity area with the view of utilizing them as inoculant sources in correspondence to promote sustainable agriculture. The plant growth-promoting abilities including phosphate solubilization, indole-3-acetic acid (IAA)-producing hormone and biocontrol activities against common phytopathogens were investigated.

Materials and methods

IMO preparation and sample collection

IMO (Indigenous Microorganism) was prepared using plastic container filled with steamed rice and covered by white paper and waist belt. The container was buried at least 2 inches deep under Ifagao bamboo soils and covered with leaf litter located in Titik Tengah Semenanjung Malaysia, Pahang state, Malaysia (03°29’33” N 102°13’10” E). After 5 days incubation, the steamed rice was fully grown with whitish mold. This process is called the cultivation of indigenous microorganisms, known as IMO1 (Indigenous Microorganism 1) . One gram of IMO sample was serially diluted into two different type of 9 mL sterile distilled water accordingly (heated and non-heated). For heat-resistant fungi isolation from IMO sample, the sample was heated in the shaking water bath water at 80 ℃ for 10 minutes. Potato dextrose agar (PDA, Oxoid Co., UK) added with 100 mg/L chloramphenicol (Sigma Aldrich, USA) was used for isolation of fungal species. The plates were incubated at 26 ℃ for 14 days under aerobic conditions. All visible fungal species were transferred onto fresh PDA for culturing. Then, pure cultures were stored in PDA slant with pH 5.5 and kept as master cell culture banking for further experiments. The slant agars were incubated at 26 ± 2 ℃ for another 21 days followed by storing in the fridge at 4 ℃.

Morphological and biochemical characterizations

Pure isolated fungi were subjected to morphological characterization analysis. All isolates were recultured aseptically from the slant agar and transferred onto fresh PDA and incubated at 26 ± 2 ℃ for 6 days. Following

the protocol of Naher *et al*. (2021), a slide culture preparation procedure was used in the microscopic examination. Regular observations were performed according to the following characteristics: conidia, conidiophores branching patterns and phialides. After 2 days, a thin section of the agar slide culture was observed under light compound microscope (Olympus CX31, Japan) with 100x and 400x magnifications. The isolates were tested for plant-growth promoting traits such as phosphate solubilisation, indole acetic acid (IAA) production and antagonistic activity.

Phosphate solubilization assay

The assay was carried out using 250 mL flask containing 50 mL of Pikovskaya (PVK) broth medium contained per L: glucose, 10 g; $Ca_3(PO_4)_2$, 5 g; $(NH_4)_2SO_4$, 0.5 g; NaCl, 0.2 g; $MgSO_4 \cdot 7H_2O$, 0.1 g; KCl, 0.2 g; yeast extract, 0.5 g; $MnSO_4 \cdot H_2O$, 0.002 g; and $FeSO_4 \cdot 7H_2O$, 0.002 g at pH 5.5. The 50 mL of PVK broth was inoculated with 2 culture discs for each isolated strain (from a 14-day-old culture) and incubated at 26 ℃ for 14 days at 150 rpm. After incubation period, the cultures were harvested by centrifugation at 6000 rpm for 15 min. Sterile uninoculated medium served as control. 500 µL of culture supernatant was estimated using for phosphate determination by mixing with 500 µL of 10 % Trichloroacetic acid in a test tube to which was added 4 mL of colour reagent (1:1:1:2 ratio of 3M H_2SO_4, 2.5 % (w/v) ammonium molybdate, 10 % (w/v) ascorbic acid and distilled water and incubated at room temperature for 15 min (Ganesh *et al.*, 2012; Mohd Din *et al.*, 2019). The absorbance of blue colour change of soluble phosphorus was determined using the calibration curve with KH_2PO_4 at 820 nm. All experiments were made in triplicate.

Screening and quantification of Indole-3-Acetic Acid (IAA)

Production of IAA was based on the colorimetric method with minor modifications (Sasirekha *et al.*, 2012). The 50 mL of PVK broth was inoculated with 2 culture discs for each isolated strain (from a 14-day-old culture) and incubated in incubator shake at 26 ℃ for 14 days at 150 rpm. Each isolate was inoculated in 50 mL NPT (2 g/L nutrient broth, 5 g/L potato dextrose, 6 g/L tryptic soy broth, 2 g/L MES hydrate, pH 5.75), with 500 mg/L tryptophan as the precursor for IAA biosynthesis. Non-inoculated media were considered as controls. After 14 days incubation in the incubator shaker at 26 ℃ with 150 rpm, the culture was centrifuged at 5,500 x g for 10 min, and 1 mL of supernatant was mixed with 4 mL of Salkowski's reagent (150 mL concentrated H_2SO_4, 250 mL distilled H_2O, 7.5 mL 0.5 M $FeCl_3.6H_2O$). The

emergence of pink color indicated IAA production. Equivalent mixtures with non-inoculated media served as blanks for the spectrophotometry absorbance were measured at 535 nm. The amount of produced IAA was determined by comparing it with a standard IAA graph. All experiments were made in triplicate.

In vitro antagonism assay

Dual culture assay was used to test the antagonistic abilities of isolated fungi against common phytopathogen fungi, namely as *Fusarium oxysporum* ICA F02 and *Botrytis cinerea* ICA F03. ICA F02 and ICA F03 were routinely cultured in PDA at 26 ± 2 ℃. Six mm diameter mycelial plugs were taken and spot-inoculated on to the edge of PDA. Meanwhile, mycelial plugs grown with isolated fungal species were placed opposite to a plugs of phytopathogen fungi (B). PDA plates containing only phytopathogen plugs were considered as a control treatment (A). The mycelial growth rate was checked after 14 days. The diameter of the pathogen growth was measured and compared with that of the control. Percent of inhibition of mycelial growth was calculated by using the following formula: (A-B)/(A X 100) (Skidmore and Dickinson, 1976). The assay was repeated thrice.

Molecular identification

DNA extraction, amplification and sequencing

The isolates having the most positive result for plant growth promotion were subjected to molecular identification. Seven-day-old pure cultures of the morphologically similar isolates were used for genomic DNA extraction. DNA extraction was carried out using a commercial kit (Apical, Kuala Lumpur, Malaysia) following the manufacturer's instructions. The polymerase chain reaction (PCR) was carried out using the primer pair ITS1 (5'-TCCGTAGGTGAACCTGCGG-3') and ITS4 (5'-TCCTCCGCTTATTGATATGC-3') to amplify the internal transcribed spacer (ITS) region in the DNA using a thermocycler (Eppendorf, Hamburg, Germany). The PCR mixture was 25 uL and was prepared by adding 12.5 µL of Taq 2X PCR master mix, 1 µL each of primers ITS 1 and ITS 4 (10 µM), 9.5 µL of double-sterilized distilled water (ddH_2O), and 1 µL of the DNA template with ddH_2O, used as the template for the control reaction instead of the DNA. The PCR program used was carried out according to the following temperature profile: An initial step of 2 min at 98 ℃, for initial denaturation; 25 cycles (98 ℃ for 15 sec; 60 ℃ for 30 sec; 72 ℃ for 30 sec) for annealing and extension,

and 1 cycle (72 ℃ for 10 min) for final extension of the amplified DNA. The expected size of the amplicon was 550 bp. The PCR products were purified by standard method and directly sequenced using Big Dye® Terminator v3.1 Cycle Sequencing Kit (Applied Biosystems, USA). Gel electrophoresis: Electrophoresis of PCR-amplified products was performed in 1.5 % agarose gels for 1.5 h at 7.0 V. The PCR products were stained with ethidium bromide and visualized with 305 nm ultraviolet light.

Phylogenetic analysis

The result of sequencings was used for similarity searches on the GenBank database using basic local aligment search tool (BLAST) algorithm. The fungal ITS sequences at this study have been deposited in GenBank under accession numbers ON527246, ON527248-ON527251. Similar sequences were aligned and the phylogenetic tree was constructed in MEGA v7.0 (Kumar *et al.,* 2018). The tree was built using Neighbor-Joining approach with 1000 bootstrap replications.

Statistical analysis

Experiments were carried out in triplicate and the mean results are presented. Comparison between means was carried out using a one-way analysis of variance (ANOVA) and the significant of difference between means was determined by t-test. All statistical analysis was carried out using the statistical package in SPSS v 16.0 at 95% least significant difference ($p < 0.05$).

Results

Morphological and molecular identification

By using macroscopic observation, the physical appearance and the color of the colony were compared with other reported fungi isolates from the literatures that literally grown under same condition on general growth medium agar plate such as PDA. Only 5 colonies were able to identify and selected in this study (Figure 1). On the basis of culture observation, 4 indigenous fungi colonies known as IMX A1, IMX 1, IMX 3 and IMX 8 had a fluffy green black filamentous mycelium after 14 days incubation. Out of these 4 colonies, only IMX A1 was isolated from sample heated at 80 ℃ for 10 minutes in the shaking water bath. Of all isolations were performed, only strain IMX 2 showed cotton white mycelium on PDA after 14 days of incubation which might be belong to *Engyodontium* or *Parengyodontium* or *Tritrachium* (Figure 1).

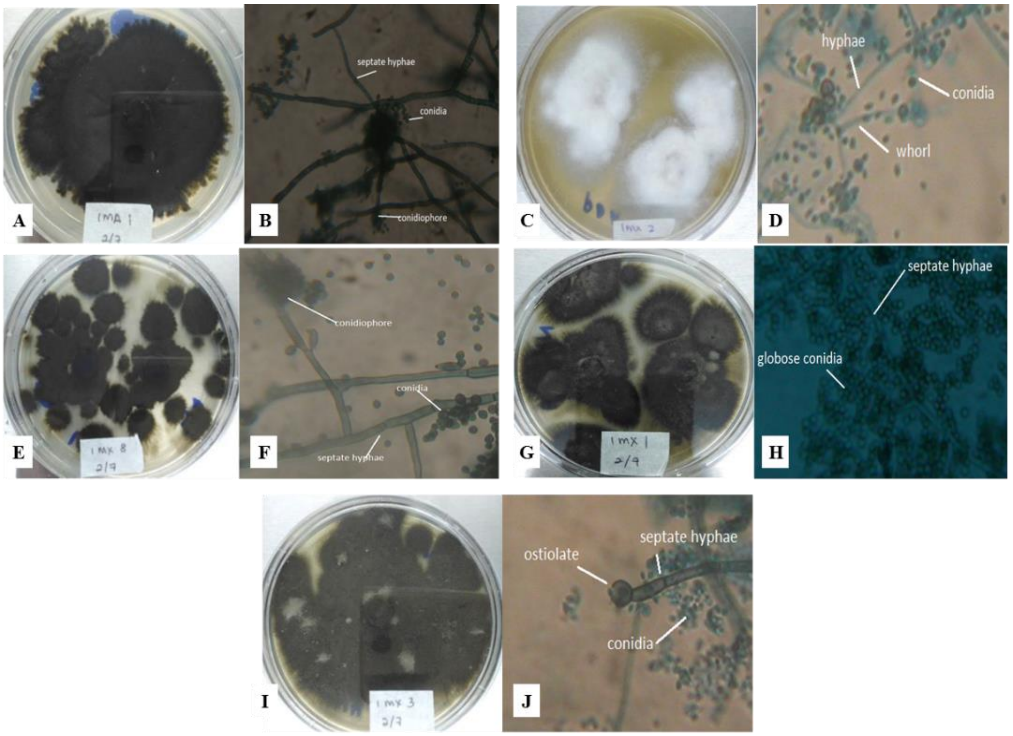

Figure 1. Representative fungal strain morphology with each microscopic characteristic: (A) Colony of IMX A1; (B) Microscopic feature of IMX A1; (C) Colony of IMA 2; (D) Microscopic feature IMA 2; (E) Colony of IMX 8; (F) Microscopic feature of IMX 8; (G) Colony of IMX 1; (H) Microscopic feature of IMX 1; (I) Colony of IMX 3; (J) Microscopic feature of IMX 3. All microscopic images are observed under 400 x magnification

In this study, 5 fungi were identified which based on internal transcribed spacer (ITS) sequence analysis, fungal isolates were identified as: *Aspergillus tamarii* (IMX A1), *Cladosporium sphaerospermum* (IMX 1), *Parengyodontium album* (IMX 2), *Cladosporium endophytica* (IMX 3) and *Aspergillus pseudotamarii* (IMX 8). The BLAST analysis of these fungal strains reveled 98-99% identity with ITS sequences of the related species (Table 1). The phylogenetic tree clearly showed the similar affliations, in line with morphological identification (Figure 2).

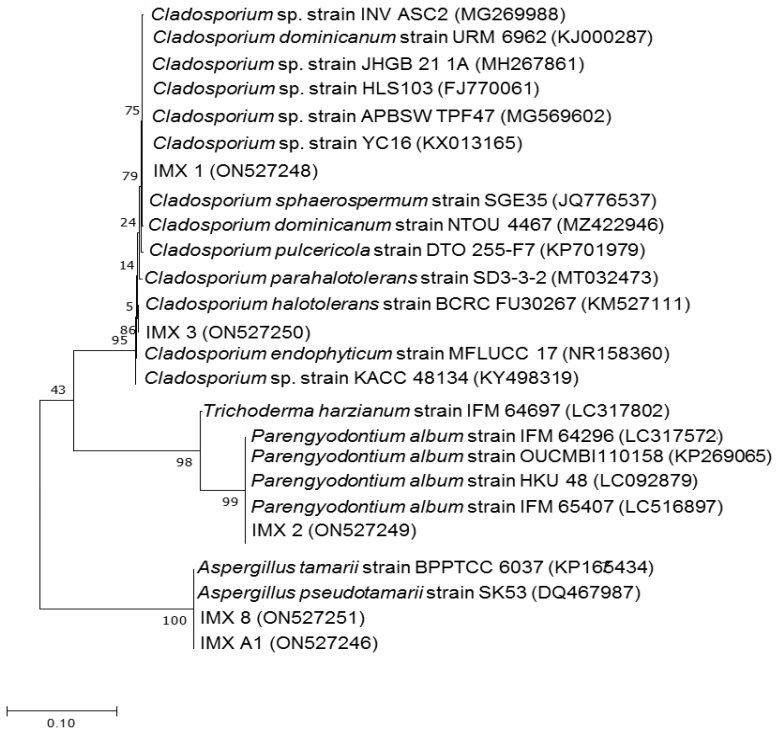

Figure 2. Phylogenetic analysis of ITS sequences of isolated fungal strains with reference sequences retrieved from NCBI (National Center for Biotechnology Information): IMX A1, IMX1, IMX 2, IMX 3, IMX 8 refer to the ITS sequences of fungal isolates. The analysis was generated using maximum likelihood with 1000 bootstrap replications

Table 1. List of isolated strains obtained from sequencing the ITS region of 5 fungal isolates from root vicinity of bamboo tree*

Isolates	Sequence Size	GenBank Accession number	Closest match (GenBank accession number)	Homology identical (%)
IMX A1	554 bp	ON527246	*Aspergillus tamarii* (MG772816)	99.50
IMX 1	554 bp	ON527248	*Cladosporium sphaerospermum* (JQ776537)	99.27
IMX 2	601 bp	ON527249	*Parengyodontium album* (MT102852)	99.49
IMX 3	554 bp	ON527250	*Cladosporium endophytica* (NR158360)	99.81
IMX 8	606 bp	ON527251	*Aspergillus pseudotamarii* (DQ467987)	100.00

*ITS: Internal Transcribed Spacers. Isolates with an ITS match > 99% similarity were considered as the same species as the NCBI acession.

Assessment of fungal strains on plant growth-promoting traits

Phosphate solubilization activity

The present study revealed that all isolated fungi were able to solubilize and utilize inorganic phosphate in the form of tricalcium phosphate [$Ca_3(PO_4)_2$] as the sole phosphate source in the PVK broth with exception of strain IMX A1. The strain of IMX 2 showed the highest phosphate solubilisation activity when compared with others isolated fungi strain. IMX 2 exhibited the ability to solubilize the phosphate of PVK broth with 34.55 % (Table 2).

Indole-3-acetic acid (IAA) production assay

In this study, analyses of phytohormone production by isolated fungi have detected production of IAA from each fungus are significantly differed, ranging between 0.51 and 21.10 µg/mL. The strain of IMX 2 exhibited the highest IAA production compare to other strain. The ability of IMX 2 to synthesize IAA was determined by reaction of liquid culture with Salkowski's reagent, which detects IAA and its intermediates (Table 2). The reaction produces a proportionate amount of tric-(indole-3-acetato) iron (III) complex, which is pink in color and is quantified by measuring absorbance at 535 nm. IMX 2 produced an average of 21.10 ± 1.04 µg/mL of IAA (or its intermediates) when grown with 500 mg/L tryptophan (the precursor for IAA biosynthesis) in liquid NPT medium.

Table 2. Plant growth-promotion traits of isolated fungal strains

Isolates	Indole-3-acetic acid (IAA) production (µg/mL)	Phosphate solubilization assay (%)	*In vitro* antagonism assay (% of inhibition)	
			F. oxysporum	***B. cinerea***
IMX A1	0.51 ±0.32^{a}	ND	28.46 ±0.77^{d}	15.35 ±0.45^{b}
IMX 1	1.81 ±0.03^{b}	0.04 ±0.01^{a}	49.64 ±0.70^{f}	76.73 ±4.84^{c}
IMX 2	21.10 ±1.04^{e}	34.55 ±0.76^{d}	5.95 ±0.11^{b}	14.22 ±0.78^{b}
IMX 3	6.03 ±0.05^{d}	0.97 ±0.09^{b}	36.64 ±0.90^{e}	12.88 ±0.15^{b}
IMX 8	2.5 ±0.11^{b}	4.62 ±0.23^{c}	3.08 ±0.06^{a}	2.07 ±0.06^{a}

ND-Not detected, Different letters on the same column are significantly different at $P < 0.05$

In vitro antagonism assay

All isolated fungi showed antagonistic activity against tested phytopathogenic fungi (Table 1). Based on the *in vitro* antagonism assay, strain IMX 1 showed the highest growth inhibition for *Fusarium oxysporum* ICA F2 and *Botrytis cinerea* ICA F3 as compared to other isolated strains (Table 2). Among all the isolated strains, IMX A1 and IMX 3 showed moderate inhibition

against both pathogens. Meanwhile, for IMX 8, showed a very weak inhibition for *in vitro* antagonism assay against of *Fusarium oxysporum* ICA F2 and *Botrytis cinerea* ICA F3.

Discussion

In this study, the fungal isolation was carried out using PDA medium before identification process took place with regard to morphological and molecular characteristics. The morphological and microscopic characteristics of isolated fungal strain IMX A1 and IMX 8 had a feature resemblance to genus *Aspergillus* according microscopic observation by Zhang *et al.* (2012) and Rustamova *et al.* (2020). These two strains (IMX A1 and IMX 8) were observed to have septate hyphae, biseriate conidial heads and small conidia (Fig.1). The morphological characteristics of strain IMX 1 showed the presence of a dark olive-green colony colour. The IMX 1 conidia are formed on densely compacted, non-specialized, slightly determinant and conidial in shape of globose to pyriform. This fungal isolate shared a sequence similarity with *Cladosporium sphaerospermum* strain. Hamayun *et al.* (2009) reported for the first time regarding the gibberilin-producing capability of this isolate so far and accessed their role in soybean growth. For IMX 3, it was observed to be grey to green of massive mycelial mat with pycnidia grey to black globose which showed a short neck with ostiolate. All these features which known as dark septate endophytes (DSE) were similarly shared with the genus *Cadaphora and Talaromyces* as indicated by Day and Currah (2011) and Likar and Regvar (2013) in their observations. This DSE-like feature likely provides crucial ecological signficance for plant host to withstand abiotic stress (Berthelot *et al.*, 2017). The microscopic characters of IMX 2 under light microscope showed the presence of whorls and conidia that can be attributed to the genus *Parengyodontium* (Tsang *et al.*, 2016). They reported that this fungus was an environmental saprobic mould, found in skin human lesion. Interestingly, some species identified in this study were not commonly reported elsewere in plant-soil association and not considered as an endophytic fungi. With an exception, Wu *et al.* (2013) and Belfiori *et al.* (2019) reported that *Parengyodontium album* was found as an endophyte of *Panax ginseng* and *Crocus sativus* saffron roots respectively. Most of the isolates in this study were described previously as pathogenic fungi, but we found them from IMO, which considered as an organic amendment for plant-growth promoting tool. We cannot infer any further explanation although hypothesis of a contamination was likely to occur. But the growth-promoting activities and biocontrol of these isolates outweight

those assumptions. To the best of our knowledge, our identified fungal species were different from reported literature, thus enriching the fungal diversity.

Phosphorus (P) is one of the important macronutrients for plant growth requirement (Malhotra *et al.,* 2018, Mohd Din *et al.,* 2019). Phosphate solubilisation trait is key critical mechanisms for plant growth promoting fungi (PGPF) to maintain P solubility in soil and function as a promising tool to alleviate P deficiency (Rashid *et al.,* 2016; Wang *et al.,* 2018). Thus, the application of PGPF is an eco-friendly strategy to encounter P insolubility due to excessive chemical P fertilizer. The performance of isolated fungal strains to solubilize phosphate into soluble form in the following manners IMX 2>IMX 8>IMX 3>IMX 1 and IMX A1 were not possessing phosphate solubilizing activity. The results were in agreement with the results demonstrated by Jain *et al.* (2017), as they reported a potential of P solubilisation by fungal isolates in their cultures. It is expected that these fungi has greater phosphate solubilizing activity in the soils because their mycelium could serve for them to spread and grow deeper in the soil as compared to bacteria (Sharma *et al.,* 2013). Several filamentous fungi such as genera of *Aspergillus*, *Penicillium*, *Trichoderma*, *Rhizoctonia* and *Absidia* have been reported and investigated in details as a good phosphate solubilization activity as well (Whitelaw *et al.,* 1999; Sharma *et al.,* 2013).

It is known that fungus have ability to produce IAA that can help to stimulate plant cell elongation and cell division, increasing root size and distribution which leads to greater nutrient absorption from the soil. Appropriate concentrations of exogenous IAA can help to stimulate the growth and development of plant root systems by activating root branching and lateral root development (Fonseca *et al.,* 2018). There are strong evidences that most of phytohormone biosynthesis may stimulate symbiosis between plants and consortium of soil fungus for colonization in soil which indirectly stimulate plant growth and defense against pathogens (Farias *et al.,* 2018; Shen *et al.,* 2018). Our study indicated that all isolates are capable to synthesize IAA in presence of tryptophan with variable measurements. These results are compatible with Khalil *et al.* (2021), who reported that the variation in IAA production from same source of origin were attributed to genetic factors of particular species. IAA-producing PGPF was known to improve the absorption of nutrients and water from the soil through facilitating root growth development (Fu *et al.,* 2016).

*I*n *vitro* antagonism assay of all isolated fungal strain showing different trend against both plant pathogens. The biological control attributes of PGPF indirectly are reported to be able to induce plant resistance against phytopthogens including *F*usarium *oxysporum* and *Botrytis cinerea* (Zhang *et*

al., 2018; Muslim *et al.,* 2019). Ability to colonize plant roots effectively and enzyme (glucose oxidase) secretion are considered the most important mechanism involved in preventing pathogenic infection (Kriaa *et al.,* 2015; Hossain *et al.,* 2017). Typically, if this isolated fungal species were free-living and posed a growth-promoting trait, they supposed to be able to combat phytopathogen attack. We are still not clear of the reduction trend of isolate (IMX 8) with regards to inhibition activity against phytopathogens. The logical reason behind this, may be due to presence of pathogen-like fungi living together with beneficial fungi to create a good-habitable environment. This situation has been reported by Venturi and da Silva (2012) whose mentioned that interspecies interaction might be occurred between these ecosystems as a result of multifaceted harmless bacteria-pathogen signalling. The present observations suggested that most of indigenous fungi probably produce considerable amounts of antimicrobial metabolites and posses mycoparatism and antibiosis that suppress the growth of plant and soilborne pathogens with exception for IMX 8.

From this research, all these isolated fungi strains have great potential of plant-growth promoting properties with regards to phosphate solubilisation activity, IAA production and antagonistic activity although showing variable results. The most promising fungal strain as demonstrated by IMX 2 through the highest phosphate solubilisation activity and IAA production as compared to others. However, the capability of certain traits can be further enhanced through formulation in the form of a consortium to boost the synergistic effect. With the above notions in considerations, an important implication of this research is to understand beneficial contribution of indigenous fungus as a plant growth promoter and facilitator for plant nutrient uptake. In addition, this study provides novel insights regarding the fungal diversity inhabiting IMO fermented amendment sample. Apart from acting as a plant growth promoter, this application of PGPF particularly strain IMX 2 can be a potential candidate for commercial usage in wider scale in the field, aimed at cutting the dependency of chemical fertilizer and promoting sustainable agriculture.

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