

A Potential Use of *Talaromyces* species as Biological Agents Against Plant Pathogenic Fungi

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Abstract

Soil samples were collected from agricultural and forest soils in the Eastern and Northern Thailand for isolation of *Talaromyces* spp. The alcohol and heat treatment methods and glucose ammonium nitrate agar media were employed. Identification was based on morphological characters such as colony growth and color on different agar media. Microscopic features were examined using light- and scanning electron microscopes. Seven species of *Talaromyces* were found including *T. austrocalifornicus*, *T. helicus* var. *major*, *T. indigoticus*, *T. rotundus*, *T. wortmannii*, *T. thailandiasis* and *Talaromyces* sp.1 (KUFC 3383). Isolates of these species were used for antagonistic activity tests against six species of plant pathogenic fungi *in vitro*. The results revealed that *T. thailandiasis* could inhibit more than 70 percent mycelial growth of *Colletotrichum capsici*, *Fusarium oxysporum* f.sp. *lycopercisi*, *Phytophthora palmivora* and *Lasiodiplodia theobromae*, whereas it could control nearly 30 percent mycelium growth of *Sclerotium rolfsii* and *Rhizoctonia solani*. Other fungi, including *T. helicus* var. *major*, *T. indigoticus*, *T. rotundus*, *T. wortmannii* and *Talaromyces* sp.1 (KUFC 3383), inhibited 40-55 percent mycelium growth of *C. capsici*, *F. oxysporum* f.sp. *cubense* and *P. palmivora*, but they failed to control *L. theobromae*.

Keywords: *Talaromyces*, soil fungi, taxonomy, antagonistic test, plant pathogenic fungi

Introduction

Talaromyces is an ascomycete erected by Benjamin in 1955 with *T. vermiculatus* as a type species. It belongs to the Class Ascomycetes, Order Eurotiales, Family Trichocomaceae Fischer (syn. Eurotiaceae Clem. & Shear) (Kirk et al., 2001). The anamorphic state most commonly belongs to *Penicillium*, section Biverticillate-Symmetrica (Stolk and Samson, 1972; Pitt et al., 2000; Heredia et al., 2001). *Talaromyces* produces white, yellow to red, soft, globose ascomata. Asci are evanescent, mostly 8 ascospores, globose to subglobose or slightly ellipsoidal, borne in chains. Ascospores are globose or ellipsoidal, smooth or showing various ornamentations (Stolk and Samson, 1972).

Forty six species and 6 varieties of *Talaromyces* were recorded from soil, debris, manure, agricultural and industrial wastes, dung, and food, with a worldwide distribution (Pitt et al., 2000; Samson and Pitt, 2000; Stolk and Samson, 1972). In Thailand, 9 species and 2 varieties of *Talaromyces* were reported (Manoch, 2004; Manoch et al., 2004).

Several species of *Talaromyces* can produce bioactive compounds, such as talarodexines A and B from *T. dextrii* having activity against *Bacillus subtilis* (Suzuki et al., 1992), and *T. convolutus* isolated from barley in Japan can produce talaroconvolutins, which inhibit plant and human pathogenic fungi including *Aspergillus fumigatus*, *A. niger*, *Candida albicans* and *Cryptococcus neoformans* (Suzuki et al., 2000). Several new

compounds were reported from other species of *Talaromyces*, such as wortmanilactones A-D from *T. wortmannii* (Dong *et al.*, 2006) and three new azaphilones, luteusins A-E from *T. luteus* (Yoshida *et al.*, 1996). In addition, *T. flavus* is the most common species of *Talaromyces* from soil with a worldwide distribution and is well known for antagonistic activity against many plant pathogenic fungi including *Alternaria alternata*, *Fusarium moniliforme*, *Magnaporthe grisea*, *Sclerotium rolfsii* and *Verticillium albo-atrum* (Duo-Chuan *et al.*, 2005; Inglis and Kawchuk, 2002; Naraghi *et al.*, 2010).

Relatively little is known of the genus *Talaromyces* in Thailand, especially with respect to diversity, taxonomy, antagonistic activity against plant pathogenic fungi, and the production of secondary metabolites (Manoch *et al.*, 2004; Luangsa-ard *et al.*, 2004). Previous studies were limited in taxonomy and phylogeny. However, Dethoup *et al.*, (2007b) studied the morphology and distribution of *T. flavus* and its potential use as biological control agent against plant pathogenic fungi. Dethoup *et al.* (2006, 2007a) reported novel bioactive compounds from *T. bacillisporus* and *T. thailandiasis*. Therefore, further investigation of *Talaromyces* species is needed in this tropical region to discover new taxa, evaluate some species as potential biological control agents against plant pathogenic fungi, and analyze secondary metabolites for possible use in industrial, pharmaceutical and agricultural enterprises. A prerequisite for all of such studies is the morphological characterization of *Talaromyces* species.

The aims of this study were 1) to study the morphology of seven *Talaromyces* species collected from soil in Thailand, and 2) to test for antagonistic activity of these *Talaromyces* species against five plant pathogenic fungi *in vitro*.

Materials and Methods

Fungal Isolation and Identification

Soil samples were collected from agricultural fields, non-agricultural fields, forests and roadsides. Samples were labeled with locations, dates, and names of the collector and brought to the laboratory for isolation of fungi.

Alcohol Treatment Method

A modification of Warcup and Baker (1963), 0.03 g, soil samples were placed in 65% ethanol for 10-20 min. The liquid was decanted, bits of the treated soil were dispensed into several sterile Petri dishes, and the plates were immediately poured with glucose ammonia nitrate agar. The plates were then placed in covered boxes for incubation in darkness at room temperature. Hyphal tips were transferred onto PDA and maintained as pure cultures for identification.

Heat Treatment Method

A modification of Warcup and Baker (1963), 1 g of soil was placed in a sterile test tube in a water bath at 60-80°C for 20-30 min. Excess water was drained off and soil particles were placed into Petri dishes. The same procedures (1.1) described previously were followed.

Identification of *Talaromyces* Species

(Stolk and Samson, 1972; Ramirez, 1982; Manoch *et al.*, 2004)

Macroscopic Examination

Morphological characteristics of colonies were determined as growth pattern, color and texture on different media, such as CZA, MEA, CYA, CMA, OMA and G25N agar after incubation for 7 to 14 days, at 25°C. Diameters of colonies were measured in mm, most effectively by transmitted light from the reverse side.

Colony characteristics were examined under a stereoscopic microscope and with the naked eye. The microscope was used for assessing texture of colonies and the appearance of penicilli and conidial chains. For judgement of conidial and colony colours, Rayner's "A Mycological Colour Chart" (Rayner, 1970) was employed.

Microscopic Examination

Microscopic characteristics were examined on a slide preparation using sterile distilled water and lactophenol as mounting media under a light microscope (Olympus BH-2 with Normaski Interference Contrast). Camera lucida drawings were employed. Photomicrographs of fungal structures were taken under stereo, light and scanning electron microscopes.

Study of ascospore ornamentation was conducted using Scanning Electron Microscopy. Mature ascomata and ascospores of *Talaromyces* from dry culture agar media were transferred with a fine needle onto double-strick scotch tape on aluminium stubs. The specimens were coated with gold for 5-7 min. and examined in a JEOL JSM 6400 scanning electron microscope (Manoch et al., 2004).

Identification was based on morphological characteristics examined under stereo, light and scanning electron microscopes. *Talaromyces* were identified following the research described in previous reports (Stolk and Samson, 1972; Takada and Udagawa, 1988; Yaguchi et al., 1993).

Antagonistic Tests of *Talaromyces* Species *in vitro*

Seven species of *Talaromyces* spp. were used to test for antagonistic activity against 5 species of plant pathogenic fungi including *Colletotrichum capsici*, *Fusarium oxysporum* f.sp. *lycopersici*, *Lasiodiplodia theobromae*, *Phytophthora palmivora*, *Rhizoctonia solani* and *Sclerotium rolfsii*. Fungi were cultivated as a dual cultures on PDA for 14 days at 28°C. The young mycelium from the colony margin of *Talaromyces* spp. and the specific plant pathogenic fungus was cut with sterile cork borer (0.8 cm diam) and placed on PDA, 7 cm apart. All Petri-dishes were incubated at room temperature (28°C) for 14 days. The inhibition levels were calculated by using the formula: $G_1 - G_2 / G_1 \times 100$ where G_1 = colony radius of plant pathogenic fungi in the control and G_2 = colony radius of plant pathogenic fungi in the dual culture test (Intana et al., 2003). Each treatment was performed with three replicates.

Results and Discussion

Fortyeight isolates of the genus *Talaromyces* comprising 6 species and 1 unidentified species were found from 30 soil samples collected from 17 provinces in Thailand including *T. austrocalifornicus*, *T. helicus* var. *major*, *T. indigoticus*, *T. rotundus*, *T. wortmannii*, *T. thailandiasis* and *Talaromyces* sp.1 (KUFC 3383). *Talaromyces austrocalifornicus* and *T. indigoticus* are new records for Thailand.

Two isolates of *Talaromyces austrocalifornicus* were found in soil from Northern Thailand, whereas two isolates *T. helicus* var. *major* were found in agricultural soil from Chanthaburi. In addition, *Talaromyces thailandiasis* was isolated from forest soil from Trat and nonagricultural soil from Chiang Mai, whereas only one isolate of *Talaromyces* sp. 1 (KUFC 3383) was found in forest soil from Trat. Among these species, *T. wortmannii* was the most dominate, follow by *T. rotundus* and *T. indigoticus*. Morphological characteristics of anamorph and teleomorph states of each species are described in Table 1 whereas colony characteristics are given below.

Talaromyces austrocalifornicus

Yaguchi et Udagawa (Figures 1A-H)

Strains examined: KUFC 3351 forest soil, Mae Hong Son

Reference: Yaguchi et al., 1993

Stat. Anam. *Penicillium austrocalifornicum* Yaguchi et Udagawa

Colonies on MEA growing moderately, attaining a diameter of 20 mm within 7 days at 25°C, plane, more or less funiculose, with central area raised up to 3-4 mm deep floccose, consisting of a compact mycelial felt, Pure Yellow (Rayner 14); producing young ascomata intermixed with yellow mycelial hyphae; conidiogenesis inconspicuous and sparse; margins entire; exudates as pale yellow drops, abundantly produced in central area; odor musty; reverse Cinnamon (R 62) or Sepia (R 63). Colonies on MEA at 28°C, reaching 22 mm and 35-37 mm in diameter within 7 and 14 days respectively, plane, fasciculate, consisting of a compact basal felt, Pure Yellow (R 14); developing abundant ascomata which form a continuous layer over the entire surface; conidiogenesis absent; reverse Amber (R 47) to Umber (R 9).

Talaromyces helicus

C.R. Benjamin var. *major* Stolk & Samson (Figures 1I-P)

Strains: KUFC 3598 agricultural soil, Chanthaburi

Reference: Stolk and Samson, 1972

Stat. Anam. *Penicillium helicum* Raper & Fennell

Table 1 Comparison of some diagnosis characters between seven species of *Talaromyces*.

<i>Talaromyces</i> spp.	Penicilli	Cleistothecium	Ascus	Ascospores
<i>T. austrocalifornicus</i>	biverticillate, sometime terverticillate or quaterverticillate	globose to subglobose, (150-) 200-300 (-400) μm	globose to ovoidal, 6-7.5 x 4.5-5.5 μm	pale yellow, broadly ellipsoidal, 2.2-3.3 x 2.2-2.5 μm , finely spinulose with sparse spines
<i>T. helicus</i> var. <i>major</i>	biverticillate or monoverticillate	globose to subglobose, (150-) 170-210 μm	broadly subglobose to globose, 6.5-10 x 6-8 μm	broadly ellipsoidal, smooth-walled, 3.3-4 x 2.3-2.6 μm
<i>T. indigotidus</i>	biverticillate, sometimes monoverticillate	globose to subglobose, 250-480 μm	ovoidal or subglobose, 7.5-8.5 x 6-7 μm	ascospores at first hyaline to pale yellow, soon becoming blue, finally indigo-blue, ellipsoidal, 3.5-4.5 x 2.4-2.85 μm , spinose
<i>T. rotundus</i>	monoverticillate	pale orange, globose to subglobose, 400-530 μm	subglobose to globose, 10-11 x 9.5-10 μm	globose, (3.5-) 4-5.5 (-6) μm in diam., spinulose
<i>T. wortmanii</i>	biverticillate	subglobose to globose, variable in size ranging 300-450 (-550) μm	globose to ovoidal, 10-11.5 x 8-9.5 μm	ellipsoidal, 4.67- 5.33 x 3.3-3.5 μm , spinulose
<i>T. thailandiasis</i>	biverticillate	subglobose to ellipsoidal, 200-540 μm	subglobose to globose, 7.5-9.5 μm	broadly ellipsoidal, 3.5-4 x 2-2.5 μm , spinulose
<i>Talaromyces</i> sp.1 (KUFC 3383)	biverticillate	subglobose to ellipsoidal, yellow, (170-) 200-300 μm	subglobose to globose, 10 x 8-10 μm	broadly ellipsoidal, 4-4.5 x 3.5-3.8 μm , microtuberculate to tuberculate

Colonies on CZA growing slowly, attaining a diameter of 15 mm within 7 days at 25°C, plane, consisting of a very thin basal mycelial felt, Straw (R 46); ascomata and conidiogenesis absent; exudates absent; reverse uncolored to Pale Luteous (R 11). Colonies on MEA spreading broadly, reaching a diameter of 65-70 mm within 7 days at 25°C, plane, more or less funiculose, consisting of a compact mycelial felt, producing abundant ascomata over the entire surface, Sulphur Yellow (R. 15) to Pure Yellow (R 14); conidiogenesis inconspicuous and sparse, margins entire; exudates abundant at central area, as pale yellow drops; reverse Luteous (R 11).

Talaromyces indigoticus

Takada et Udagawa (Figures 2A-F)

Strain examined: KUFC 3366 forest soil, Sakon Nakhon

Reference: Takada and Udagawa, 1993

Stat. Anam. *Penicillium indigoticum* Takada et Udagawa

Colonies on MEA growing moderately, attaining a diameter of 25-27 mm within 7 days at 25°C, floccose, plane, very thin, producing limit developing ascomata in the central area, white; conidiogenesis limited; reverse Pale Luteous (R 11). Colonies on MEA at 28°C, reaching a diameter of 35 mm within 7 days, plane, consisting of a thin basal mycelial felt, white; raised to umbonate at the margin; ascomata and conidiogenesis absent; reverse Straw (R 46).

Talaromyces rotundus

C.R. Benjamin (Figures 2G-J)

Strains examined: KUFC 3359 agricultural soil, Chiang Mai

References: Stolk and Samson, 1972

Stat. Anam. *Penicillium rotundum* Raper and Fennell

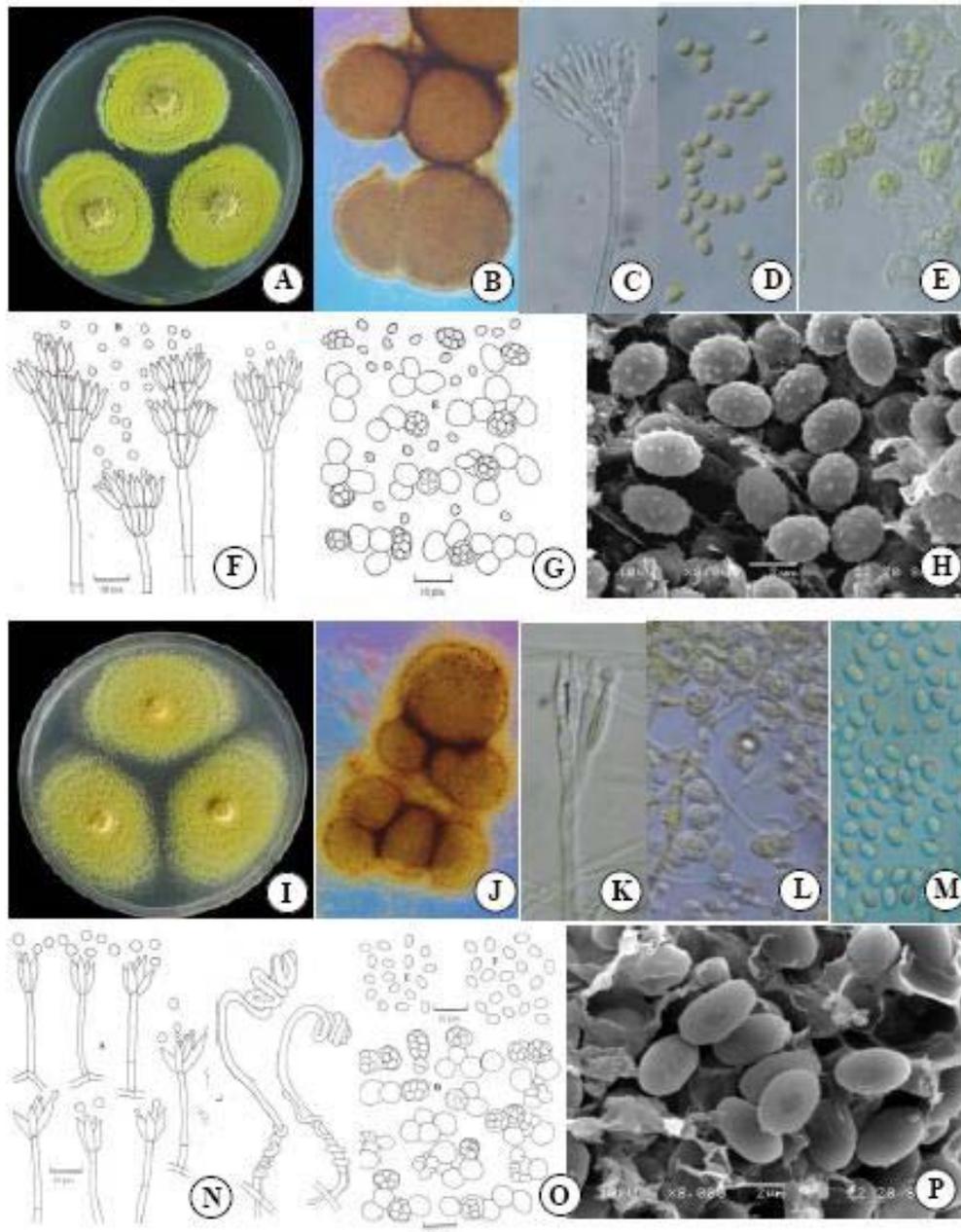


Figure 1 A-H. *T. austrocalifornicus*; colony on MEA, incubated for 7 days at 25°C (A); ascomata (B); penicilli (C); asci and ascospores (D-E); camera lucida drawings of penicilli (F); asci and ascospores (G); SEM of ascospores (H); I-P. *T. helicus* var. *major*; colony on MEA incubated for 7 days at 25°C (I); ascomata (J); penicilli (K); asci and ascospores (L); ascospores (M); camera lucida drawings of penicilli (N); ascumatal initials, asci and ascospores (O); SEM of ascospores (P).

Colonies on CZA growing slowly, attaining a diameter of 15-18 mm within 7 days at 25°C, velvety, plane but centrally wrinkled and sulcate, consisting of a thin basal mycelial felt, Pure Yellow (R 14); ascomata and conidiogenesis absent; exudates and soluble pigment absent; margins entire and white; reverse brown shades ranging

Sienna (R 8) to Umber (R 9). Colonies on MEA growing rapidly, attaining a diameter of 28-30 mm within 7 days at 25°C, plane, funiculose, consisting of a thin basal mycelial felt, Saffron (R 10); ascomata absent; conidiogenesis inconspicuous and sparse; margins white and broad; exudates absent; reverse Ochreous (R 44) to Amber (R 9).

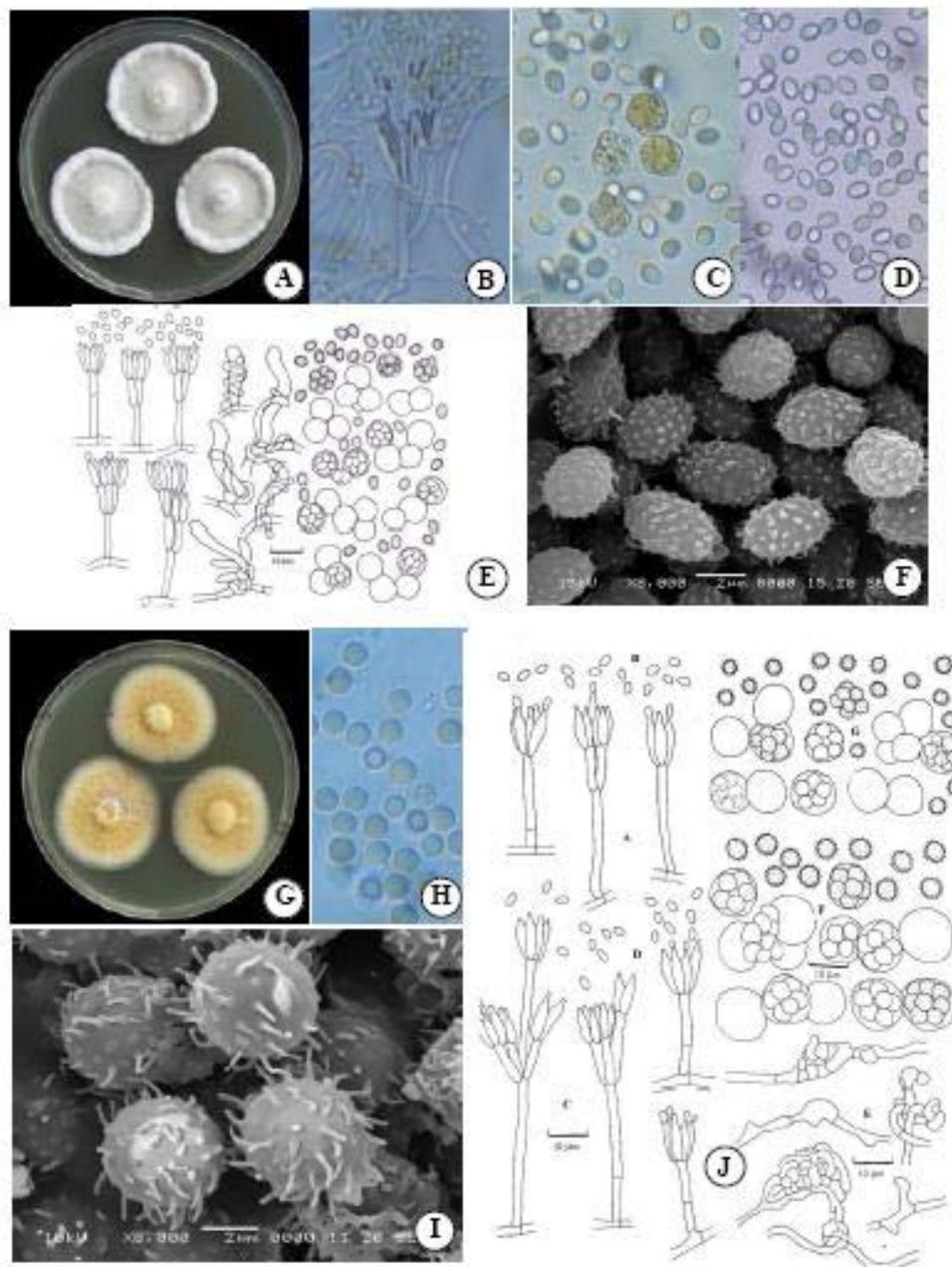


Figure 2 A-F. *T. indigoticus*; colony on MEA, incubated for 7 days at 25°C (A); penicilli (B); ascus and ascospores (C); ascospores (D); camera lucida drawings of penicilli, ascus and ascospores (E); SEM of ascospores (F); G-J. *T. rotundus*; colony on MEA incubated for 7 days at 25°C (G); ascospores (H); SEM of ascospores (I); camera lucida drawings of penicilli; ascumatal initials, asci and ascospores (J).

Talaromyces wortmannii

C.R. Benjamin (Figures 3A-H)

Strains examined: KUFC 3333 forest soil, Mae Hong Son

References: Stolk and Samson, 1972

Stat. Anam. *Penicillium wortmannii* Klöcker

Colonies on MEA growing moderately, attaining a diameter of 21-22 mm within 7 days at

25°C, plane, velvety, more or less wrinkled or radially furrowed, consisting of a compact mycelial felt in which abundant yellow ascomata soon develop with accompanying Greening Glauous (R 91) color from the profuse conidia; margins entire; exudates absent; reverse Pale Luteous (R 11) to Luteous (R 12). Colonies on MEA at 28°C, reaching 35 mm in diameter within 7 days, plane,

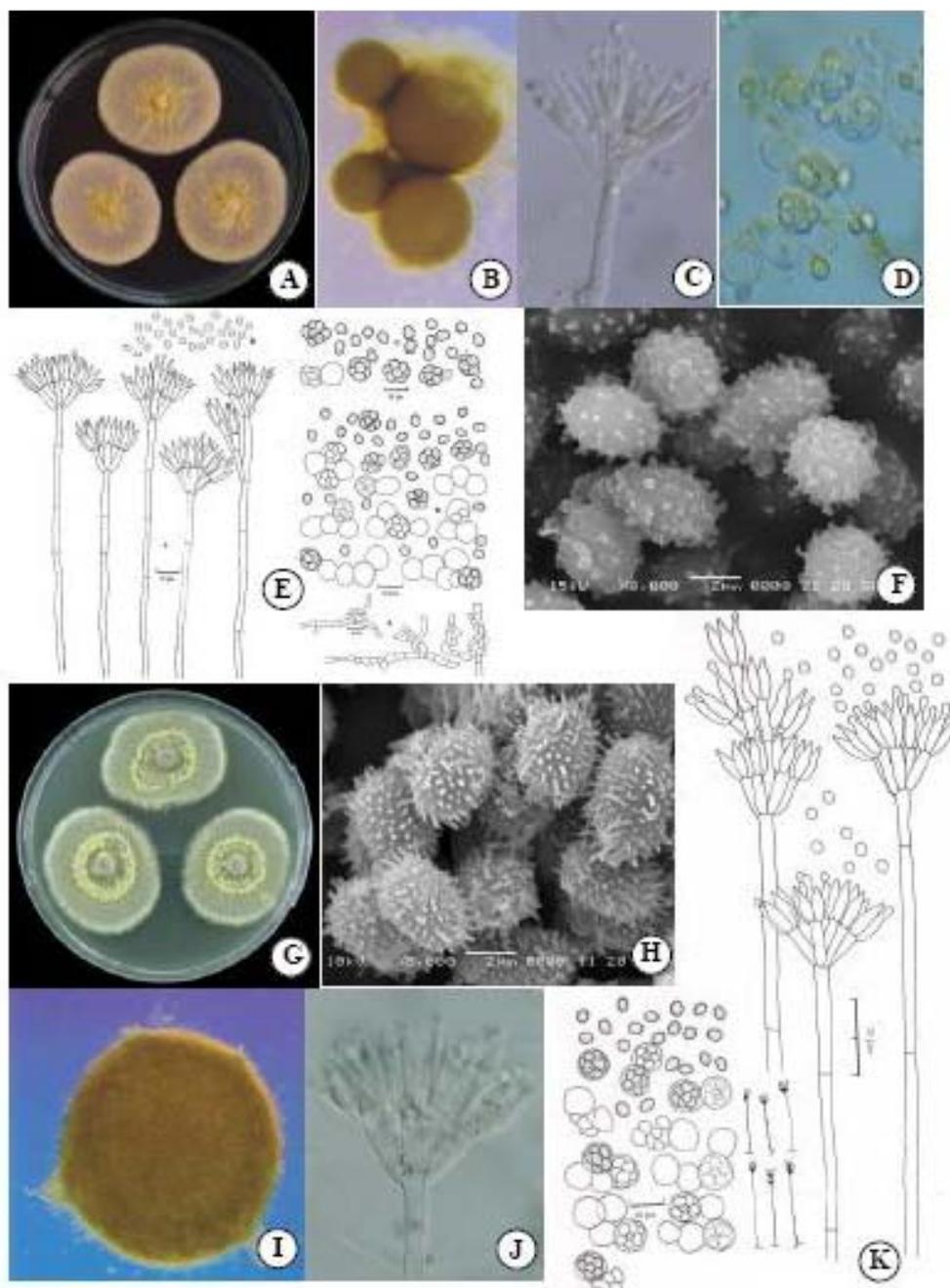


Figure 3 A-H. *T. wortmannii*; colony on MEA, incubated for 7 days at 25°C (A); ascomata (B); penicilli (C); ascus and ascospores (D); camera lucida drawings of penicillin, ascus and ascospores (E); SEM of ascospores (F); G-K. *T. thailandiasis*; colony on MEA incubated for 7 days at 25°C (G); SEM of ascospores (H); ascoma (I); penicilli (J); camera lucida drawings of penicillus, ascum, asci and ascospores (K).

velvety, central colony area lightly wrinkled, consisting of a compact basal felt which numerous ascomata develop, showing Luteous (R 12) to Pale Orange (R 7) shade; conidiogenesis abundant, Pale Greening Glaucous (R 123) color from the profuse conidia; in some strain such as KUFC 3354, producing only umbonate, white mycelium;

margins entire; exudates present as orange drops; reverse Pale Luteous (R 11) to Luteous (R 12).

Talaromyces thailandiasis

(Figures 3G-K)

Strain examined: KUFC 3399 forest soil, Trat Stat. Anam. *Penicillium* sp. 1

Colonies on MEA growing moderately, attaining a diameter of 25-30 mm within 7 days 25°C, velvety, consisting of a thin basal felt, producing abundant conidiogenesis over the entire surface, Pale Olivaceous Grey (R 120); ascomata limited; margins entire, broad and submerged; exudates absent; odor musty; reverse Straw (R 46). Colonies on MEA at 28°C, reaching 30 mm and 45 mm in diameter within 7 and 14 days respectively, velvety, consisting of a thin basal felt, producing moderately ascomata at central area showing Pure Yellow (R 14) shade; conidiogenesis abundant, Pale Olivaceous Grey (R 120); margins entire; exudates absent; reverse Straw (R 46) to Luteous (R 12).

Talaromyces sp. 1

(Figures 4A-F)

Strain examined: KUFC 3383 forest soil, Trat Stat. Anam. *Penicillium* sp. 1

Colonies on MEA growing moderately, attaining a diameter of 22-25 mm within 7 days 25°C, velvety to more or less funiculose, plane,

slightly sulcate, consisting of a compact basal felt, producing moderately developing yellow ascomata in the central area, Pure Luteous (R 11); conidiogenesis abundant produced at the periphery, Pale Greenish Grey (R 123); reverse Pale Luteous (R 11) to Luteous (R 12).

Antagonistic Effect of *Talaromyces* spp. against Plant Pathogenic Fungi *in vitro*

The results reveal that the most effective species was *T. thailandiasis*, which controlled more than 70 % of the mycelium growth of *C. capsici*, *F. oxysporum*, *P. palmivora* and *L. theobromae* and caused moderate inhibition of the radial growth of *R. solani* and *S. rolfsii*. *T. austrocalifornicus* showed low efficacy in controlling mycelium growth of all plant pathogenic fungi (Figure 5A-D).

Most species of *Talaromyces* spp. effectively inhibited mycelial growth of the two plant pathogenic fungi including *C. capsici* and *P. palmivora* on PDA, at 28°C, but failed to inhibit *L. theobromae in vitro* except *T. thailandiasis* and *Talaromyces* sp.1 (KUFC 3383) (Table 2, Figure 5).

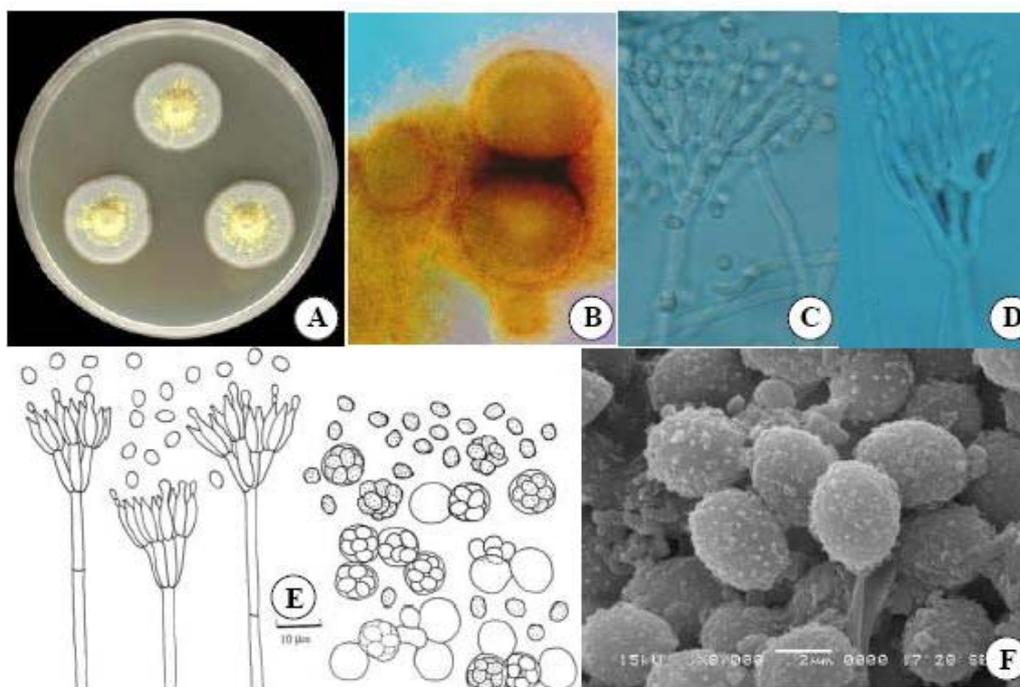


Figure 4 A-F. *Talaromyces* sp. (KUFC 3383); colony on MEA, incubated for 7 days at 25°C (A); ascomata (B); penicilli (C-D); camera lucida drawings of penicilli, asci and ascospores (E); SEM of ascospores (F).

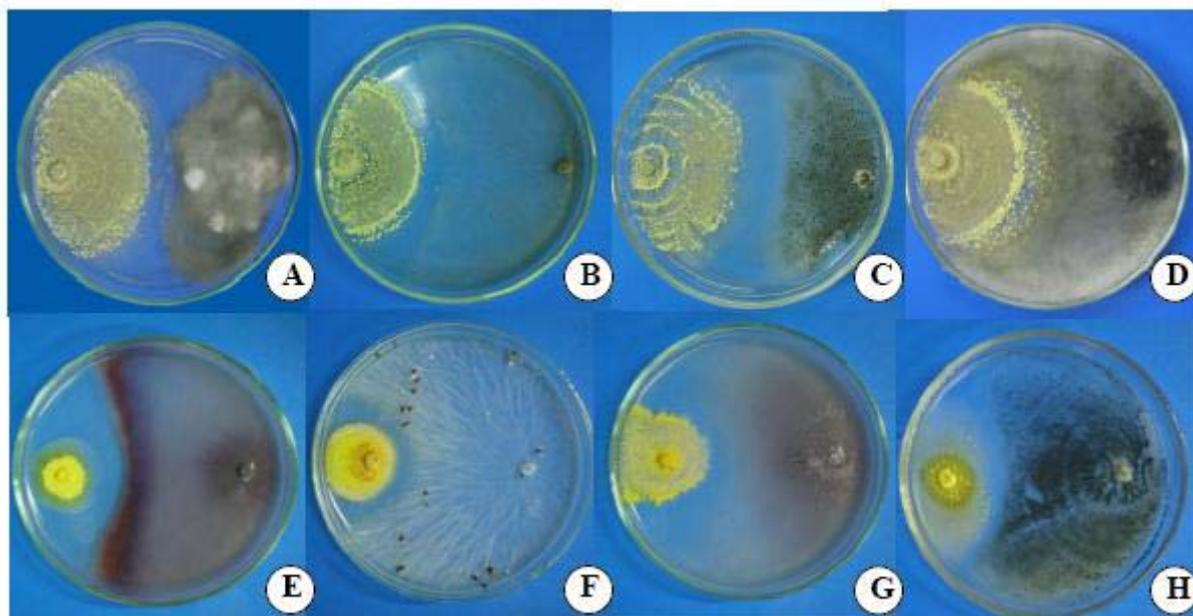


Figure 5 Antagonistic test as dual culture of different *Talaromyces* spp. (left) and plant pathogenic fungi (right) on PDA incubated for 14 days at 28°C. *Talaromyces thailandiasis* vs *H. maydis* (A), *R. solani* (B), *C. capsici* (C), *L. theobromae* (D); *T. austrocalifornicus* vs *F. oxysporum* f.sp. cubense (E), *T. helicus* var. *major* vs *S. rolfsii* (F), *F. oxysporum* f.sp. cubense (G), *T. rotundus* vs *C. capsici* (H).

Table 2 Percent inhibition on mycelial growth of six plant pathogenic fungi by seven species of *Talaromyces* cultivated on PDA as dual culture at 28 °C for 14 days.

Talaromyces species	Inhibition (%)						
	KUFC	<i>Fusarium oxysporum</i> f.sp. <i>lycopersici</i>	<i>Colletotrichum capsici</i>	<i>Lasiodiplodia theobromae</i>	<i>Phytophthora palmivora</i>	<i>Rhizoctonia solani</i>	<i>Sclerotium rolfsii</i>
<i>T. austrocalifornicus</i>	3364	38.89	33.33	∟	50	18.88	27.28
<i>T. helicus</i> var. <i>major</i>	3564	44.44	55.55	-	47.77	-	33.33
<i>T. indigotidus</i>	3363	33.33	55.55	-	50	28.88	-
<i>T. rotundus</i>	3359	33.33	42.22	-	52.22	27.77	27.78
<i>T. wortmanii</i>	3333	27.78	44.44	-	44.44	27.22	-
<i>T. thailandiasis</i>	3399	77.77	77.77	72.22	72.22	27.22	27.78
<i>Talaromyces</i> sp.1	3383	38.89	44.44	33.33	52.22	27.22	27.78

∟ plant pathogenic fungi overgrew the colony of *Talaromyces* spp.

Mycelial growth inhibition for the two basidiomycetous plant pathogenic fungi by *Talaromyces* spp. was less than 30%. Two species, *T. indigotidus* and *T. wortmanii* did not control mycelial growth of *S. rolfsii*, while *T. helicus* var. *major* failed to inhibit *R. solani*.

Most of *Talaromyces* spp. failed to control *L. theobromae*, a pathogen of mangosteen, but could inhibit mycelium growth of other plant pathogenic fungi such as *C. capsici* and *P. palmivora*. Three species of *Talaromyces* spp. inhibited more than 50% of the radial growth of *C. capsici*. causal agent

of anthracnose of chilli, whereas other species produced moderate inhibition of the radial growth of this plant pathogen. In addition, 5 species of *Talaromyces* could inhibit more than 50 % mycelium growth of *P. palmivora*, while *T. helicus* var. *major* and *Talaromyces* sp. (KUFC 3383) caused less than 50% inhibition of radial growth of *P. palmivora*.

From several studies it has been confirmed that *Talaromyces* species have antagonist and biological control potential against plant pathogen especially strains of *T. flavus*. Dethoup et al. (2007b) reported that *T. flavus* isolates could inhibit the mycelial growth of *Phytophthora palmivora*, *P. parasitica*, *Peronophythora litchii*, *Colletotrichum capsici*, *C. gloeosporioides*, *Pestalotiopsis guepinii*, *Phyllosticta* sp., *Curvularia lunata*, *Helminthosporium maydis*, *H. oryzae* and *Fusarium oxysporum*. However, none of the isolates controlled *Pythium aphanidermatum*, *Lasiodiplodia theobromae*, *Rhizoctonia solani* and *Sclerotium rolfsii* *in vitro*. In the present study, many species of *Talaromyces* could inhibit mycelium growth of *C. capsici* and *F. oxysporum*.

Different mechanisms are involved in the biological control of *T. flavus* against *S. rolfsii* and *V. dahliae*. Mycoparasitism is the main mechanism controlling *S. rolfsii*, as indicated by the colonization of its sclerotia and hyphae by *T. flavus* as well as the high positive correlation found between mycoparasitism and disease suppression (Madi et al., 1997). Sclerotia of *S. rolfsii*, mycelium and microsclerotium of *V. dahliae* were very sensitive to the antibiotic activity present in the culture filtrate of *T. flavus*. The antibiotic metabolite(s) secreted by *T. flavus* inhibited melanization of newly formed *V. dahliae* microsclerotia, whereas melanization of newly formed *S. rolfsii* sclerotia was not affected. Although melanization in both fungi was inhibited by H₂O₂, but *V. dahliae* was fourfold more sensitive to inhibit by H₂O₂ as compare with *S. rolfsii* (Madi et al., 1997).

Conclusions

Seven species of *Talaromyces* were found from forest and agricultural soil in Mae Hong Son, Chaingmai, Chanthaburi, Trat and Sakon Nakhon Provinces: *T. austrocalifornicus*, *T. helicus* var.

helicus, *T. indigoticus*, *T. luteus*, *T. rotundus*, *T. wortmanii*, *T. thailandiasis* and *Talaromyces* sp.1 (KUFC 3383). *Talaromyces austrocalifornicus* and *T. indigoticus* were new records for Thailand, and *T. thailandiasis* proved to be new taxon. Molecular analysis is underway to confirm the results from the present morphological study.

The antagonistic activity tests against 6 species of plant pathogenic fungi revealed that 7 species of *Talaromyces* species could moderately inhibit mycelial growth of *F. oxysporum*, *P. palmivora* and *C. capsici*, whereas a new species of *T. thailandiasis* from forest soil in Trat province provided the most effective inhibition of the radial growth of *F. oxysporum*, *C. capsici*, *P. palmivora* and *L. theobromae* and also inhibited mycelial growth inhibition of *S. rolfsii* and *R. solani* to a lesser degree.

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