JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Fungicidal Property of *Curcuma longa* L. Rhizome-Derived Curcumin against Phytopathogenic Fungi in a Greenhouse

MOO-KEY KIM,† GYUNG-JA CHOI,‡ AND HOI-SEON LEE*,†

Faculty of Biotechnology and Institute of Agricultural Science & Technology, College of Agriculture, Chonbuk National University, Chonju 561-756, Screening Division, Korea Research Institute of Chemical Technology, Taejeon 305-600, South Korea

Fungicidal activity of *Curcuma longa* rhizome-derived materials against *Botrytis cineria*, *Erysiphe graminis*, *Phytophthora infestans*, *Puccinia recondita*, *Pyricularia oryzae*, and *Rhizoctonia solani* was tested using a whole plant method *in vivo*. It was compared with synthetic fungicides and four commercially available compounds derived from *C. longa*. The response varied with the tested plant pathogen. At 1000 mg/L, the hexane extract of *C. longa* showed fungicidal activities against *E. graminis*, *P. infestans*, and *R. solani*, and the ethyl acetate extract of *C. longa* showed fungicidal activities against *B. cineria*, *P. infestans*, *Pu. recondita*, and *R. solani*. Curcumin was isolated from the ethyl acetate fraction using chromatographic techniques and showed fungicidal activities against *P. infestans*, *Pu. recondita*, and *R. solani* with 100, 100, and 63% control values at 500 mg/L and 85, 76, and 45% control values at 250 mg/L, respectively. In the test with components derived from *C. longa*, turmerone exhibited weak activity against *E. graminis*, but no activity was observed from treatments with borneol, 1,8-cineole, sabinene, and turmerone. In comparison, potent fungicidal activity with chlorothalonil against *P. infestans* at 50 mg/L and dichlofluanid against *B. cinerea* at 50 mg/L was exhibited. These results may be an indication of at least one of the fungicidal actions of curcumin derived from *C. longa*.

KEYWORDS: ***Curcuma longa*; curcumin; fungicidal activity; phytopathogenic fungi**

INTRODUCTION

Preharvest losses due to fungal diseases in world crop production may amount to 12% or even more in developing countries (*1*−*4*). Synthetic fungicides have effectively controlled plant diseases for a number of years, but increasing concern over environmental effects of the currently used fungicides has highlighted the need for the development of alternative types of selective control or of methods of crop protection with or without reduced use of conventional fungicides (*2*, *3*). Research into plant-derived fungicides for agriculture is now being intensified as it becomes evident that plant-derived fungicides still have enormous potential to influence modern agrochemical research (*3*). Although it is difficult to define the ecological significance of most of the synthetic fungicides, there is good reason to suppose that the secondary metabolism of plants has evolved to protect them from attack by microbial pathogens (*3*).

Plant extracts may be an alternative to currently used fungicides for controlling phytopathogenic fungi, because they constitute a rich source of bioactive chemicals (*4*, *5*). Since these are often active against a limited number of specific target species, are biodegradable to nontoxic products, and are potentially suitable for use in integrated management programs, they could lead to the development of new classes of possibly safer disease control agents. Therefore, efforts have focused on secondary plant metabolites for potentially useful products as commercial fungicides or as lead compounds (*6*, *7*). In East Asia, the rhizome from *Curcuma longa* L., belonging to the Zingiberaceae family, has long been considered to have natural medicinal properties, for instance, as an analgesic in the treatment of menstrual disorders, rheumatism, and traumatic diseases, because it contains a number of monoterpenoids, sesquiterpenoids, and curcuminoids (*8*). Furthermore, it has been noted that the extract of *C. longa* L. has insecticidal (*9*, *10*), repellent (*11*, *12*), and antifeeding activities (*11*) against some stored-product insects such as *Schistocerca gregaria* Forsk and *Dysdercus koenigiin* Walk (*13*). The insect repellent and antifeeding constituents in *C. longa* L. are turmerone and *ar*-turmerone (*12*, *14*) and curcuminoids (*13*), respectively. However, little work has been done to manage fungi populations or their damage by using the *Curcuma* rhizome despite its excellent pharmacological actions (*8*). In the greenhouse studies described herein, we assessed *in vivo* the fungicidal activities of *C. longa* L. rhizome-derived materials, the synthetic fungicides chlo-

* To whom correspondence should be addressed: Faculty of Biotechnology and Institute of Agricultural Science & Technology, College of Agriculture, Chonbuk National University, Chonju 561-756, South Korea. Phone: +82-63-270-2544. Fax: +82-63-270-2550. E-mail: hoiseon@moak.chonbuk.ac.kr.
† Chonbuk National University.
‡ Korea Research Institute of Chemical Technology.

10.1021/jf0210369 CCC: $25.00
Published on Web 02/14/2003

rothalonil and dichlofluanid, and four commercially available compounds derived from *C. longa* L. (*15*) against six phytopathogenic fungi.

MATERIALS AND METHODS

Chemicals. Borneol, 1,8-cineole, and sabinene were purchased from Fluka Chemical Corp. (Milwaukee, WI). Turmerone was kindly provided by B.-S. Park (Chonbuk National University). Chlorothalonil ($C_8Cl_4N_2$, MW = 265.90) and dichlofluanid ($C_9H_{11}Cl_2FN_2O_2S_2$, MW = 333.24) were purchased from Dongbu HanNong Chemical Co., and all other chemicals were reagent grade.

Extraction and Isolation. The dried rhizome (5 kg) from *C. longa* was purchased from a medicinal herb shop, Kyungdong Market (Seoul, South Korea). It was finely powdered, extracted twice with methanol (3 L) at room temperature (24−26 °C) for 2 days, and filtered. The combined filtrate was concentrated under vacuum at 35 °C to yield ~10%, based on the weight of the dried rhizome. The extract (20 g) was sequentially partitioned into hexane (7.9 g), chloroform (4.8 g), ethyl acetate (3.8 g), butanol (0.7 g), and water (2.8 g) for a subsequent bioassay. The organic solvent portions were concentrated to dryness by rotary evaporation at 35 °C, while the water portion was freeze-dried.

Because of its good fungicidal activity against *Phytophthora infestans*, *Puccinia recondita*, and *Rhizoctonia solani*, the ethyl acetate fraction was chromatographed on a 95 cm × 10 cm (inside diameter) silica gel column (Merck 230−400 mesh, 750 g) and successively eluted with 20, 30, and 40% ethyl acetate/hexane mixtures (15 L each) followed by a 10% methanol/acetone mixture (20 L). Column fractions were analyzed by TLC (silica gel G), and fractions with a similar TLC pattern were pooled. The combined fractions exhibited fungicidal activities against *P. infestans*, *Pu. recondita*, and *R. solani* and were successively rechromatographed on a 100 cm × 4 cm (inside diameter) column with 20 and 25% ethyl acetate/hexane mixtures (30 L each) as the eluent. Fungicidal activity was observed in subfractions, which were further chromatographed on a 70 cm × 2.5 cm (inside diameter) column with 20% (800 mL) and 25% (4 L) acetone/hexane mixtures as the eluent. A preparative HPLC system (Spectra System P2000, Thermo Separation Products) was used for further separation of the constituents. The column was a 300 mm × 19 mm (inside diameter) μ Porasil silica column (Waters) using an ethyl acetate/hexane mixture (4:1, v/v) at a flow rate of 4 mL/min and detected at 242 nm. Finally, a fungicidal principle (56 mg) was isolated.

Structural determination of the active isolate was made by spectroscopic analysis. ^{1}H and ^{13}C NMR spectra were recorded in deuteriochloroform with a Bruker AM-500 spectrometer at 400 and 100 MHz, respectively. Chemical shifts were reported as δ values downfield from an internal standard of Me_4Si. Mass spectra were obtained on a JEOL GSX 400 spectrometer.

***In Vivo* Fungicidal Activity.** Six plant diseases evaluated in this study were rice blast, rice sheath blight, cucumber gray mold, tomato late blight, wheat leaf rust, and barley powdery mildew caused by *Pyricularia grisea*, *R. solani*, *Botrytis cinerea*, *P. infestans*, *Pu. recondita*, and *Erysiphe graminis*, respectively. Except for *Pu. recondita* and *E. graminis*, the others were routinely maintained on potato dextrose agar (PDA) slants and V-8 agar slants, and kept for stock at 4 °C.

The fungicidal activities of test samples were determined by a whole plant method in a greenhouse, as previously described (*7*). The initial concentration of the test solution was 2000 mg/L, at which more than 60% of the compounds exhibited fungicidal activity against six test fungi; further tests employed a dilution sequence of 1000, 500, 250, and 125 mg/L.

To prepare test solutions at a concentration of 2000 mg/L, 100 mg of the test sample was dissolved in 0.5 mL of dimethyl sulfoxide (DMSO), followed by diluting it with 49.5 mL of water containing Tween 20 (250 μg/mL). Fifty milliliters of each test sample solution was sprayed onto two pots on a turntable at the same time. The treated plants were kept in a greenhouse for 1 day, before being inoculated with each pathogen. Controls were sprayed with the Tween 20 solution. All tests were replicated three times.

In a test with rice blast (RCB) caused by *Py. grisea*, rice plants at the second leaf stage (three plants per pot) were sprayed with each test solution. Treated plants were inoculated with a suspension of conidia in distilled water (1×10^6 spores/mL) and kept in a chamber (25 °C) for 24 h under 100% relative humidity (RH). Treated and control plants were then held in a lighted chamber (26 ± 2 °C and 85% RH) for 5 days, and rated for the severity of the disease. For rice sheath blight (RSB) caused by *R. solani*, each test solution was sprayed onto rice plants at the third leaf stage (three plants per pot). The plants were inoculated by injecting the inoculum at the base of the rice plants. Inoculum of *R. solani* was made by inoculating mycelial plugs in wheat bran medium at 25 °C for 7 days, and macerated at a ratio of 500 g of medium-incubated *R. solani* per liter of distilled water into the mixer. Treated and control plants were held in a lighted chamber (28 °C) for 5 days. With cucumber gray mold (CGM) caused by *B. cinerea*, cucumber plants at the first leaf stage (one plant per pot) were sprayed with each test solution. The cucumber was inoculated with conidia (1×10^6 spores/mL) of *B. cinerea* incubated on PDA medium at 20 °C for 15 days by leaf spray and then placed in a chamber (20 °C) for 4−5 days. For tomato late blight (TLB) caused by *P. infestans*, each test solution was sprayed onto tomato plants at the second leaf stage (two plants per pot). The plants were inoculated with a suspension of 1×10^5 zoosporangia/mL made from a 14-day culture of V-8 juice agar medium at 20 °C. They were kept in a chamber (18 °C) for 4 days, and disease ratings were made. For wheat leaf rust (WLR) caused by *Pu. recondita*, wheat plants at the first leaf stage (four plants per pot) were sprayed with each test solution. The plants were sprayed with a suspension (60 mg/100 mL of 250 ppm Tween 20) of uredospores collected from second leaf stage of wheat, and then placed in a moist chamber. One day after inoculation, plants were held in a growth chamber (20 °C and 70% RH). The fungicidal activities of the test samples were determined 10 days after inoculation (DAI). For barley powdery mildew (BPM) caused by *E. graminis*, barley plants with a fully expended first leaf stage (four plants per pot; ϕ = 7.5 cm) were sprayed with a suspension of the test material. Treated plants were dusted with *E. graminis* conidia formed on leaves of barley by the ratio of eight tested pots per maintained pot.

The control effect of test samples on each disease was evaluated with a control value (CV) calculated with the formula CV (%) = $[(A - B)/A] \times 100$, where A and B represent the disease area on the untreated and treated plants, respectively.

Statistical Analysis. Analysis of variance was performed with the PROC GLM procedure (SAS Institute, Cary, NC). If $P > F$ was less than 0.01, means were separated with the least significant difference (LSD) pest at the $P = 0.05$ level.

RESULTS AND DISCUSSION

During the initial experiments, we observed that a methanolic extract of *C. longa* dried rhizomes possessed fungicidal activity against *E. graminis*, *P. infestans*, *Pu. recondita*, and *R. solani* with a 100% control value at a concentration of 2000 mg/L. However, this methanolic fraction exhibited no fungicidal activity against *Pyricularia oryzae*. Further solvent fractionation showed strong fungicidal activities in the ethyl acetate fraction against *P. infestans* and *Pu. recondita* with 100 and 90% control values, respectively, at a concentration of 1000 mg/L and moderate fungicidal activity against *R. solani* at the same concentration (**Table 1**). Furthermore, the hexane fraction had fungicidal activities against *E. graminis*, *P. infestans*, and *R. solani* with 75, 84, and 60% control values, respectively, at 1000 mg/L. However, little fungicidal activity was produced from the other organic solvent fractions (**Table 1**). One active isolate from the ethyl acetate fraction exhibited potent fungicidal activities against *B. cinerea*, *P. infestans*, *Pu. recondita*, and *R. solani* (**Table 2**), and it was characterized by spectroscopic analyses as curcumin (**Figure 1**). The compound was identified on the basis of the following evidence: orange-yellow solid; mp 182−184 °C; EI-MS (70 eV) *m/z* (% relative intensity) M^+

Table 1. Antifungal Activities of *C. longa* L. Rhizome-Derived Materials against Phytopathogenic Fungi *in Vivo*[a]

material	concn (mg/L)	control value[b] (%) RCB	RSB	CGM	TLB	WLR	BPM
methanol extract	2000	0	100	70	100	100	100
	1000	0	88	54	91	90	85
hexane fraction	2000	0	85	0	100	0	100
	1000	0	60	0	84	0	75
chloroform fraction	2000	0	0	0	0	15	0
	1000	0	0	0	0	0	0
ethyl acetate fraction	2000	0	100	65	100	100	0
	1000	0	75	46	100	90	0
butanol fraction	2000	0	0	0	15	0	0
	1000	0	0	0	0	0	0
water fraction	2000	0	20	0	0	0	0
	1000	0	0	0	0	0	0
LSD[c] (0.05)		–	9.7	3.9	9.8	7.4	8.5

[a] Activity is a preventive value (%): 100% for complete killing and 0% for zero killing. [b] RCB, rice blast caused by *Py. grisea* on rice; RSB, rice sheath blight caused by *R. solani* on rice; CGM, cucumber gray mold caused by *B. cinerea* on cucumber; TLB, tomato late blight caused by *P. infestans* on tomato; WLR, wheat leaf rust caused by *Pu. recondita* on wheat; BPM, barley powdery mildew caused by *E. graminis* on barley. [c] Least significant difference.

Table 2. Antifungal Activities of *C. longa* L. Rhizome-Derived Components and Commercial Fungicides against Phytopathogenic Fungi *in Vivo*[a]

material	concn (mg/L)	control value[b] (%) RCB	RSB	CGM	TLB	WLR	BPM
turmerone	1000	0	0	0	0	0	65
	500	0	0	0	0	0	40
curcumin	1000	0	85	70	100	100	0
	500	0	63	40	100	100	0
	250	0	45	15	85	76	0
	125	0	26	0	57	63	0
borneol	1000	0	0	0	0	0	0
	500	0	0	0	0	0	0
1,8-cineole	1000	0	0	0	0	0	0
	500	0	0	0	0	0	0
sabinene	1000	0	0	0	0	0	0
	500	0	0	0	0	0	0
chlorothalonil[c]	50	0	0	0	95	0	0
dichlofluanid	50	0	0	92	0	0	0
LSD[d] (0.05)		–	10.2	5.7	7.9	8.3	10.5

[a] Activity is a preventive value (%): 100% for complete killing and 0% for zero killing. [b] RCB, rice blast caused by *Py. grisea* on rice; RSB, rice sheath blight caused by *R. solani* on rice; CGM, cucumber gray mold caused by *B. cinerea* on cucumber; TLB, tomato late blight caused by *P. infestans* on tomato; WLR, wheat leaf rust caused by *Pu. recondita* on wheat; BPM, barley powdery mildew caused by *E. graminis* on barley. [c] Commercial name. [d] Least significant difference.

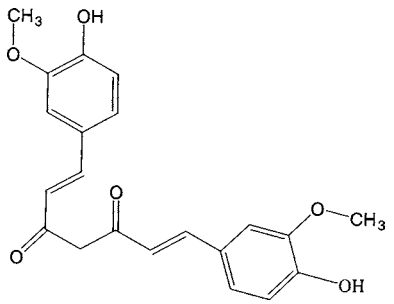

Figure 1. Structure of curcumin isolated from *C. longa* L. rhizome.

368 (63), 350 (58), 320 (22), 272 (20), 232 (23), 217 (27), 177 (100), 145 (49), 137 (48), 117 (18); ^{13}C NMR (acetone-d_6, 100 MHz) δ 184.5, 150.0, 148.8, 141.4, 128.2, 123.8, 122.3, 116.2, 111.6, 101.6, 56.3; CIMS *m*/*z* 369 $(M + 1)^+$; ^{1}H NMR δ 3.89 (s, 6H, 2× OCH_3), 5.88 [s, 1H, C(OH)═CH, enol form], 6.53 (d, 2H, J = 16 Hz, 2,6-H), 6.87 (d, 2H, J = 8 Hz, 5′,5″-H, aromatic), 7.04−7.07 (dd, 2H, J = 2 and 8 Hz, 6′,6″-H), 7.10 (d, 2H, J = 2 Hz, 2′,2″-H aromatic), 7.54 (d, 2H, J = 16 Hz, 1,7-H), 9.25 (2H, Ar-OH); IR (KBr) ν 3427 (O-H str), 2950−3000 (α,β-unsaturated and aryl C-H str), 1287, 1207 cm^{-1} (C-O str). Elemental analysis calculated for $C_{21}H_{20}O_6$: C, 68.47; H, 5.47. Found: C, 68.32; H, 5.29. The spectroscopic analyses of curcumin from *C. longa* are identical to the data of curcumin isolated from *Curcuma zedoaria* (*16*).

The fungicidal activities of curcumin isolated from *C. longa* rhizomes against six phytopathogenic fungi when treated with 1000, 500, 250, and 125 mg/L were determined *in vivo* (**Table 2**). Curcumin exhibited strong and moderate fungicidal activity against *B. cinerea*, *P. infestans*, *Pu. recondita*, and *R. solani in vivo*, except for rice blast caused by *Py. grisea* on rice and barley powdery mildew caused by *E. graminis* on barley. This study is the first to report the fungicidal function of the component isolated from *C. longa* rhizomes. The extract of *C. longa* L. is used in foods as a condiment. It is also used as an essential ingredient in medicine as a carminative, as an anthelmintic, as a laxative, and as a cure for liver ailments (*17*). The use of *C. longa* as an insect repellent, insecticide, and for antibacterial activity has long been known (*12*, *14*, *18*). Among these components, turmeric oils and curcuminoids constitute a major group of secondary metabolites. Turmerone and *ar*-turmerone isolated from *C. longa* L. have been reported to be repellency to *Tribolium castaneum* (Hbst.) (*12*). Furthermore, *ar*-turmerone has potent insecticidal activity against *Nilaparvata lugens* Stål female adults and *Plutella xylostella* L. larvae (*14*). Curcuminoids such as curcumin, demethoxycurcumin, and bismethoxycurcumin have potent antioxidant, antibacterial, antitumorigenetic, and anti-inflammatory properties (*15*−*18*).

To determine the fungicidal activity of other components derived from *C. longa* L., four commercially available compounds derived from this plant species (*15*) against six phytopathogenic fungi were determined (**Table 2**). Turmerone exhibited 65% control values against *E. graminis* when treated with 1000 mg/L. However, little or no activity was observed for borneol, 1,8-cineole, sabinene, and turmerone when treated at 1000 and 500 mg/L. Because of the fungicidal activity of curcumin against *P. infestans*, *Pu. recondita*, and *R. solani*, this compound was compared with chlorothalonil and dichlofluanid as currently used fungicides (**Table 2**). Potent fungicidal activity was observed with chlorothalonil against *P. infestans* at 50 mg/L and dichlofluanid against *B. cinerea* at 50 mg/L, whereas no fungicidal activity was produced for chlorothalonil against *B. cineria*, *Py. grisea*, *R. solani*, and *Pu. recondita* and for dichlofluanid against *Py. grisea*, *P. infestans*, *Pu. recondita*, and *R. solani*. In this study, although curcumin is more than 10 times less active than synthetic fungicides that were used, curcumin may be useful for a lead product for developing new types of fungicides for controlling plant pathogens on crops.

Certain plant extracts and phytochemicals act in many ways on various types of disease complex, and may be applied to the crop in the same way as other agricultural chemicals. They are being considered as potential alternatives for synthetic fungicides (*19*, *20*), or lead compounds for new classes of synthetic fungicides such as podoblastin produced by *Podophyllum peltatum* (*20*, *21*). Several naturally occurring compounds such as tryptanthrin, indole 3-acetonitrile, and *p*-coumaric acid methyl ester have been isolated and identified from woad, *Isatis tinctoria* L., against *Coniophora puteana* Schum.

Fr. Karst (*22*). Various compounds, including phenolics, terpenoids, alkaloids, and lignans, exist in plants (*4*, *8*). On the basis of our results and these earlier findings, the fungicidal activity *in vivo* of curcumin derived from *C. longa* rhizomes may be valuable for a useful lead product of possibly safer disease control agents.

LITERATURE CITED

(1) Agrios, G. N. Significance of plant diseases. In *Plant Pathology*, 4th ed.; Academic Press: San Diego, 1997; pp 25−37.

(2) Lee, S. E.; Kim, J. E.; Lee, H. S. Insecticide resistance in increasing interest. *Agric. Chem. Biotechnol*. **2001**, *44*, 105−112.

(3) Benner, J. P. Pesticidal compounds from higher plants. *Pestic. Sci*. **1993**, *39*, 95−102.

(4) Swain, T. Secondary compounds as protective agents. *Annu. Rev. Plant Physiol*. **1977**, *28*, 479−501.

(5) Wink, M. Production and application of phytochemicals from an agricultural perspective. In *Phytochemistry and Agriculture*; van Beek, T. A., Breteler, H., Eds.; Proceedings of the Phytochemical Society Europe; Clarendon Press: Oxford, U.K., 1993; Vol. 34, pp 171−213.

(6) Balandrin, M.; Klocke, J.; Wurtele, E. S.; Bollinger, W. H. Natural plant chemicals: Sources of industrial and medicinal materials. *Science* **1985**, *228*, 1154−1160.

(7) Lee, H. S.; Lee, S. W.; Cho, K. Y.; Kim, M. K.; Ahn, Y. J. Fungicidal activities of 51 fruit extracts against six phytopathogenic fungi. *Agric. Chem. Biotechnol*. **2001**, *44*, 147−153.

(8) Tang, W.; Eisenbrand, G. *Chinese Drugs of Plant Origin*; Springer-Verlag: Berlin and Heidelberg, Germany 1992; pp 401−415.

(9) Chander, H.; Kulkarni, S. G.; Berry, S. K. Effectiveness of turmeric powder and mustard oil as protectants in stored milled rice against the rice weevil *Sitophilus oryzae*. *Int. Pest Control* **1991**, *33*, 94−97.

(10) Chander, H.; Kulkarni, S. G.; Berry, S. K. Studies on turmeric and mustard oil as protectants against infestation of red flour beetle, *Tribolium castaneum* (Herbst) in stored rice. *J. Insect Sci*. **1992**, *5*, 220−222.

(11) Jilani, G.; Saxena, R. C. Repellent and feeding deterrent effects of turmeric oil, sweetflag oil, neem oil and a neem-based insecticide against lesser grain borer (Coleoptera: Bostrychidae). *J. Econ. Entomol*. **1990**, *83*, 629−634.

(12) Su, H. C. F.; Horvat, R.; Jilani, G. Isolation, purification, and characterization of insect repellents from *Curcuma longa* L. *J. Agric. Food Chem*. **1982**, *30*, 290−292.

(13) Chowdhury, H.; Walia, S.; Saxena, B. S. Isolation, characterization and insect growth inhibitory activity of major turmeric constituents and their derivatives against *Schistocerca gregaria* (Forsk) and *Dysdercus koenigii* (Walk). *Pest Manage. Sci*. **2000**, *56*, 1086−1092.

(14) Lee, H. S.; Shin, W. K.; Song, C.; Cho, K. Y.; Ahn, Y. J. Insecticidal activities of *ar*-turmerone identified *Curcuma longa* rhizome against *Nilaparvata lugens* (Homoptera: Delphacidae) and *Plutella xyostella* (Lepidoptera: Yponomeutidae). *J. Asia-Pacific Entomol*. **2001**, *4*, 181−185.

(15) Gopalan, B.; Goto, M.; Kodama, A.; Hirose, T. Supercritical carbon dioxide extraction of turmeric (*Curcuma longa*). *J. Agric. Food Chem*. **2000**, *48*, 2189−2192.

(16) Syu, W., Jr.; Shen, C. C.; Don, M. J.; Ou, J. C.; Lee, G. H.; Sun, C. M. Cytotoxicity of curcuminoids and some novel compounds from *Curcuma zedoaria*. *J. Nat. Prod*. **1998**, *61*, 1531−1534.

(17) Srimal, R. C. Turmeric: A brief review of medicinal properties. *Fitoterapia* **1997**, *68*, 483−493.

(18) Negi, P. S.; Jayaprakasha, G. K.; Jagan Mohan Rao, L.; Sakariah, K. K. Antibacterial activity of turmeric oil: A byproduct from curcumin manufacture. *J. Agric. Food Chem*. **1999**, *47*, 4297−4300.

(19) Hedin, P. A. New concepts and trends in pesticide chemistry. *J. Agric. Food Chem*. **1982**, *30*, 201−215.

(20) Hostettman, K.; Potterat, O. Strategy for the isolation and analysis of antifungal, molluscicidal, and larvicidal agents from tropical plants. In *Phytochemicals for Pest Control*; Hedin, P. A., Hollingworth, R. M., Masler, E. P., Miyamoto, J., Thompson, D. G., Eds.; ACS Symposium Series No. 387; American Chemical Society: Washington, DC, 1997; pp 14-26.

(21) Miyakado, M. The search for new insecticidal and fungicidal compounds from plants. *J. Pestic. Sci*. **1986**, *11*, 483−492.

(22) Seifert, K.; Unger, W. Insecticidal and fungicidal compounds from *Isatis tinctoria* L. *Z. Naturforsch*. **1994**, *44C*, 44−48.

Received for review October 11, 2002. Revised manuscript received December 31, 2002. Accepted January 2, 2003. This work was supported by a grant from the BioGreen 21 Program, Rural Development Administration, South Korea.

JF0210369