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Isolation and Identification of Secondary Metabolites of *Clitocybe nuda* Responsible for Inhibition of Zoospore Germination of *Phytophthora capsici*

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ABSTRACT: Secondary metabolites of *Clitocybe nuda* displayed antimicrobial activity against *Phytophthora capsici*. The culture filtrate of *C. nuda* was extracted with ethanol and chromatographically separated on a Sephadex LH-20 column and fractionated on a silica gel column to give eight fractions. These fractions were tested for the ability to inhibit zoospore germination of *P. capsici*. The most active fraction was further purified by silica gel column chromatography to yield three compounds: 2-methoxy-5-methyl-6-methoxymethyl-*p*-benzoquinone (**1**), 6-hydroxy-2*H*-pyran-3-carbaldehyde (**2**), and indole-3-carbaldehyde (**3**), all new to *C. nuda*. At a concentration of 500 mg/L, compound **3** showed complete inhibition of zoospore germination, while compounds **1** and **2** showed inhibition rates of 97 and 86%, respectively. To our knowledge, compound **1** is a newly discovered compound and, for the other two compounds, this is the first report in *C. nuda*. These compounds are potential candidates for new edible fungi-derived pesticides for the control of plant diseases.

KEYWORDS: *Clitocybe nuda, secondary metabolite, antifungal substance, Phytophthora capsici, indole-3-carbaldehyde, p-benzoquinone*

INTRODUCTION

Phytophthora blight, caused by *Phytophthora capsici*, is a devastating disease of peppers. *P. capsici* attacks pepper roots, stems, leaves, and fruits and causes severe yield losses.[1] Its hosts include cucumber, watermelon, and honeydew melon.[1] For the control of this soil-borne disease, crop rotation and chemical application have been widely used. Chemical treatments have shown promising results in the control of the disease.[2] However, the chemicals have also led to the development of fungicide resistance and the environmental pollution, which may pose a serious risk to animal and human health.[3] In recent years, eco-friendly biological control of plant diseases and nonchemical control of crop pests have received great public attention worldwide.

A large number of biologically active compounds are produced by basidiomycetes,[4,5] which possess antibacterial, antifungal, and antiviral activities.[6−8] Several compounds that inhibit the growth of a large spectrum of saprophytic and phytopathogenic fungi have been isolated from wild macrofungi (mushrooms).[4,9,10] The fruiting body and mycelia of *Clitocybe nuda* (wood blewit) have been reported to show antimicrobial activity against the human pathogens *Staphylococcus aureus*[11−13] and *Candida albicans*.[11,12] Sangita and Agate[14] also reported that *C. nuda* showed antibacterial activity against *S. aureus*, and the water-soluble substrates were found to be polyacetylenic compounds by ultraviolet (UV) absorption spectra. Recently, we found that the culture filtrate of *C. nuda* was reported to strongly inhibit certain plant pathogenic fungi and bacteria and to effectively reduce the disease incidences of Phytophthora blight of pepper caused by *P. capsici* and leaf spot of pepper caused by *Xanthomonas axonopodis* pv. *vesicatoria*.[15] The inhibitory substances have molecular weights between 1000 and 100, are negatively charged, and have hydrophilic characteristics. The objective of this study was to identify the key chemical substances in the culture filtrate of *C. nuda* strain LA82 that are responsible for the inhibition of zoospore germination of *P. capsici*.

MATERIALS AND METHODS

General Experimental Procedures. The compounds were identified with the following spectrometric apparatuses. The UV spectra were obtained on a Hitachi U-3210 spectrophotometer (Tokyo, Japan). The infrared (IR) spectra were recorded on a Jasco FT-IR-5300 infrared spectrophotometer (Tokyo, Japan). All of the nuclear magnetic resonance (NMR) spectra were recorded with deuterated solvent on a Varian INOVA Oxford-500 FT-NMR spectrometer (Vernon Hills, IL) and a Varian INOVA Oxford-400 FT-NMR spectrometer (Vernon Hills, IL). Electron impact mass spectrometry (EI−MS) and high-resolution (HR)-EI−MS data were recorded on a JEOL JMS-700 spectrometer (Tokyo, Japan) and a Shimadzu QP2010 mass spectrometer (Tokyo, Japan). Silica gel 60 (230−400 mesh; Merck, Darmstadt, Germany) was used for column chromatography. Thin-layer chromatography (TLC) was carried out on precoated Kieselgel 60 F_{254} (0.2 mm; Merck).

Organisms and Media. *C. nuda* strain LA82 was obtained through breeding for high yield at the Taiwan Agricultural Research Institute and maintained on compost extract agar (CEA), consisting of

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compost extract and 2% agar.[16] For compost extract preparation, 25 g of dried rice straw compost described previously[16] and 100 g of corn meal were boiled in 1 L of water for 20 min and filtered through two layers of cheesecloth. *P. capsici* PCM81 was isolated from infected pepper tissues and maintained on 10% V8 agar consisting of 10% V8 juice, 0.03% $CaCO_3$, and 2% agar.

Spore Production. Sporangia of *P. capsici* PCM81 was produced by growing the organism on 10% V8 agar at 24 °C in a 12 h light−dark cycle, with cool white fluorescent irradiation for 5 days. For the release of zoospores from sporangia, the culture was cut into small pieces (ca. 10 × 10 mm), transferred to 15 mL of sterile distilled water in a 9 cm Petri plate, and incubated under light. After 1 days at 24 °C, the water was replaced with 15 mL of sterile distilled water and the Petri plate was placed in a refrigerator at 4 °C for 1 h. The concentration of zoospore suspension was adjusted to 10^5 spores/mL with a Pipetman microliter pipet.[17]

Production of Culture Filtrate. The *C. nuda* strain LA82 culture block (10 × 10 × 3 mm) was placed in 200 mL of potato dextrose broth (PDB, Difco) in a 500 mL flask, which was incubated at 24 °C for 21 days on a shaker at 120 revolutions per minute (rpm). The culture fluid was harvested by filtration through a Whatman no. 1 filter paper and a 0.22 μm filter (Millipore, Billerica, MA). A total of 2000 mL of 21-day-old culture filtrate of *C. nuda* strain LA82 was concentrated under vacuum and freeze-dried to give a 10 g residue for chemical analysis.

Extraction and Isolation. The 10 g of residue was extracted with 1000 mL of 95% ethanol. The ethanol extract was concentrated under vacuum to produce 1.5 g of crude extract. The crude extract was chromatographically separated on a Sephadex LH-20 column (25−100 μm particle diameters) and eluted with 50% methanol in H_2O to generate two fractions (fractions 1 and 2). Fraction 2 but not fraction 1 showed significant activity and was further chromatographically separated on a silica gel column (230−400 mesh, 1.5 × 60 cm) with *n*-hexane/acetone (2:1) to produce eight subfractions (subfractions 2-1−2-8). Among these eight subfractions, only fractions 2-3 and 2-4 prevented zoospore germination of *P. capsici*. Fraction 2-3 was further chromatographically separated on a silica gel column (230−400 mesh, 1.5 × 30 cm) with *n*-hexane/EtOAc (2:1) to yield fraction 2-3-1 (compound **1**; 14 mg) and fraction 2-3-2 (compound **2**; 1.8 mg). Fraction 2-4 was chromatographically separated on a silica gel column (230−400 mesh, 1.5 × 30 cm) with *n*-hexane/EtOAc (2:3) to yield fraction 2-4-1 (compound **3**; 1.7 mg).

Compound ***1***. Yellow powder. UV (MeOH) λ_{max}: 248, 272, 292, 364, and 416 nm. IR (neat) ν_{max}: 1721, 1640, 1612 cm^{-1}. ^1H NMR ($CDCl_3$, 500 MHz) δ: 1.99 (s, 3, 5-CH_3), 2.28 (s, 3, 6-$CH_2OC\underline{H}_3$), 3.67 (s, 2, 6-$C\underline{H}_2OCH_3$), 3.81 (s, 3, 2-OCH_3), 5.94 (s, 1, H-2). ^{13}C NMR ($CDCl_3$, 125 MHz) δ: 12.8 (5-CH_3), 30.1 (6-$CH_2O\underline{C}H_3$), 40.8 (6-$\underline{C}H_2OCH_3$), 56.2 (2-OCH_3), 107.5 (C-3), 136.2 (C-6), 144.1 (C-5), 158.3 (C-2), 181.4 (C-1), 186.9 (C-4). EI−MS *m*/*z*: 197 $[M + 1]^+$, 133, 119, 97, 69, 57. HR-EI−MS *m*/*z*: 196.0738 (calculated for $C_{10}H_{12}O_4$, 196.0736 $[M]^+$).

Compound ***2***. ^1H NMR ($CDCl_3$, 500 MHz) δ: 4.63 (br s, 2, H-2), 6.46 (d, 1, *J* = 3.6 Hz, H-4), 7.18 (d, 1, *J* = 3.6 Hz, H-5), 9.47 (s, 1, H of aldehyde). ^{13}C NMR ($CDCl_3$, 125 MHz) δ: 57.1 (C-2), 109.9 (C-4), 123.5 (C-5), 152.0 (C-3), 161.0 (C-6), 177.8 (CHO). EI−MS *m*/*z*: 126 $[M]^+$, 109, 97, 69.

Compound ***3***. ^1H NMR ($CDCl_3$, 400 MHz) δ: 7.32 (m, 1, H-5), 7.34 (m, 1, H-5), 7.45 (d, 1, *J* = 8.8 Hz, H-7), 7.85 (d, 1, *J* = 3.2 Hz, H-2), 8.33 (d, 1, *J* = 8.8 Hz, H-4), 8.80 (br s, 1, H-1), 10.1 (s, 1, H of aldehyde). ^{13}C NMR ($CDCl_3$, 100 MHz) δ: 111.5 (C-7), 119.8 (C-3), 122.0 (C-4), 123.1 (C-5), 124.4 (C-9), 124.5 (C-6), 135.2(C-2), 136.1 (C-8), 185.2 (CHO). EI−MS *m*/*z*: 145 $[M]^+$, 116, 103, 73, 59.

Assay of Antimicrobial Activity. The *n*-hexane/acetone (2:1, v/v) extract was separated into eight fractions after evaporation of the solvents. Each fraction was dissolved in distilled water to give concentrations of 500 and 1000 ppm to assay for the antimicrobial activity. Fractions 2-3 and 2-4 were further separated into three subfractions 2-3-1, 2-3-2, and 2-4-1 by a chromatographic technique as described above, and each subfraction was assayed for antimicrobial activity. To assay the antimicrobial activity of the three subfractions against the zoospore germination of *P. capsici* strain PCM81, 10 μL of spore suspension was mixed with an equal volume of culture fraction in a well of a sterile eight-cavity slide. The slides were kept moist by placing them on L-shaped glass rods on moistened paper towels in 9 cm plastic Petri dishes sealed with Parafilm. A spore suspension mixed with distilled water was used as a control. Zoospore germination was recorded after incubation at 24 °C for 8 h, and 100 spores were counted for each of the four replicates. All experiments were repeated twice.

Statistical Analysis. Data were analyzed with Duncan's multiple range test using a standard statistical package (SAS/STAT) (version 9.1, SAS Institute, Inc., Cary, NC) to assess the statistical significance between each column.

RESULTS AND DISCUSSION

Antimicrobial Activities of the Extract. The *C. nuda* partially purified subfractions were extracted with *n*-hexane/acetone (2:1, v/v) and subjected to antimicrobial activity assays. Eight subfractions (subfractions 2-1−2-8) were separated from fraction 2 and adjusted to 1000 ppm for assay of zoospore germination. The results showed that fractions 2-3 and 2-4 exhibited excellent antimicrobial activities against *P. capsici* with 100% inhibition at 1000 ppm, while the other samples were innocuous to *P. capsici*. Subfraction 2-3-1 was separated on a silica gel column with *n*-hexane/acetone (2:1, v/v) to yield 14 mg of compound **1** and 1.8 mg of compound **2**. Compounds **1** and **2** showed strong antimicrobial activity at 500 ppm, reducing the germination from 77% in the control to 0 and 14.5%, respectively (Table 1). Subfraction 2-4-1 was separated on a silica gel column with *n*-hexane/EtOAc (2:3) to yield 1.7 mg of compound **3**. Compound **3** showed complete inhibition of zoospore germination at 500 ppm (Table 1).

Table 1. Antifungal Activities of Three Chromatographic Subfractions of the Culture Filtrate of *C. nuda* Strain LA82 on Zoospore Germination of *P. capsici*

	zoospore germination (%)		
compound	1000 ppm	500 ppm	100 ppm
1	0 b[a]	3.0 c	65.0 a
2	0 b	14.5 b	77.0 a
3	0 b	0.0 c	57.0 b
control	78.0 a	77.0 a	75.0 a

[a]Means within the same column followed by the same letter are not significantly different at $p = 0.05$ according to Duncan's multiple range test.

Structural Characterization of Compound 1. Compound **1** was obtained as a yellow powder. HR-EI−MS of compound **1** showed a molecular ion peak at *m*/*z* 196.07382, which corresponded to a molecular formula of $C_{10}H_{12}O_4$. The IR absorption peaks were indicative of a C═O group (1721 cm^{-1}), a C═C double bond (1640 and 1612 cm^{-1}), and the presence of a *p*-benzoquinone moiety.[18] UV absorptions were observed at 248, 272, 292, 364, and 416 nm. The ^1H NMR spectrum of compound **1** showed a low field signal at δ 5.94 (s, 1, H-3), which indicated a tri-substituted *p*-benzoquinone. A methyl proton signal at δ 1.99 (s, 3, 5-CH_3), two methoxy proton signals at δ 2.28 (s, 3, 6-$CH_2OC\underline{H}_3$) and 3.81 (s, 3, 2-OCH_3), and an oxygen-bearing methylene proton signal at δ 3.67 (s, 2, 6-$C\underline{H}_2OCH_3$) were also observed in the ^1H NMR spectrum. The ^{13}C NMR spectrum of compound **1** showed two carbonyl carbons at δ 181.4 and 186.9, three quaternary carbons, a tertiary carbon at δ 107.5, a secondary carbon at δ

40.8, and three primary carbon at δ 12.8, 30.1, and 56.2, which indicated a *p*-benzoquinone ring substituted with a methyl group, a methoxy group, and a methoxymethyl group. The nuclear Overhauser effect spectrometry (NOESY) cross-peak of H-3 at δ 5.94 with 2-OCH_3 and 6-$C\underline{H}_2OCH_3$ at δ 3.67 with 5-CH_3 and 6-$CH_2OC\underline{H}_3$ was observed. The heteronuclear multiple-bond correlation (HMBC) of H-3 at δ 5.94 with C-2, C-4, and C-5, 2-$OC\underline{H}_3$ at δ 3.81 with C-2, 5-$C\underline{H}_3$ at δ 1.99 with C-4 and C-5, 6-$C\underline{H}_2OCH_3$ at δ 3.67 with C-1 and C-5, and 6-$CH_2OC\underline{H}_3$ at δ 2.28 with 6-$\underline{C}H_2OCH_3$ were observed (Figure 1). The substitute groups were determined to be at C-

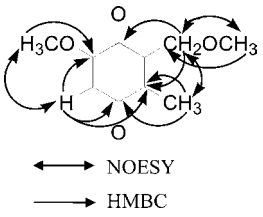

Figure 1. NOESY and HMBC of compound **1**.

2, C-5, and C-6 by NOESY and HMBC experiments. On the basis of the above evidence and literature data,[19−21] the structure of compound **1** was determined to be 2-methoxy-5-methyl-6-methoxymethyl-*p*-benzoquinone (Figure 2), which is

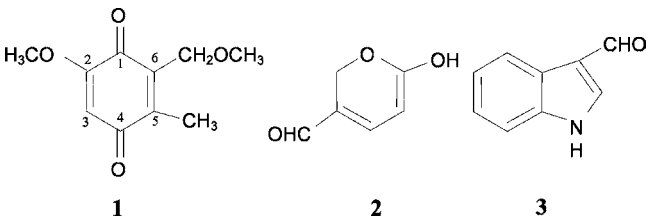

Figure 2. Structures of compounds **1**−**3**: compound **1**, 2-methoxy-5-methyl-6-methoxymethyl-*p*-benzoquinone; compound **2**, 6-hydroxy-2*H*-pyran-3-carbaldehyde; and compound **3**, indole-3-carbaldehyde.

a new natural product. *p*-Benzoquinone or 1,4-benzoquinone is the basic structure of quinonoid compounds. Many studies indicated that 1,4-benzoquinone chemical derivatives exhibit excellent pharmacological applications, such as antibacterial,[22] antitumor,[23] cytotoxic and antioxidant,[24] anticoagulant,[25] and herbicidal[26] activities. A large number studies of *p*-benzoquinone compounds including occurrence in nature or syntheses have been reported.[27] To our knowledge, this is the first report of a new compound with strong inhibitory activity against an oomycete plant pathogen.

Structural Characterization of Compound 2. Compound **2** was identified by NMR and MS analyses, compared to literature data,[28,29] and identified as 6-hydroxy-2*H*-pyran-3-carbaldehyde (Figure 2). 6-Hydroxy-2*H*-pyran-3-carbaldehyde has been isolated from a traditional medicinal plant *Crinum yemense*, and this compound is a tyrosinase inhibitor, which could be used as a skin-whitening agent.[29] This compound was discovered for the first time from the secondary metabolites of *C. nuda* and other microorganisms as well. It exhibited strong antibiotic activity against zoospore germination of *P. capsici*. Its dimer 6−6′-bis(2*H*-pyran-3-carbaldehyde) isolated from *Pseudallescheria boydii* showed strong inhibitory activity against *Alternaria brassicicola*.[30]

Structural Characterization of Compound 3. Compound **3** was identified by NMR and MS analyses, compared to literature data,[31,32] and identified as indole-3-carbaldehyde (Figure 2). Indole-3-carbaldehyde was isolated previously from extracellular fluids of an unknown fungus YL185 and was found to be a tyrosinase inhibitor, inhibiting oxidation and melanin biosynthesis.[33] Yue et al.[34] also isolated this compound from liquid fermentation cultures of *Epichloë festucae*, which showed antifungal activity against chestnut blight caused by *Cryphonectria parasitica*. Some reports demonstrated that indole compounds can be produced by basidiomycetes.[35−37] The indole compounds and their derivatives possess anti-inflammatory, analgesic therapeutics,[38] and antimicrobial activities against Gram-positive bacteria and fungi.[39] The indole compound found in this study possessed inhibitory activity against zoospore germination of *P. capsici*.

Compounds **2** and **3** were tyrosinase inhibitors and inhibitory to melanin biosynthesis.[29,33] Whether such activity is related to the antimicrobial activity remains to be investigated. Antagonistic microorganisms and their secondary metabolites have been evaluated for their potential to be used as biocontrol agents for plant disease control. The antibacterial and antifungal activities of a broad range of basidiomycetes have been considered to be prolific and easily manipulated sources of antibiotics.[4] The inhibitory substances of *C. nuda* were isolated and identified as three compounds. Molecular formulas of compounds **1**, **2**, and **3** are $C_{10}H_{12}O_4$, $C_{12}H_{10}O_5$, and C_9H_6NO, and molecular weights are 196, 126, and 145, respectively. The results are consistent with the molecular-weight dialysis test. The inhibitors have molecular weights between 1000 and 100 and are negatively charged, and the inhibitors are also not proteins.[15] The three compounds reported in this study are excellent potential candidates for new edible fungi-derived compounds for the biocontrol of plant diseases.

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Notes

The authors declare no competing financial interest.

REFERENCES

(1) Alizadeh, A.; Tsao, P. H. *Phytophthora capsici*. In *Phytophthora Diseases Worldwide*; Erwin, D. C., Ribeiro, O. K., Eds.; American Phytopathological Society (APS) Press: St. Paul, MN, 1996; pp 262−268.

(2) Hwang, B. K.; Kim, D. H. Phytophthora blight of pepper and its control in Korea. *Plant Dis.* **1995**, *79*, 221−227.

(3) Muto, M.; Takahashi, H.; Ishihara, K.; Yuasa, H.; Huang, J. W. Antimicrobial activity of medicinal plant extracts used by indigenous people in Taiwan. *Plant Pathol. Bull.* **2005**, *14*, 13−24.

(4) Anke, T. Basidiomycetes: A source for new bioactive secondary metabolites. *Prog. Ind. Microbiol.* **1989**, *27*, 51−66.

(5) Arnone, A.; Nasini, G.; Vajna de Pava, O. Marasmane sesquiterpenes from the basidiomycete *Clitocybe hydrogramma*. *Phytochemistry* **1997**, *46*, 1099−1101.

(6) Benedict, R. G.; Brady, L. R. Antimicrobial activity of mushroom metabolites. *J. Pharm. Sci.* **1972**, *61*, 1820−1822.

(7) Hautzel, R.; Anke, H. Screening of basidiomycetes and ascomycetes for plant growth regulating substances. Introduction of

the gibberellic acid induced de novo synthesis of hydrolytic enzymes in embryoless seeds of *Triticum aestivum* as a test system. *Z. Naturforsch.* **1990**, *45*, 1093−1098.
(8) Korzybski, T.; Kowszyk, G. Z.; Kurylowicz, W. Antifungal antibiotics (polyenes). In *Antibiotics: Origin, Nature and Properties*; Korzybski, T., Ed.; Oxford: London, U.K., 1967; pp 769−820.
(9) Anke, T. The antifungal strobilurins and their possible ecological role. *Can. J. Bot.* **1995**, *73*, 940−945.
(10) Anke, T. Strobilurins. In *Fungal Biotechnology*; Anke, T., Ed.; Chapman and Hall: London, U.K., 1997; pp 206−212.
(11) Suay, I.; Arenal, F.; Asensio, F. J.; Basilio, A.; Cabello, M. A.; Díez, M. T.; García, J. B.; Val, A. G.; Gorrochategui, J.; Hernández, P.; Peláez, F.; Vicente, M. F. Screening of basidiomycetes for antimicrobial activities. *Antonie van Leeuwenhoek* **2000**, *78*, 129−139.
(12) Dulger, B.; Ergul, C. C.; Gucin, F. Antimicrobial activity of the macrofungus *Lepista nuda*. *Fitoterapia* **2002**, *73*, 695−697.
(13) Mustafa, Y.; Bilgili, F. Antimicrobial activities of fruit bodies and/or mycelial cultures of some mushroom isolates. *Pharm. Biol.* **2006**, *44*, 660−667.
(14) Sangita, D.; Agate, A. D. Antibacterial activity of some Indian mushrooms. *Int. J. Med. Mushrooms* **2000**, *2*, 141−150.
(15) Chen, J. T.; Huang, J. W. Control of plant diseases with secondary metabolites of *Clitocybe nuda*. *New Biotechnol.* **2009**, *26*, 193−198.
(16) Chen, J. T.; Peng, J. T.; Chen, M. H.; Chien, S. Y. Effects of indoor composting and conventional composting on the yield of *Agaricus bisporus*. *J. Taiwan Agric. Res.* **2010**, *59*, 19−28.
(17) Ko, W. H.; Chase, L.; Kunimoto, R. A microsyringe method for determining concentration of fungal propagules. *Phytopathology* **1973**, *63*, 1206−1207.
(18) Mehrotra, R.; Vishwakarma, R. A.; Thakur, R. S. Anietane diterpenoid from *Coleus zeylanicus*. *Phytochemistry* **1989**, *28*, 3135−3137.
(19) Höfle, G. ^{13}C-NMR-spektroskopie chinoider verbindungen—I: Substituierte *p*-benzochinone und 2-cyclohexenone. *Tetrahedron* **1976**, *32*, 1431−1436.
(20) Kiuchi, F.; Takashima, H.; Tsuda, Y. Dimerization of 2,5-dihydroxybezoquinones in water. *Chem. Pharm. Bull.* **1998**, *46*, 1129−1234.
(21) Machado, G.; Carrera, P.; Pomini, A. M.; Marsaioli, A. J. Chemical defense in harvestmen (arachnida, opiliones): Do benzoquinone secretions deterinvertebrate and vertebrate predators. *J. Chem. Ecol.* **2005**, *31*, 2519−2539.
(22) Lana, E. J. L.; Carazza, F.; Takahashi, J. Antibacterial evaluation of 1,4-benzoquinone derivatives. *J. Agric. Food Chem.* **2006**, *54*, 2053−2056.
(23) Kwon, H. C.; Choi, S. U.; Lee, K. R. Phytochemical constituents of *Artemisia stolonifera*. *Arch. Pharm. Res.* **2001**, *24*, 312−315.
(24) Muhammad, I.; Takamatsu, S.; Walker, L. A.; Mossa, J. S.; Fong, H. H. S.; El-Feraly, F. S. Cytotoxic and antioxidant activities of alkylated benzoquinones from *Maesa lanceolata*. *Phytother. Res.* **2003**, *17*, 887−891.
(25) Dowd, P.; Zheng, Z. B. On the mechanism of the anticlotting action of vitamin E quinine. *Proc. Natl. Acad. Sci. U.S.A.* **1995**, *92*, 8171−8175.
(26) Gonzalez-Ibarra, M.; Farfán, N.; Trejo, C.; Uribe, S.; Lotina-Hennsen, B. Selective herbicide activity of 2,5-di(benzylamine)-*p*-benzoquinone against the monocot weed *Echinochloa crusgalli*. An in vivo analysis of photosynthesis and growth. *J. Agric. Food Chem.* **2005**, *53*, 3415−3420.
(27) Abraham, I.; Joshi, R.; Pardasani, P.; Pardasani, R. T. Recent advances in 1,4-benzoquinone chemistry. *J. Braz. Chem. Soc.* **2011**, *22*, 385−421.
(28) Schulte, K. E.; Rücker, G.; El-Kersch, M. Nicotin and 3-formoyl-4-hydroxy-2*H*-pyranaus *Herpestis moniera*. *Phytochemistry* **1972**, *11*, 2649−2651.
(29) Abdel-Halim, O. B.; Marzouk, A. M.; Mothana, R.; Awadh, N. A new tyrosinase inhibitor from *Crinum yemense* as potential treatment for hyperpigmentation. *Pharmazie* **2008**, *63*, 405−408.
(30) Su, H. J.; Lin, M. J.; Tsou, Y. J.; Ko, W. H. Pseudallin, a new antibiotic produced by the human pathogenic fungus *Pseudallescheria boydii*, with ecological significance. *Bot. Stud.* **2012**, *53*, 239−242.
(31) Bernart, M.; Gerwick, H. W. 3-(Hydroxyacetyl)indole, a plant growth regulator from the Oregon red alga *Prionitis lanceolata*. *Phytochemistry* **1990**, *29*, 3697−3698.
(32) Wu, P. L.; Hsu, Y. L.; Jao, C. W. Indole alkaloids from *Cephalanceropsis gracilis*. *J. Nat. Prod.* **2006**, *69*, 1467−1470.
(33) Shimizu, K.; Geng, X.; Hashiguchi, M.; Suhara, H.; Fukunaga, S.; Yasutake, S.; Kondo, R.; Tsutsui, M.; Sato, I. Indole-3-carbaldehyde: A tyrosinase inhibitor from fungus YL185. *J. Wood Sci.* **2003**, *49*, 349−354.
(34) Yue, Q.; Miller, C. J.; White, J. F.; Richardson, M. D. Isolation and characterization of fungal inhibitors from *Epichloe festucae*. *J. Agric. Food Chem.* **2000**, *48*, 4687−4692.
(35) Turner, W. B. *Fungal Metabolites*; Academic Press: London, U.K., 1983; Vol. *1*, p 446.
(36) Muszyńska, B.; Sułkowska-Ziaja, K.; Ekiert, H. Indole compounds in fruiting bodies of some edible Basidiomycota species. *Food Chem.* **2011**, *125*, 1306−1308.
(37) Muszyńska, B.; Sułkowska-Ziaja, K. Analysis of indole compounds in edible Basidiomycota species after thermal processing. *Food Chem.* **2012**, *132*, 455−459.
(38) Morgan, Y. R.; Turner, P.; Kennedy, B. J.; Hambley, T. W.; Lay, P. A.; Biffin, J. R.; Regtop, H. L.; Warwick, B. Preparation and characterization of dinuclear copper−indomethacin anti-inflammatory drugs. *Inorg. Chim. Acta* **2001**, *324*, 150−161.
(39) Gutierrez-Lugo, M. T.; Woldemichael, G. M.; Singh, M. P.; Suarez, P. A.; Maiese, W. M.; Montenegro, B. N. Isolation of three new naturally occurring compounds from the culture of *Micromonospora* sp. P1068. *Nat. Prod. Res.* **2005**, *19*, 645−652.