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Diversity of Marine Fungi in Eastern Thailand

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ABSTRACT

An investigation on the diversity of marine fungi was carried out by isolating fungi from twigs collected from several beaches in Chanthaburi, Rayong and Chon Buri provinces. Each fungus was directly isolated on half strength potato dextrose agar with 70% sea water. A total of 152 marine fungal isolates were found, comprising 26 genera and 31 species: *Algialus parvus*, *Antennospora quadricornuta*, *Arenariomyces trifurcatus*, *Camerosporium* sp., *Clavatospora bulbosa*, *Cladobotryum* sp., *Corollospora pulchella*, *Dactylospora haliotrepha*, *Didymosphaeria maritime*, *Emericella nidulans*, *E. variecolor*, *Eurotium* sp., *Halosphaeria quadricornuta*, *H. quadriremis*, *H. hamata*, *Herpotrichella* sp., *Kallichroma tethys*, *Lulworthia grandispora*, *Lulworthia* sp., *Marinosphaera mangrovel*, *Nais inornata*, *Pestalotia* sp., *Pleospora* sp., *Pontoponae* sp., *Savoryella paucispora*, *Spirodesmium eupatoriicola*, *Torpedospora radiata*, *Varicosporina ramulosa*, *Verruculina enalia*, *Zalerion varium* and an unidentified sp.1. Pure cultures are being maintained in a culture collection at the Department of Plant Pathology, Faculty of Agriculture, Kasetsart University, Bangkok for further study on enzyme and secondary metabolites, which could be very useful in agriculture and medicine.

Key words: diversity, marine fungi, twigs, Ascomycetes, secondary metabolites

INTRODUCTION

Fungi from the marine environment have shown great potential as an important source of pharmacologically active metabolites. The biological activity of their metabolites is mainly of interest because of their antibiotic and anticancer properties, and to a lesser extent for other selective activities, such as cell cycle inhibition, antagonism of platelet activating factors, antiviral activity and radical scavenging activity (Bugni and Ireland, 2004). Several of the metabolites from marine fungi have shown biological activity as antibiotics, antitumor or immunosuppressive agents and as enzymes (Bhadury *et al*., 2006). In Thailand, 154 marine fungi were reported from 16 provinces in Thailand, including 118 Ascomycota, 26 Deuteromycota, 3 Basidiomycota, 28 anamorphic fungi and 7 Stramenopiles and 33 were new records for the country (Jones *et al*., 2006). The common species found in Thailand were: *Antennospora quadrinuta, A. salina, Cirrenalia pygmea, Corollospora maritime, Dactylospora haliotrepha, Halocyphina villosa, Halorosellinia oceanica, Lignincola leavis, Lulworthia grandispora, Periconia prolifica, Saagaromyces abonnis, Trichocladium melhae, Torpedospora radiata*, *Verruculina enalia* and *Zalerion varium*. They were mostly isolated from sand, driftwood and decayed wood of mangrove trees (Sakayaroj *et al*., 2004; Jones *et al*., 2006). In addition, Manoch *et al*. (2007)

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reported 278 fungal isolates from a mangrove swamp at the Kung Krabaen Bay Royal Development Study Center, Chanthaburi province. Later, Suetrong *et al*. (2007) studied marine fungal diversity in Thailand. They reported 10 new records of marine fungi from driftwood and attached decaying mangrove wood from central, eastern and southern Thailand including: *Aigialus* cf. *mangrovel*, *Dendryphiella arenaria, Lindra thallasiae, Mycosphaerella avicenniae, Manglicola guatemalensis*, *Patellaria* sp., *Pontoporiae* sp., *Sporomiella* sp., *Swampomyces aegyptiacus* and *Varicosporina prolifica*.

The purposes of this study were to: 1) isolate and study species diversity and the distribution of marine fungi from twigs in Chanthaburi, Rayong and Chon Buri provinces, and 2) maintain cultures to find new taxa capable of producing secondary metabolites, which could be very useful in agriculture and medicine.

MATERIALS AND METHODS

Twigs were collected from beaches in Chanthaburi, Rayong and Chon Buri provinces (Table 1). The isolation technique involved using a fine needle to pick up the fruiting bodies of fungi from twigs and transfering them to a Petri-dish containing $^1/_2$ potato dextrose agar ($^1/_2$ PDA) with 70% sea water (Ebel, 2007). They were incubated at 28°C for 5 d and then the hyphal tips were transferred onto $^1/_2$ PDA slant and maintained as pure culture for identification. Slides were prepared with sterile distilled water and lactophenol. Specimens were examined under stereo- (Olympus SZ-PT) and light microscopes (Olympus BH-2) with Nomarski Interference Contrast. Identification of the fungal isolates was based on the morphological characteristics of fruiting bodies and spore ornamentation examined under stereo- and light microscopes.

RESULTS

The results revealed that 152 marine fungal isolates, comprising 26 genera and 31 species were found on the beaches in Chanthaburi, Chonburi and Rayong (Table 2). Most species of marine fungi were similar to those previous reported (Sakayaroj *et al*., 2004; Jones *et al*., 2006). The most common species were: *Dactylospora haliotrepha*, *Halosphaeria quadricornuta* (Figure 1F), *Lulworthia grandispora*, *Torpedospora radiata* (Figure 2C-D), and *Verruculina enalia*, whereas *Camerosporium* sp. isolated from a twig at Mae Pim Beach was the first record of this species in Thailand (Figure 1G). *Antennospora quadricornuta*, *Camerosporium* sp., *Marinosphaera mangrovel*, *Pontoponae* sp. and an unidentified sp.1 were isolated only from twigs at Mae Pim Beach, Rayong. However, the fungi found only at Bang Sa Rai Beach, Chonburi were *Didymosphaeria maritime* and *Emericella variecolor*, whereas *Emericella nidulans* and *Savoryella paucispora* were isolated from Sattahip Beach, Chon Buri. *Corollospora pulchella* and *Pestalotia* sp. were found at Sai Kaew Beach and

Table 1 Locations and date of collection.

No.	Location	Date of collection
1	Chanthaburi Beach, Chanthaburi	11/7/08
2	Sri Ra Cha Beach, Chon Buri	5/12/07
3	Bang Sa Rai Beach, Chon Buri	11/5/08
4	Sattahip Beach, Chon Buri	2/7/08
5	Sai Kaew Beach, Chon Buri	22/9/08
6	Mae Pim Beach, Rayong	9/10/08

Sri Ra Cha Beach, Chon Buri, respectively. In this study, many species of marine fungi were found that could produce important compounds, such as *Emericella variecolor*, *Algialus parvus*, *Pestalotia* sp.. Pure cultures are being maintained in a culture collection at the Department of Plant Pathology, Faculty of Agriculture, Kasetsart University, Bangkok for further study.

DISCUSSION

The results of this study were compared with those of the survey by Jones *et al*. (2006), who analyzed the occurrence of marine fungi on driftwood samples along the coastal beaches of Eastern Thailand. They found 45 species, out of which 14 were detected in the present survey:

Table 2 Marine fungi isolated from twigs at different locations.

Fungi	Class	Location
Algialus parvus	Ascomycetes	2, 3
Antennospora quadricornuta	Ascomycetes	6
Arenariomyces trifurcatus	Ascomycetes	5, 6
Camerosporium sp.	Hyphomycetes	6
Clavatospora bulbosa	Hyphomycetes	1, 3, 4
Cladobotryum sp.	Hyphomycetes	2
Corollospora pulchella	Ascomycetes	5
Dactylospora haliotrepha	Ascomycetes	1, 2, 3, 4, 5, 6
Didymosphaeria maritime	Ascomycetes	3
Emericella nidulans	Ascomycetes	4
Emericella variecolor	Ascomycetes	3
Eurotium spp.	Ascomycetes	2,6
Halosphaeria quadricornuta	Ascomycetes	1, 2, 3, 4, 5, 6
Halosphaeria quadriremis	Ascomycetes	3, 4
Halosphaeria hamata	Ascomycetes	1, 2
Herpotrichella spp.	Ascomycetes	3, 5
Kallichroma tethys	Ascomycetes	1, 4, 5
Lulworthia grandispora	Ascomycetes	1, 2, 3, 4, 5, 6
Lulworthia spp.	Ascomycetes	2, 5, 6
Marinosphaera mangrovel	Ascomycetes	6
Nais inornata	Ascomycetes	2, 5
Pestalotia sp.	Coelomycetes	2
Pleospora spp.	Ascomycetes	4, 5
Pontoponae sp.	Ascomycetes	6
Savoryella paucispora	Ascomycetes	4
Spirodesmium eupatoriicola	Hyphomycetes	1, 2, 3, 6
Torpedospora radiata	Ascomycetes	1, 2, 3, 4, 5, 6
Varicosporina ramulosa	Hyphomycetes	4, 5
Verruculina enalia	Ascomycetes	1, 2, 3, 4, 5, 6
Zalerion varium	Hyphomycetes	2, 3, 4
unidentified sp.1	Ascomycetes	6

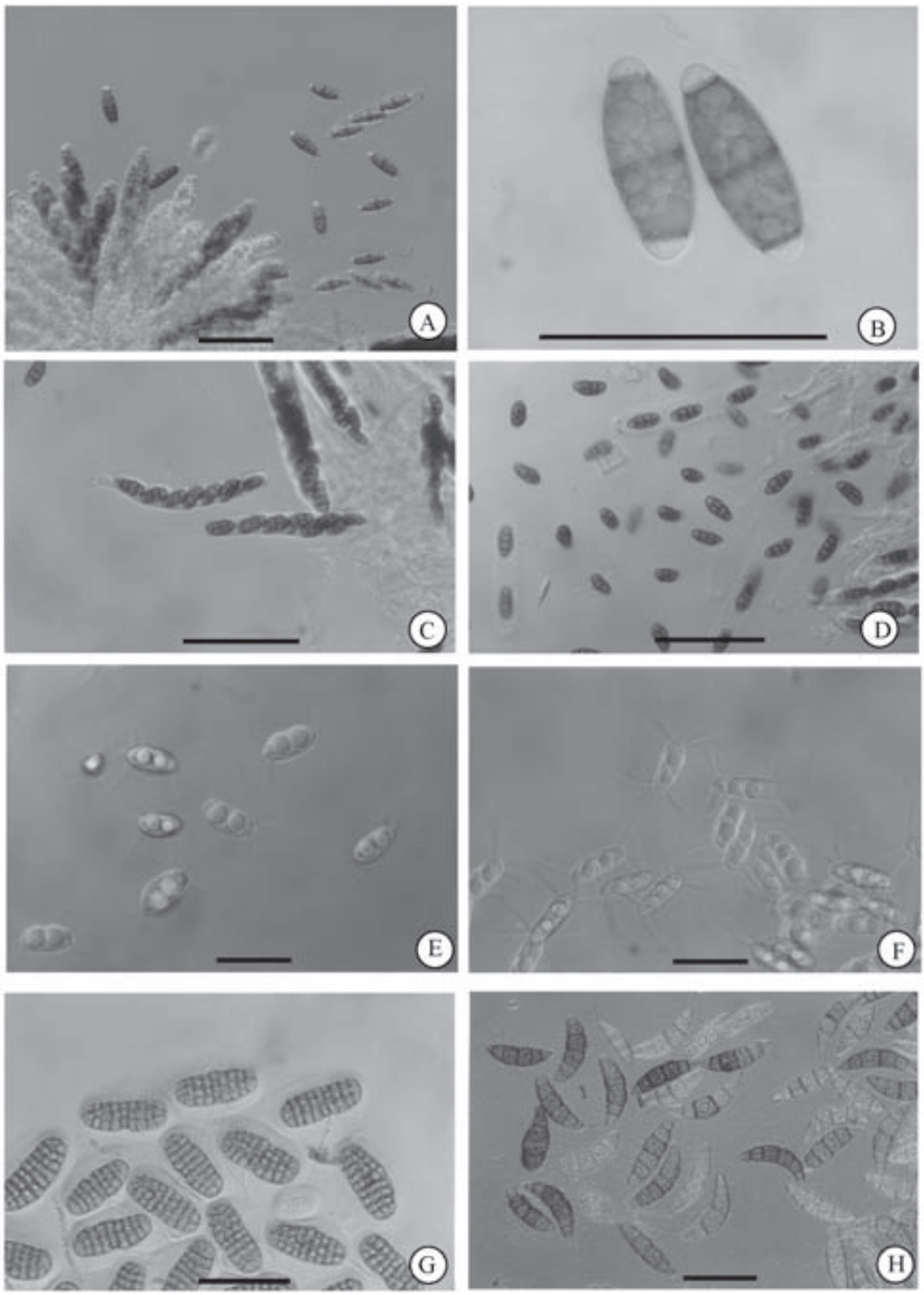

Figure 1 A-B. Asci and ascospores of *Savoryella paucispora*, C-D. *Pleospora* sp., E. Ascospores of *Halosphaeria quadriremis*, F. *Halosphaeria quadricornuta*, G. Conidia of *Camerosporium* sp. and H. Ascospores of an unidentified sp.1. (Scale bars: A, C, G, H = 50 μm; B, D, E, F = 30 μm)

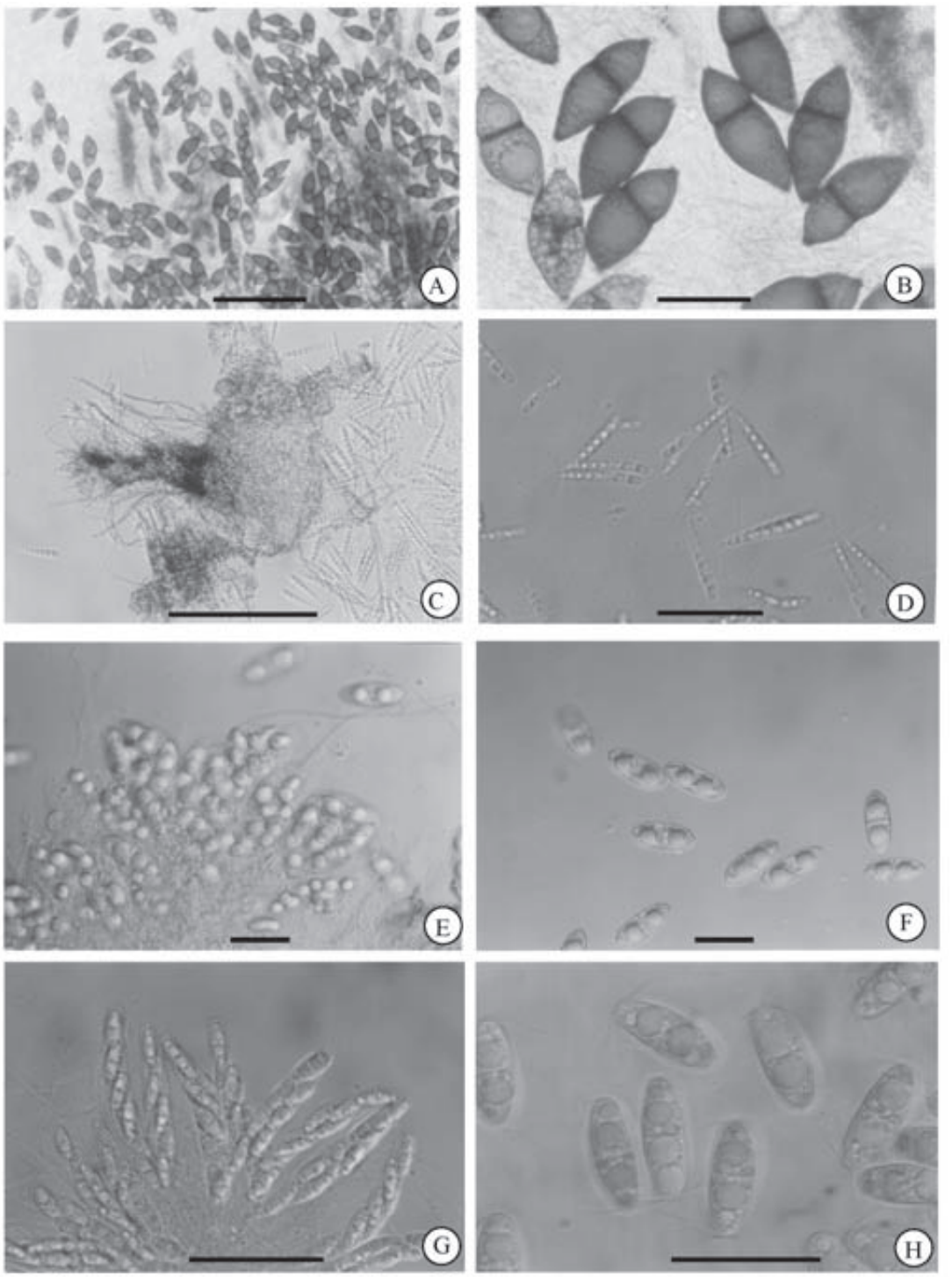

Figure 2 A-B. Asci and ascospores of *Didymosphaeria maritime*, C-D. Perithecia and ascospores of *Torpedospora radiata*, E-F. Asci and ascospores of *Nais inornata*, G-H. *Arenariomyces trifurcatus* (Scale bars: A, C, G = 50 μm; B, D, E, F, H = 30 μm)

Algialus parvus, *Camerosporium* sp., *Didymosphaeria maritime*, *Halosphaeria quadricornuta*, *H. quadriremis*, *H. hamata*, *Herpotrichella* sp., *Kallichroma tethys*, *Lulworthia grandispora*, *Lulworthia* sp., *Nais inornata*, *Pleospora* sp., *Pontoponae* sp. and *Spirodesmium eupatoriicola*. Marine fungi play an important role in the decomposition of plant material in their habitats and can produce novel compounds with great potential as pharmaceuticals, nutritional supplements, cosmetics and agrichemicals where these marine bioproducts have a strong potential market value (Faulkner, 2001). This current study has started to isolate marine fungi that may produce compounds with potential in the fields of agriculture and medicine. Many species of marine fungi have been found that produce important compounds, such as *Emericella variecolor, Algialus parvus* and *Pestalotia* sp..

Pornpakakul *et al*. (2007) have reported 4 xanthones, shamixanthone, 14-methoxy tajixanthone -25-acetate, tajixanthone methanoate, and tajixanthone hydrate from *Emericella variecolor,* an endophytic fungus isolated from *Croton oblongifolius* in Thailand. All compounds showed moderate activity against gastric carcinoma, colon carcinoma and breast carcinoma. Malstrom *et al*. (2002) recorded similar compounds and varixanthone from *E. variecolor* (sponge derived) in Venezuelan waters of the Caribbean Sea. Isaka *et al*. (2002) reported that aigialomycin from the mangrove fungus *Aigialus parvus* showed *in vitro* antimalarial activity. In the current study, *Algialus parvus* was isolated from decaying wood at Sri Ra Cha and Bang Sa Rai Beaches, Chon Buri. In addition, Chinworrungsee *et al*. (2001) reported halorosellinic acid from *Halorosellinia oceanica* showed antimalarial properties. Pestalone, a new chlorinated benzophenone compound isolated from the marine fungus *Pestalotia* sp., showed potent antibiotic activity against methicillin-resistant *S. aureus* and vancomycin-resistant *Enterococcus faecium* (Bhadury *et al*., 2006). In this study, *Pestalotia* sp. was isolated from decaying wood, Sri Ra Cha Beach, Chon Buri.

CONCLUSION

A total of 152 isolates of marine fungi was found on twigs from beaches in Chanthaburi, Rayong and Chonburi provinces. *Dactylospora haliotrepha*, *Halosphaeria quadricornuta Lulworthia grandispora*, *Torpedospora radiata* and *Verruculina enalia* were common marine fungi, whereas *Camerosporium* sp. was a new record for Thailand.

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