New record of *Chaetomium* species isolated from soil under pineapple plantation in Thailand

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*Chaetomium* species were isolated from soil in pineapple plantations in Phatthalung and Rayong provinces by soil plate and baiting techniques. Taxonomic study was based on available dichotomously keys and monograph of the genus. Five species are recorded as follows: *C. aureum*, *C. bostrychodes*, *C. cochliodes*, *C. cupreum* and *C. gracile*. Another four species are reported to be new records in Thailand as follows: *C. carinthiacum*, *C. flavigenum*, *C. perlucidum* and *C. succineum*.

Key words: *Chaetomium*, Taxonomic study

Introduction

*Chaetomium* is a fungus belonging to Ascomycota of the family Chaetomiaceae which established by Kunze in 1817 (von Arx *et al*., 1986). *Chaetomium* Kunze is one of the largest genera of saprophytic ascomycetes which comprise more than 300 species worldwide (von Arx *et al*., 1986; Soytong and Quimio, 1989; Decock and Hennebert, 1997; Udagawa *et al*., 1997; Rodriguez *et al*., 2002). Approximately 20 species have been recorded in Thailand (Table 1). *Chaetomium* species are well known as coprophilous, seed and soil fungi (Somrithipol, 2004; Somrithipol *et al*., 2004), and also found in organic compost (Soytong, 1990). They degrade cellulose and other organic material and act as antagonist against plant fungal pathogens (Soytong, 2001). *C. globosum* is reported by several researchers to be a strong cellulose decomposer

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(Umikalsom, et al., 1997; Umikalsom, et al., 1998) and expressed a very effective antagonist of various soil microorganisms (Aggarwal, et al., 2004; Dhingra, et al., 2003; Soytong et al., 2001). In Thailand Chaetomium species were screened for using as antagonist in 1989 (Soytong et al., 2001). It has also been reported that some isolates of C. globosum produce antibiotics that can suppress damping-off of sugar beet caused by Pythium ultimum (Di-Pietro et al., 1991). C. cupreum and C. globosum have been reported to reduce leaf spot disease of corn caused by Curvularia lunata, rice blast caused by Pyricularia oryzae, sheath blight of rice caused by Rhizoctonia oryzae and tomato wilt caused by Fusarium oxysporum f.sp. lycopersici (Soytong, 1992a, 1992b).

Moreover, Chaetomium species are noted for their secondary metabolite content with biological activities. Several types of compounds have been investigated from Chaetomium spp. e.g. benzoquinone derivatives (Brewer et al., 1986), a new anthraquinone-chromanone compound named chaetomanone and seven known compounds, ergosterol, ergosteryl palmitate, chrysophanol, chaetoglobosin C, alternariol monomethyl ether, echinuline and isochaetoglobosin D were found from C. globosum KMITL-N0802 and also reported that chaetomanone and echinulin showed activity towards Mycobacterium tuberculosis (Kanokmedhakul et al., 2001). Three new azaphilones named rotiorinols A-C, two new stereoisomers, (-)-rotiorin and epi-isochromophilone II and a known compound, rubrorotiorin, were isolated from the fungus C. cupreum CC3003 of which compounds, rotiorinols A, rotiorinols C, (-)-rotiorin and rubrorotiorin act as antifungal activity against Candida albicans (Kanokmedhakul et al., 2006). Four new dimeric spiro-azaphilones, cochliodones A-D, two new azaphilones, chaetoviridines E and F, a new epi-chaetoviridin A, and known compounds, chaetoviridin A, ergosterol, chaetochalasin A were isolated from C. cochlidiodes VT 01 and C. cochlidiodes CTh 05. Chaetoviridines E and chaetochalasin A exhibited antimalarial activity against Plasmodium falciparum while cochliodones C, chaetoviridines E and F, chaetochalasin A expressed antimycobacterial activity against M. tuberculosis. Furthermore, C. cochlidiodes VT 01 and C. cochlidiodes CTh 05 were reported to be antagonistic to Fusarium oxysporum f.sp lycopersici causing tomato wilt (Phonkerd et al., 2008), Chaetominedione is reported as a new tyrosine kinase inhibitor isolated from the algicolous marine fungus Chaetomium sp.(Abdel-Lateff, 2008) etc.

Chaetomium species are traditionally identified by morphological data, the type of terminal hair and lateral hairs or ascomatal hairs (straight, hooked, spiral, coiled etc.) covering the ascomata, the shape and size of asci and ascospores according to von Arx et al. (1986) and Seth (1970).
The objective of this research was to investigate the species of *Chaetomium* isolated from soil in pineapple plantations from Phatthalung and Rayong provinces, Thailand.

**Table 1.** List of *Chaetomium* species in Thailand.

<table>
<thead>
<tr>
<th>Species</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>C. ampullare</em> Chivers</td>
<td>Soytong, 1991</td>
</tr>
<tr>
<td><em>C. apiculatum</em> Lodha</td>
<td>Udagawa, 1973</td>
</tr>
<tr>
<td><em>C. aureum</em> Chivers</td>
<td>Soytong, 1991; Petcharat and Soytong, 1991</td>
</tr>
<tr>
<td><em>C. bostrychodes zopf</em></td>
<td>Soytong, 1991</td>
</tr>
<tr>
<td><em>C. cocholiodes</em> Palliser</td>
<td>Soytong, 1991</td>
</tr>
<tr>
<td><em>C. cupreum</em> Ames</td>
<td>Soytong, 1991; Petcharat and Soytong, 1991;</td>
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<tr>
<td></td>
<td>Somrithipol, 2004</td>
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<tr>
<td><em>C. deceptivum</em> Malloch &amp; Benny</td>
<td>Soytong, 1991</td>
</tr>
<tr>
<td><em>C. floriforme</em> Gené &amp; Guarro</td>
<td>Gené and Guarro, 1996</td>
</tr>
<tr>
<td><em>C. fusiforme</em> Chivers</td>
<td>Petcharat and Soytong, 1991</td>
</tr>
<tr>
<td><em>C. globosum</em> Kunze</td>
<td>Soytong, 1991; Petcharat and Soytong, 1991;</td>
</tr>
<tr>
<td></td>
<td>Somrithipol, 2004; Somrithipol et al., 2004</td>
</tr>
<tr>
<td><em>C. gracile</em> Udagawa</td>
<td>Soytong, 1991</td>
</tr>
<tr>
<td><em>C. hamadae</em> (Udagawa) v. Arx</td>
<td>Soytong, 1991</td>
</tr>
<tr>
<td><em>C. homopilatum</em> Omvik</td>
<td>Soytong, 1991</td>
</tr>
<tr>
<td><em>C. indicum</em> Corda</td>
<td>Somrithipol et al., 2004</td>
</tr>
<tr>
<td><em>C. longicolleum</em> Krezm. &amp; Badura</td>
<td>Soytong, 1991</td>
</tr>
<tr>
<td><em>C. lucknowense</em> Rai &amp; Tewari</td>
<td>Soytong, 1991; Petcharat and Soytong, 1991</td>
</tr>
<tr>
<td><em>C. malaysiense</em> v. Arx</td>
<td>Soytong, 1991</td>
</tr>
<tr>
<td><em>C. megasporum</em> Sorgel</td>
<td>Soytong, 1991</td>
</tr>
<tr>
<td><em>C. seminudum</em> Ames</td>
<td>Soytong, 1991</td>
</tr>
<tr>
<td><em>C. thermophilum</em> La Touche</td>
<td>Somrithipol, 2004</td>
</tr>
<tr>
<td><em>C. tortile</em> Bainier</td>
<td>Somrithipol et al., 2004</td>
</tr>
<tr>
<td><em>C. variosporum</em> Udagawa et Horie</td>
<td>Udagawa, 1973</td>
</tr>
<tr>
<td><em>C. venezuelense</em> Ames</td>
<td>Udagawa, 1973</td>
</tr>
<tr>
<td><em>C. vitellinum</em> Carter</td>
<td>Soytong, 1991</td>
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</tbody>
</table>

**Materials and methods**

**Source of isolates**

Soil samples for the recovery of *Chaetomium* spp. were collected from pineapple plantations in Phatthalung and Rayong provinces, Thailand, during August to November 2007. Soil samples were kept in clean plastic bags, brought to the laboratory at King Mongkut’s Institute of Technology Ladkrabang, Bangkok, Thailand.
Isolation and identification

Chaetomium species were originally isolated by soil plate technique and baiting technique according to the method described by Soytong (1989).

Soil plate technique, soil samples were dried and ground to fine particles; 0.005-0.015 g of each soil sample were placed to sterilized Petri dishes and then overlaid with glucose-ammonium nitrate agar (GANA) medium (10 g glucose, 1 g NH₄NO₃, 1 g Difco bacto yeast extract, 0.5 g K₂HPO₄, 0.5 g MgSO₄.7H₂O, 20 g agar, 0.06 g rose bengal, 0.03 g streptomycin, 1,000 ml distilled water). After 2-7 d incubation at room temperature in the dark, Chaetomium spp. were observed under stereo microscope and isolated into pure culture by single spore isolation.

Baiting technique, each soil sample (ca 10 g) were placed to sterilized Petri dishes and moistened with sterile distilled water before baited with small pieces of sterilized straws, filter paper, tissue paper and pineapple leaves. After 21 d incubation at room temperature, Chaetomium spp. on baits were daily observed and picked their ascomata to glass slide with small amount of sterilized water before spread on water agar (WA) in a 9-cm-diameter Petri dish. The WA plates were incubated for 12 h at room temperature, then single colony was transferred onto PDA plates and isolated into pure culture. All isolates were kept in culture collection, Herbarium of Thai Mycological Association (H-TMA) at King Mongkut’s Institute of Technology Ladkrabang, Thailand.

Results and discussion

Isolation and identification

Thirty isolates of Chaetomium were obtained in pure culture and identified into 9 species as presented in Table 1 and Fig.1-9. C. cupreum was the most common species which was found in soil from pineapple plantations taken from both Phatthalung and Rayong provinces. The most isolates were obtained by baiting technique, except C. cupreum S1 which was found by soil plate technique.

Identification of Chaetomium species are usually considered morphological characters (Arx et al., 1986; Seth, 1970; Soytong and Quimio, 1989; Gené and Guarro, 1996; Rodríguez et al., 2002) and molecular methods were used in the taxonomy of Chaetomium by Lee and Hanlin (1999). In the GenBank database (2008) sequences of 29 identified and of 44 unidentified Chaetomium species are now deposited. It is needed to do more identification
work both morphological and molecular data to confirm species in the near future.

**Table 2.** *Chaetomium* species isolated from soil in pineapple plantation at different locations in Thailand.

<table>
<thead>
<tr>
<th>Methods</th>
<th>Species</th>
<th>Phatthalung Isolates</th>
<th>Rayong Isolates</th>
</tr>
</thead>
<tbody>
<tr>
<td>soil plate</td>
<td><em>C. cupreum</em></td>
<td>S1, MB601, MB608, MB603, MB103</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td><em>C. aureum</em></td>
<td>MB601, MB608, MB603, MB103</td>
<td>RY102</td>
</tr>
<tr>
<td></td>
<td><em>C. bostrychodes</em></td>
<td>PR1, PR2, PR3, NB701</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td><em>C. carinthiacum</em></td>
<td>NB501</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td><em>C. cochlodes</em></td>
<td>-</td>
<td>RY301</td>
</tr>
<tr>
<td>baiting</td>
<td><em>C. cupreum</em></td>
<td>NB201, MB303, MB301, V4B1</td>
<td>RY201, RY202, RY203, RY204</td>
</tr>
<tr>
<td></td>
<td><em>C. flavigenum</em></td>
<td>MB607, MB402, MB606, MB611, MB604</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td><em>C. gracile</em></td>
<td>NB401, MB605</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td><em>C. perlucidum</em></td>
<td>NB202, NB501</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td><em>C. succineum</em></td>
<td>MB305, MB304</td>
<td>-</td>
</tr>
</tbody>
</table>

Five species are recorded as follows:-


Young colonies usually are white by aerial mycelium. Mature colonies become red by a red pigment exudate. Ascomata are pale green, ovate in shape, 78.5-142.6 x 90.6-180.3 μm. Ascomatal hairs arcuate, septate. Asci are clavate in shape with 8 ascospores per ascus. Ascospores are elliptical, 5.0-7.7 x 8.5-12.5 μm, with two apical germ pores (Fig.1).

Isolate examined: MB607.


Colonies are rapidly growing, young colonies usually are white by aerial mycelium, occasionally with a purple pigment exudate. Mature colonies become green to brown with ascomata. Ascomata are olivaceous, maturing within 10-14 days, dark green to brown when old, ovate in shape, 190.2-349.8 x 272-419.8 μm. Ascomatal hairs usually spirally coiled. Asci are clavate in shape with 8 ascospores per ascus. Ascospores are lemoniform, 7.5-9.9 x 8.6-11.3 μm, with an apical germ pore (Fig.2).

Isolate examined: PR1.
Fig. 1. *Chaetomium aureum* MB601. A. 10-day-old-culture on PDA, B. ascomata, C. ascomatal hairs, D. young asci, E. 8 ascospores in an ascus, F. ascospores.

Fig. 2. *Chaetomium bostrychodes* PR1. A. 10-day-old-culture on PDA, B. ascomata, C. ascomatal hairs, D. young asci, E. 8 ascospores in an ascus, F. ascospores.

Colonies are rapidly growing, young colonies usually are white by aerial mycelium, occasionally with a purple pigment exudate. Mature colonies become green to brown with ascomata. Ascomata are olivaceous, maturing within 10-14 days, dark green to brown when old, ovate in shape, 107.1-143.1x122.0-209.2 µm. Ascomatal hairs usually irregularly sinuous. Asci are clavate in shape with 8 ascospores per ascus. Ascospores are broadly ovate to lemon-shaped, 4.2-5.9x7.1-10.0 µm, with an apical germ pore (Fig.3).

Isolate examined: RY301.

**Fig.3.** *Chaetomium cochliodes* RY301. A. 10-day-old-culture on PDA, B. ascomata, C. ascomatal hairs, D. young asci, E. 8 ascospores in an ascus, F. ascospores.


Colonies usually are red due to a red pigment exudate. Ascomata are red, maturing within 10-14 days, ovate in shape, 79.7-142.7 x 94.7-151.5 µm. Ascomatal hairs arcuate, apically circinate or coiled, septate. Asci are clavate in shape with 8 ascospores per ascus. Ascospores are reniform, 4.7-6.7 x 6.7-10.0 µm, with a single apical germ pore (Fig.4).

Isolate examined: RY202
Fig. 4. Chaetomium cupreum RY202. A. 10-day-old-culture on PDA, B. ascomata, C. ascomatal hairs, D. young asci, E. 8 ascospores in an ascus, F. ascospores.

Fig. 5. Chaetomium gracile NB401. A. 10-day-old-culture on PDA, B. ascomata, C. ascomatal hairs, D. young asci, E. 8 ascospores in an ascus, F. ascospores.
Colonies usually yellow due to yellow pigment exudates. Ascomata are olivaceous grey, maturing within 10-14 days, ovate in shape, 75.4-161.4x110.2-202.8 µm. Ascomatal hairs arcuate. Asci are clavate in shape with 8 ascospores per ascus. Ascospores are fusiform, 5.1-6.9x12.7-17.7 µm, with two apical germ pores (Fig.5).
Isolate examined: NB401.

Four species are reported to be new records in Thailand as follows:-

Colonies usually are white by aerial mycelium, without a pigment exudate. Mature colonies become green to brown with ascomata. Ascomata are olivaceous, maturing within 10-14 days, dark green when old, ovate in shape, 96.1-146.4x101.8-153.2 µm. Ascomatal hairs irregularly sinuous with roughened hairs. Asci are clavate in shape with 8 ascospores per ascus. Ascospores are elliptical, 4.3-6.2x8.4-11.6 µm, with an apical germ pore (Fig.6).
Isolate examined: NB501.

Colonies usually are white by aerial mycelium, becoming red or orange due to a red pigment exudate. Mature colonies become green to brown with ascomata. Ascomata are olivaceous to brown, maturing within 10-14 days, dark grey-green when old, ovate in shape, 92.5-134.9 x 113.2-190.3 µm. Ascomatal hairs arcuate. Asci are clavate in shape with 8 ascospores per ascus. Ascospores are fusiform, 4.0-6.3 x 6.6-11.2 µm, with two apical germ pores (Fig.7).
Isolate examined: MB601.

Colonies usually are white or greylish by aerial mycelium, without a pigment exudate. Mature colonies dark grey to black with ascomata. Ascomata are grey, ovate in shape, 91.9-145.5x120.1-190.6 µm. Ascomatal hairs undulate and irregularly sinuous. Asci are clavate in shape with 8 ascospores per ascus. Ascospores are elliptical, 3.4-5.5x6.7-9.2 µm, with an apical germ pore (Fig.8).
Isolate examined: NB202.
Fig. 6. *Chaetomium carinthiacum* NB501. A. 10-day-old-culture on PDA, B. ascomata, C. ascomatal hairs, D. young asci, E. 8 ascospores in an ascus, F. ascospores.

Fig. 7. *Chaetomium flavigenum* MB607. A. 10-day-old-culture on PDA, B. ascomata, C. ascomatal hairs, D. young asci, E. 8 ascospores in an ascus, F. ascospores.
Fig. 8. **Chaetomium perlucidum** NB202. A. 10-day-old-culture on PDA, B. ascomata, C. ascomatal hairs, D. young asci, E. 8 ascospores in an ascus, F. ascospores.

Fig. 9. **Chaetomium succineum** MB304. A. 10-day-old-culture on PDA, B. ascomata, C. ascomatal hairs, D. young asci, E. 8 ascospores in an ascus, F. ascospores.
**Chaetomium succineum** Ames. Mycologia. 41: 445 (1949).

Colonies usually are dark green or greyish by aerial mycelium, without a pigment exudate. Mature colonies dark grey to black with ascomata. Ascomata are grey, ovate in shape, 107.1-143.1x122.0-209.2 µm. Ascomatal hairs loosely hairs. Asci are clavate in shape with 8 ascospores per ascus. Ascospores are elliptical, 4.2-5.9x7.1-10.0 µm, with an apical germ pore (Fig.9).

Isolate examined: MB304.

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**References**


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