JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Article

pubs.acs.org/JAFC

Acaricidal Potentials of Active Properties Isolated from *Cynanchum paniculatum* and Acaricidal Changes by Introducing Functional Radicals

Min-Gi Kim, Ji-Yeon Yang,* and Hoi-Seon Lee*

Department of Bioenvironmental Chemistry and Institute of Agricultural Science and Technology, College of Agriculture and Life Sciences, Chonbuk National University, Jeonju 561-756, Republic of Korea

ABSTRACT: This study evaluated the acaricidal activities of acetophenone and its derivatives for their potentials as natural acaricides using an impregnated fumigant bioassay against *Dermatophagoides* spp. and *Tyrophagus putrescentiae*. On the basis of the LD_{50} values against *D. farinae*, 3′-methoxyacetophenone (0.41 $\mu g/cm^2$) was 89.9 times more toxic than DEET (36.87 $\mu g/cm^2$), followed by 4′-methoxyacetophenone (0.52 $\mu g/cm^2$), 2′-methoxyacetophenone (0.75 $\mu g/cm^2$), 2′-hydroxy-5′-methoxyacetophenone (1.03 $\mu g/cm^2$), 2′-hydroxy-4′-methoxyacetophenone (1.29 $\mu g/cm^2$), acetophenone (1.48 $\mu g/cm^2$), 2′-hydroxyacetophenone (1.74 $\mu g/cm^2$), 2′,5′-dimethoxyacetophenone (1.87 $\mu g/cm^2$), 2′,4′-dimethoxyacetophenone (2.10 $\mu g/cm^2$), and benzyl benzoate (9.92 $\mu g/cm^2$). In regard to structure−activity relationships between acaricidal activity and functional radicals (hydroxyl and methoxy groups) on the acetophenone skeleton, a monomethoxy group (2′-, 3′-, and 4′-methoxyacetone) on the acetophenone skeleton was more toxic than were the other groups (2′,4′- and 2′,5′-dimethoxyacetophenone, 2′- and 4′-hydroxyacetophenone, 2′-hydroxy-4′-methoxyacetophenone, 2′-hydroxy-5′-methoxyacetophenone, and 4′-hydroxy-3′-methoxyacetophenone). These results indicated that acaricidal activity against three mite species changed with the introduction of functional radicals (hydroxyl and methoxy groups) onto the acetophenone skeleton.

KEYWORDS: *acaricide, 2′-hydroxy-5′-acetophenone, Dermatophagoides farinae, D. pteronyssinus, Tyrophagus putrescentiae*

INTRODUCTION

The prevalence of allergic diseases such as allergic rhinitis, allergic rhinoconjunctivitis, atopic dermatitis, and bronchial asthma has increased not only in developing countries but also in industrialized countries.[1] House dust and stored food mites, factors affecting the development and expression of allergic disease, are typical indoor inhalant allergens.[2] House dust mites, *Dermatophagoides farinae* (Hughes) and *Dermatophagoides pteronyssinus* (Trouessart), are well-known producers of allergens.[3] In addition, stored food mites, *Tyrophagus putrescentiae* (Schrank) from the family Acaridae, are prevalent in barn dust, dried eggs, ham, poultry, stored grains, and straw.[4] These stored food mites may cause acute enteritis, diarrhea, systemic anaphylaxis, and urinary tract infection when contaminated foods are handled, ingested, or inhaled.[5] In this regard, commercial acaricides, such as benzyl benzoate, dibutyl phthalate, and *N*,*N*-diethyl-*m*-toluamide (DEET), have been used to control house dust and stored food mites.[6] However, repeated use of commercial acaricides results in potential problems such as residual toxicity and resistance.[6,7] These problems indicate that development of selective mite control alternatives is needed.[8]

Many studies have sought to develop natural acaricides.[9,10] Chemotherapeutic agents, such as carvacrol, leptospermone, and quinone, have been previously reported to have acaricidal effects against house dust and stored food mites.[11,12] In the Asclepiadaceae family, *Cynanchum paniculatum*, which is chiefly distributed in East Asia (China, Japan, and Korea), has yielded several acetophenones, alkaloids, steroids, and fatty diolefins.[13,14] Acetophenone and its derivatives have shown many interesting pharmacological actions such as analgesic properties and antiaggregatory, anti-inflammatory, antimicrobial, antioxidant, and cytotoxic activities.[15] Relatively few studies have been conducted to evaluate the acaricidal activities of acetophenone derivatives against house dust and stored food mites, despite their biological activities. Although the acaricidal toxicities of acetophenone derivatives containing methyl groups from *Angelicae koreana* roots were evaluated,[16] this study was conducted to assess the acaricidal activities of acetophenone derivatives containing hydroxyl and methoxy groups against house dust and stored food mites. Moreover, the active compound isolated from *C. paniculatum* roots and acetophenone derivatives containing hydroxyl and methoxy groups were compared with the commonly used benzyl benzoate and DEET to determine acaricidal activities and structure−activity relationships.

MATERIALS AND METHODS

Chemicals. Acetophenone, 2′,4′-dimethoxyacetophenone, 2′,5′-dimethoxyacetophenone, 2′-hydroxyacetophenene, 2′-hydroxy-4′-methoxyacetophenone, 2′-hydroxy-5′-methoxyacetophenone, 4′-hydroxyacetophenone, 4′-hydroxy-3′-methoxyacetophenone, 2′-methoxyacetophenone, 3′-methoxyacetophenone, 4′-methoxyacetophenone, myristic acid, and palmitic acid were purchased from Sigma-Aldrich (St. Louis, MO, USA). All other chemicals were of reagent grade.

Gas Chromatography−Mass Spectrometry. The roots of *C. paniculatum* were purchased from a local market in Jeonju, Korea. The

Received: May 28, 2013
Revised: July 10, 2013
Accepted: July 15, 2013
Published: July 15, 2013

dx.doi.org/10.1021/jf402330p | J. Agric. Food Chem. 2013, 61, 7568−7573

roots were washed three times with 500 mL of distilled water, air-dried for 7 days, and then ground to a fine powder. The essential oil (yield = 0.43%) of *C. paniculatum* roots was extracted by steam distillation as previously described[6] and analyzed on a gas chromatograph (HP6890, Agilent)−mass spectrometer (5973IV, Agilent) (GC-MS). Separation was performed on a DB-5 (0.25 mm film) fused silica capillary column (30 m × 0.25 mm i.d.; J&W Scientific, Folsom, CA, USA). GC conditions were as follows: injector temperature, 210 °C; column temperature, isothermal at 50 °C for 15 min, increased by 2 °C/min to 200 °C, and held at this temperature for 15 min; ion source temperature, 230 °C. Helium (He) was used as a carrier gas at a rate of 0.8 mL/min. The effluent of the GC column was introduced directly into the source of the mass spectrometer. Spectra were obtained in the electron ionization (EI) mode at an ionization energy of 70 eV. The sector mass analyzer was set to scan from 50 to 600 amu over 2 s. Identification of compounds was accomplished using retention times and the mass spectra obtained when authentic standards were analyzed using the GC-MS system. When an authentic sample was not available, identification was achieved by comparison with a mass spectra library.

Isolation. The essential oil of *C. paniculatum* (30 g) was isolated by silica gel column chromatography (Merck 70−230 mesh, 600 g, 550 mm i.d. × 700 mm; Rahway, NJ, USA) and successively eluted with hexane/ethyl acetate (4:1 to 1:1, gradient, v/v). Each fraction was loaded on a thin layer chromatography (TLC) plate to identify similar fraction patterns. Ultimately, six fractions were obtained and referred to as CP-1−CP-6. The acaricidal activity of each of the six fractions was evaluated against *D. farinae*, *D. pteronyssinus*, and *T. putrescentiae* at a concentration of 80 $\mu g/cm^2$. CP-3 had potent acaricidal activity against *Dermatophagoides* spp. and *T. putrescentiae*. Therefore, rechromatography of active CP-3 (10.6 g) was conducted on a silica gel column using hexane/ethyl acetate (3:1, v/v), and three fractions (CP-31−CP-33) were obtained. Among these fractions, CP-33 had a potent acaricidal activity against the three mite species. Furthermore, to departmentalize the CP-33, preparative HPLC (Japan Analytical Industry Co., Ltd., Tokyo, Japan) was used. CP-33 (7.1 g) was divided into four fractions (CP-331−CP-334) using a Jaigel GS series column (GS 310 500 mm × 2) with methanol (100%) as the mobile phase at a flow rate of 5 mL/min and with UV detection. CP-332 (2.25 g) had the highest acaricidal activity among these fractions. Next, a Jaigel W series column (W252 500 mm + W253 500 mm) with methanol (100%) as the mobile phase at a flow rate of 3.5 mL/min was used. Finally, the active component (CP-3322, 842 mg) was isolated. The structure of CP-3322 was determined using nuclear magnetic resonance (NMR) spectroscopy. ^1H NMR and ^{13}C NMR were measured using a JNM-EX600 (Jeol Ltd., Tokyo, Japan) spectrometer in deuterochloroform ($CDCl_3$) with tetramethylsilane (TMS) as an internal standard at 600 and 150 MHz, respectively.

Target Mites. The respective cultures of *D. farinae*, *D. pteronyssinus*, and *T. putrescentiae* were maintained without exposure to any known acaricide. These mites were maintained on fry feed 1 (Korea Special Feed Meal Co. Ltd., Jeonju, Korea) and dried yeast (1:1 by weight) and were reared in a plastic case (15 × 12 × 6 cm) containing 30 g of a sterilized diet. Rearing cages were kept in an incubator at 25 ± 1 °C and 75% relative humidity (RH) in continuous darkness. Fry feed consisted of protein (49.0%), lipid (4.0%), cellulose (3.0%), phosphorus (2.0%), and calcium (1.9%).

Bioassay. An impregnated fumigant bioassay was used to determine the acaricidal activities of test materials. This method was slightly modified from the method described by Lee et al.[17] Benzyl benzoate and DEET served as positive control.[18] Different quantities (80, 40, 20, 10, 5, 2.5, 1.0, 0.5, and 0.25 $\mu g/cm^2$) of each test sample were dissolved in acetone, and a sample (20 μL) was applied to individual paper disks (8 mm diameter, 1 mm thickness, Tokyo Roshi, Japan). The negative control disk received only acetone (20 μL). The paper disks were dried in a fume hood for 15 min and then placed in the cap of a microtube (2 mL, Greiner bio-one GmbH, Germany). Thirty individuals of *D. farinae*, *D. pteronyssinus*, or *T. putrescentiae* were placed in each microtube, which was then sealed using the cap containing the treated paper disks. The treated and control mites were maintained at 25 ± 1 °C and 75% relative humidity in darkness for 24 h.

Statistical Analysis. Twenty-four hours after treatment, mortalities of each group were determined under a binocular microscope (20×). Mites were considered dead if the appendages did not move when prodded with a pin. All treatments were replicated three times, and LD_{50} values were calculated by probit analysis.[19] The relative toxicity (RT) was determined as the ratio of synthetic acaricide LD_{50}/test sample LD_{50} using a previously described method.[20]

RESULTS AND DISCUSSION

The acaricidal activities of the essential oil extracted from *C. paniculatum* roots against *D. farinae*, *D. pteronyssinus*, and *T. putrescentiae* were evaluated using an impregnated fumigant bioassay and were compared to DEET, a well-known synthetic acaricide. On the basis of LD_{50} values, the essential oil extracted from *C. paniculatum* roots was about 8.93, 4.58, and 2.79 times more toxic than DEET (LD_{50} = 4.13, 3.91, and 4.87 $\mu g/cm^2$) against *D. farinae*, *D. pteronyssinus*, and *T. putrescentiae*, respectively (Table 1). Due to the excellent activity of the essential oil, identification of the oil constituents was determined by GC-MS (Table 2). The main constituents were 98.80% acetophenones (49.34% 2′-hydroxy-5′-methoxyacetophenone, 48.09% 4′-hydroxy-3′-methoxyacetophenone,

Table 1. Acaricidal Activities of *C. paniculatum* Oil and Synthetic Acaricides against *Dermatophagoides farinae*, *Dermatophagoides pteronyssinus*, and *Tyrophagus putrescentiae*

material	mite species	LD_{50} ($\mu g/cm^2$)	95% CL	RT[a]
C. paniculatum oil	*D. farinae*	4.13	3.18−5.08	8.93
	D. pteronyssinus	3.91	2.90−4.92	4.58
	T. putrescentiae	4.87	3.76−5.98	2.79
benzyl benzoate	*D. farinae*	9.92	9.83−10.01	3.7
	D. pteronyssinus	8.75	8.69−8.81	2.0
	T. putrescentiae	11.24	11.00−11.48	1.2
DEET	*D. farinae*	36.87	36.06−37.68	1.0
	D. pteronyssinus	17.93	17.00−18.86	1.0
	T. putrescentiae	13.58	12.50−14.66	1.0

[a]Relative toxicity = LD_{50} value of DEET/LD_{50} value of each chemical.

Table 2. Volatile Compounds Derived from *C. paniculatum* Oil Identified by GC-MS

compound	mass spectral data[a]	RI[b]	retention time (min)	relative content (%)
4′-hydroxyacetophenone	136, 121, 93, 65, 43	1468	9.11	1.26
4′-hydroxy-3′-methoxyacetophenone	166, 151, 123, 93, 65, 43, 15	1614	13.26	47.98
2′-hydroxy-5′-methoxyacetophenone	166, 151, 123, 95, 67, 43	1628	13.40	49.23
myristic acid	228, 185, 130, 73, 60, 57, 55, 43, 41	1400	17.77	0.32
palmitic acid	256, 213, 73, 60, 57, 55, 43, 41	1600	19.73	0.67

[a]Major fragmentation ions, base peak (listed first), and other ions in decreasing order of relative abundance. [b]RI, Kovats' index of retention.

dx.doi.org/10.1021/jf402330p | J. Agric. Food Chem. 2013, 61, 7568−7573

Table 3. Acaricidal Activities of Acetophenone Derivatives and Synthetic Acaricides against *Dermatophagoides farinae*, *Dermatophagoides pteronyssinus*, and *Tyrophagus putrescentiae*

material	mite species	LD_{50} ($\mu g/cm^2$)	95% CL	RT[a]
acetophenone	*D. farinae*	1.48	1.16−1.80	24.9
	D. pteronyssinus	1.84	1.48−2.20	9.7
	T. putrescentiae	0.56	0.39−0.73	24.3
2′-hydroxyacetophenone	*D. farinae*	1.74	1.52−1.96	21.2
	D. pteronyssinus	2.10	1.94−2.26	8.5
	T. putrescentiae	1.12	0.57−1.67	12.1
4′-hydroxyacetophenone	*D. farinae*	–[b]	–	–
	D. pteronyssinus	–	–	–
	T. putrescentiae	–	–	–
2′-methoxyacetophenone	*D. farinae*	0.75	0.59−0.91	49.2
	D. pteronyssinus	0.92	0.74−1.10	19.5
	T. putrescentiae	0.59	0.39−0.79	23.1
3′-methoxyacetophenone	*D. farinae*	0.41	0.19−0.63	89.9
	D. pteronyssinus	0.58	0.31−0.85	30.9
	T. putrescentiae	2.63	2.19−3.07	5.2
4′-methoxyacetophenone	*D. farinae*	0.52	0.38−0.66	70.9
	D. pteronyssinus	0.57	0.42−0.72	31.5
	T. putrescentiae	3.20	2.84−3.56	4.2
2′,4′-dimethoxyacetophenone	*D. farinae*	2.10	1.88−2.32	17.6
	D. pteronyssinus	1.75	1.56−1.94	10.2
	T. putrescentiae	–	–	–
2′,5′-dimethoxyacetophenone	*D. farinae*	1.87	1.60−2.14	19.7
	D. pteronyssinus	1.78	1.63−1.93	10.1
	T. putrescentiae	2.19	1.90−2.48	6.2
2′-hydroxy-4′-methoxyacetophenone	*D. farinae*	1.29	1.11−1.47	28.4
	D. pteronyssinus	1.25	1.05−1.45	14.3
	T. putrescentiae	2.28	2.01−2.55	6.0
2′-hydroxy-5′-methoxyacetophenone	*D. farinae*	1.03	0.79−1.27	35.8
	D. pteronyssinus	1.13	0.85−1.41	15.9
	T. putrescentiae	1.78	1.42−2.14	7.6
4′-hydroxy-3′-methoxyacetophenone	*D. farinae*	–	–	–
	D. pteronyssinus	–	–	–
	T. putrescentiae	–	–	–
benzyl benzoate	*D. farinae*	9.92	9.83−10.01	3.7
	D. pteronyssinus	8.75	8.69−8.81	2.0
	T. putrescentiae	11.24	11.00−11.48	1.2
DEET	*D. farinae*	36.87	36.06−37.68	1.0
	D. pteronyssinus	17.93	17.00−18.86	1.0
	T. putrescentiae	13.58	12.50−14.66	1.0

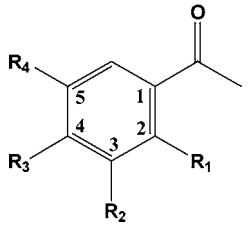
[a]Relative toxicity = LD_{50} value of DEET/LD_{50} value of each chemical. [b]No activity.

and 1.37% 4′-hydroxyacetophenone) and 1.20% fatty acids (0.78% palmitic acid and 0.42% myristic acid).

To isolate the active compound of the *C. paniculatum* oil, various chromatographic analyses were performed using single or mixed organic solvents. As a result, CP-3322 was isolated and identified by spectroscopic analyses including EI-MS, ^{1}H NMR, and ^{13}C NMR. The isolated CP-3322 was characterized as 2′-hydroxy-5′-methoxyacetophenone ($C_{14}H_8O_5$): EI-MS (70 eV) m/z M^+ 166.17; ^{1}H NMR ($CDCl_3$, 600 MHz) δ 7.252 (1H, s), 7.151−7.236 (1H, d, J = 67.2 Hz), 7.075−7.148 (1H, t, J = 43.8 Hz), 6.884−6.951 (1H, t, J = 40.2 Hz), 6.451−6.488 (1H, d, J = 22.2 Hz), 5.311−5.351 (OH, t, J = 24.0 Hz), 2.313−

dx.doi.org/10.1021/jf402330p | J. Agric. Food Chem. 2013, 61, 7568−7573

Table 4. Structure–Activity Relationships of Acetophenone Derivatives



compound	R_1	R_2	R_3	R_4	LD_{50} ($\mu g/cm^2$)		
					D. farinae	*D. pteronyssinus*	*T. putrescentiae*
acetophenone	H	H	H	H	**1.48**	**1.84**	**0.56**
2′-hydroxyacetophenone	**OH**	H	H	H	**1.74**	**2.10**	**1.12**
4′-hydroxyacetophenone	H	H	**OH**	H	–[a]	–	–
2′-methoxyacetophenone	**OCH_3**	H	H	H	**0.75**	**0.92**	**0.59**
3′-methoxyacetophenone	H	**OCH_3**	H	H	**0.41**	**0.58**	**2.63**
4′-methoxyacetophenone	H	H	**OCH_3**	H	**0.52**	**0.57**	**3.20**
2′,4′-dimethoxyacetophenone	**OCH_3**	H	**OCH_3**	H	**2.10**	**1.75**	–
2′,5′-dimethoxyacetophenone	**OCH_3**	H	H	**OCH_3**	**1.87**	**1.78**	**2.19**
2′-hydroxy-4′-methoxyacetophenone	**OH**	H	**OCH_3**	H	**1.29**	**1.25**	**2.28**
2′-hydroxy-5′-methoxyacetophenone	**OH**	H	H	**OCH_3**	**1.03**	**1.13**	**1.78**
4′-hydroxy-3′-methoxyacetophenone	H	**OCH_3**	**OH**	H	–	–	–

[a]No activity.

2.333 (3H, t, J = 12.0 Hz); ^{13}C NMR ($CDCl_3$, 150 MHz) δ 188.0, 182.1, 164.5, 163.3, 161.8, 147.6, 136.4, 133.3, 122.7, 120.2, 113.3, 108.9, 108.3, 107.0, 21.6. The research findings of 2′-hydroxy-5′-methoxyacetophenone were compared with those of previous study.[21]

Because of the potent acaricidal activity of 2′-hydroxy-5′-methoxyacetophenone, acaricidal activities of acetophenone and its derivatives based on the acetophenone skeleton were evaluated using an impregnated fumigant bioassay against the three mite species and were compared with the activities of synthetic acaricides (benzyl benzoate and DEET) (Table 3). The LD_{50} values of acetophenone were 1.48, 1.84, and 0.56 $\mu g/cm^2$ against *D. farinae*, *D. pteronyssinus*, and *T. putrescentiae*, respectively. On the basis of LD_{50} values, acetophenone was about 6.7, 4.8, and 20.1 times more toxic than benzyl benzoate (LD_{50} = 9.92, 8.75, and 11.24 $\mu g/cm^2$) and about 24.9, 9.7, and 24.3 times more toxic than DEET (LD_{50} = 36.87, 17.93, and 13.58 $\mu g/cm^2$) against *D. farinae*, *D. pteronyssinus*, and *T. putrescentiae*, respectively. These results indicate that acaricidal activity is different for each species of mite due to species-specific differences.[22]

2′-Hydroxy-5′-methoxyacetophenone derivatives were selected such as acetophenone, 2′-hydroxyacetophenone, 4′-hydroxyacetophenone, 2′-methoxyacetophenone, 3′-methoxyacetophenone, 4′-methoxyacetophenone, 2′,4′-dimethoxyacetophenone, 2′,5′-dimethoxyacetophenone, 2′-hydroxy-4′-methoxyacetophenone, and 4′-hydroxy-3′-methoxyacetophenone. On the basis of the LD_{50} values against *D. farinae*, the most toxic compound was 3′-methoxyacetophenone (0.41 $\mu g/cm^2$), followed by 4′-methoxyacetophenone (0.52 $\mu g/cm^2$), 2′-methoxyacetophenone (0.75 $\mu g/cm^2$), 2′-hydroxy-5′-methoxyacetophenone (1.03 $\mu g/cm^2$), 2′-hydroxy-4′-methoxyacetophenone (1.29 $\mu g/cm^2$), acetophenone (1.48 $\mu g/cm^2$), 2′-hydroxyacetophenone (1.74 $\mu g/cm^2$), 2′,5′-dimethoxyacetophenone (1.87 $\mu g/cm^2$), and 2′,4′-dimethoxyacetophenone (2.10 $\mu g/cm^2$). Against *D. pteronyssinus*, 4′-methoxyacetophenone (0.57 $\mu g/cm^2$) had the most acaricidal activity, followed by 3′-methoxyacetophenone (0.58 $\mu g/cm^2$), 2′-methoxyacetophenone (0.92 $\mu g/cm^2$), 2′-hydroxy-5′-methoxyacetophenone (1.13 $\mu g/cm^2$), 2′-hydroxy-4′-methoxyacetophenone (1.25 $\mu g/cm^2$), 2′,4′-dimethoxyacetophenone (1.75 $\mu g/cm^2$), 2′,5′-dimethoxyacetophenone (1.78 $\mu g/cm^2$), acetophenone (1.84 $\mu g/cm^2$), and 2′-hydroxyacetophenone (2.10 $\mu g/cm^2$). In the case of *T. putrescentiae*, acetophenone (0.56 $\mu g/cm^2$) had the most acaricidal effect, followed by 2′-methoxyacetophenone (0.59 $\mu g/cm^2$), 2′-hydroxyacetophenone (1.12 $\mu g/cm^2$), 2′-hydroxy-5′-methoxyacetophenone (1.78 $\mu g/cm^2$), 2′,5′-dimethoxyacetophenone (2.19 $\mu g/cm^2$), 2′-hydroxy-4′-methoxyacetophenone (2.28 $\mu g/cm^2$), 3′-methoxyacetophenone (2.63 $\mu g/cm^2$), and 4′-methoxyacetophenone (3.20 $\mu g/cm^2$). However, 4′-hydroxyacetophenone and 4′-hydroxy-3′-methoxyacetophenone exhibited no detectable activities against the three mite species at 80 $\mu g/cm^2$, and 2′,4′-dimethoxyacetophenone showed no acaricidal activity against *T. putrescentiae* at 80 $\mu g/cm^2$ (Table 3).

To establish structure–activity relationships between acaricidal activity and functional radicals (hydroxyl and methoxy groups) on the acetophenone skeleton (R_1, R_2, R_3, and R_4), we described the acaricidal activities of acetophenone derivatives according to LD_{50} value (Table 4). For acetophenone derivatives formed by introducing a hydroxyl group (R_1 or R_3) into the acetophenone skeleton, the acaricidal activity of 2′-hydroxyacetophenone was slightly decreased relative to that of acetophenone, whereas 4′-hydroxyacetophenone did not exhibit acaricidal activity against the three mite species. For acetophenone derivatives with the addition of a methoxy group (R_1, R_2, R_3, or R_4), 2′-methoxyacetophenone had potent acaricidal activity against the three mite species. The acaricidal activities of 3′- and 4′-methoxyacetophenone were greater than that of acetophenone against *Dermatophagoides* spp. but less than that of acetophenone against *T. putrescentiae*. Moreover, the acaricidal activities of 2′,4′- and 2′,5′-dimethoxyacetophenone containing a methoxy group in 2′-methoxyacetophenone against *Dermatophagoides* spp. were slightly decreased. For *T. putrescentiae*, 2′,4′- and 2′,5′-dimethoxyacetophenone had no or weak acaricidal activities. Compared with acaricidal activities of

dx.doi.org/10.1021/jf402330p | J. Agric. Food Chem. 2013, 61, 7568–7573

acetophenone derivatives combining hydroxyl and methoxy groups in the acetophenone skeleton, 2′-hydroxy-4′-methoxyacetophenone and 2′-hydroxy-5′-methoxyacetophenone were more toxic to *Dermatophagoides* spp. than was 2′-hydroxyacetophenone but had less acaricidal activity against *T. putrescentiae* than 2′-hydroxyacetophenone. 4′-Hydroxy-3′-methoxyacetophenone had no activity, despite the acaricidal activity of 3′-methoxyacetophenone against the three mite species.

In addition, Kerr[23] reported that insecticidal activity depends on the presence of a methoxy group in the benzene ring (eugenol, methyleugenol, and elemicin). Taken together, acetophenone derivatives with a methoxy group at R_1, R_2, or R_3 position on an acetophenone skeleton had potent acaricidal effects against the three mite species. Changes in the positions of functional radicals on an acetophenone skeleton play an important role in acaricidal effects.

Recent studies of acaricides have sought to develop newer and safer agents to replace older agents and avoid the problems of toxicity, resistance, and environmental damage.[24,25] According to the Sigma-Aldrich database,[26] the oral LD_{50} values in mammals of acetophenone, 2′-hydroxy-4′-methoxyacetophenone, and 4′-methoxyacetophenone are 815, 490, and 1720 mg/kg, respectively. Furthermore, these compounds are commonly used as components of flavors and fragrances and are found in naturally occurring materials such as honey, plums, and strawberries.[27] Taken together, these results indicate that acetophenone and its derivatives would be good replacements for commercial synthetic acaricides and would also be suitable as fumigants in vapor-phase acaricides. Further studies should be conducted to determine the effects of acetophenone derivatives on human health and the environment and to continue development of effective formulations to improve acaricidal potency and stability.

AUTHOR INFORMATION

Corresponding Author

*(H.-S.L.) Phone: +82 63 270 2544. Fax: +82 63 270 2550. E-mail: hoiseon@jbnu.ac.kr. (J.-Y.Y.) E-mail: jiyeo0527@jbnu.ac.kr.

Funding

This research was supported by the Basic Science Research Program through the National Research Foundation of Korea (NRF) funded by the Ministry of Education, Science and Technology (2012R1A1A4A01002124).

Notes

The authors declare no competing financial interest.

REFERENCES

(1) Mario, C. G.; Belinda, G. N.; Daniel, R. A. Comparative mites and cockroaches sensitization study in three cities of Mexico. *Rev. Alergia Mex.* **2008**, *55*, 234−239.

(2) Hiramatsu, Y.; Shida, S.; Miyazaki, Y. House dust mites and their sensitivity to wood oil and volatiles. *J. Wood Sci.* **2008**, *54*, 1−9.

(3) Evelyn, M.; Ana, M. D. Allergy to house dust mites and asthma. *P. R. Health Sci. J.* **2004**, *23*, 47−57.

(4) Arlian, L. G.; Morgan, M. S.; Vyszenski-Moher, D. L.; Sharra, D. Cross-reactivity between storage and dust mites and between mites and shrimp. *Exp. Appl. Acarol.* **2009**, *47*, 159−172.

(5) Aygun, O.; Yaman, M.; Durmaz, H. A. Survey on occurrence of *Trophagus putresentiae* (Acari: Acaridae) in Surk, a traditional Turkish dairy product. *J. Food Eng.* **2007**, *78*, 878−881.

(6) Lee, H. S. *p*-Anisaldehyde: acaricidal component of *Pimpinella anisum* seed oil against the house dust mites *Dermatophagoides farinae* and *Dermatophagoides pteronyssinus*. *Planta Med.* **2004**, *70*, 279−281.

(7) Ahn, Y. J.; Lee, S. B.; Lee, H. S.; Kim, G. H. Insecticidal and acaricidal activity of carvacrol and β-thujaplicine derived from *Thujopsis dolabrata* var. hondai sawdust. *J. Chem. Ecol.* **1998**, *24*, 8−12.

(8) Skelton, A. C.; Cameron, M. M.; Pickett, J. A.; Birkett, M. A. Identification of neryl formate as the airborne aggregation pheromone for the American house dust mite and the European house dust mite (Acri: Epidermoptidae). *J. Med. Entomol.* **2010**, *47*, 798−804.

(9) Yang, Y. C; Lee, S. G.; Lee, H. K.; Kim, M. K.; Lee, S. H.; Lee, H. S. A piperidine amide extracted from *Piper longum* L. fruit show activity against *Aedes aegypti* mosquito lava. *J. Agric. Food Chem.* **2002**, *50*, 7700−7703.

(10) Lim, J. H.; Kim, H. W.; Jeon, J. H.; Lee, H. S. Acaricidal constituents isolated from *Sinapis alba* L. seeds and structure-activity relationships. *J. Agric. Food Chem.* **2008**, *56*, 9962−9966.

(11) Lee, C. H.; Lee, S. G.; Lee, H. S. Acaricidal effects of *Thymus vulgaris* leaf-derived materials and monoterpene alcohols against *Dermatophagoides* spp. *J. Korean Soc. Appl. Biol. Chem.* **2010**, *53*, 170−174.

(12) Lee, H. S. Acaricidal effects of quinone and its congeners and color alteration of *Dermatophagoids* spp. with quinone. *J. Microbiol. Biotechnol.* **2007**, *17*, 1394−1398.

(13) Dou, J.; Li, P.; Bi, Z. M.; Zhou, J. L. New C_{21} steroidal glycoside from *Cynanchum paniculatum*. *Chin. Chem. Lett.* **2007**, *18*, 300−302.

(14) Sun, Y.; Liu, Z.; Wang, J.; Tian, W.; Zhou, H.; Zhu, L.; Zhang, C. Supercritical fluid extraction of paeonol from *Cynanchum paniculatum* (Bae.) Kitag. and subsequent isolation by high-speed counter-current chromatography coupled with high-performance liquid chromatography-photodiode array detector. *Sep. Purif. Technol.* **2008**, *64*, 221−226.

(15) Rajabi, L.; Courreges, C.; Montoya, J.; Aguilera, R. J.; Rrimm, T. P. Acetophenones with selective antimycobacterial activity. *Lett. Appl. Microbiol.* **2005**, *40*, 212−217.

(16) Oh, M. S.; Yang, J. Y.; Lee, H. S. Acaricidal toxicity of 2′-hydroxy-4′-methylacetophenone isolated from *Angelicae koreana* roots and structure-activity relationships of its derivatives. *J. Agric. Food Chem.* **2012**, *60*, 3606−3611.

(17) Lee, C. H.; Kim, H. W.; Lee, H. S. Acaricidal properties of piperazine and its derivatives against house-dust and stored-food mites. *Pest Manag. Sci.* **2009**, *65*, 704−710.

(18) Yang, J. Y.; Lee, H. S. Acaricidal activities of the active component of *Lycopus lucidus* oil and its derivatives against house dust and stored food mites (Arachnida: Acari). *Pest Manag. Sci.* **2012**, *68*, 564−572.

(19) SAS Institute. *SAS/STAT User's Guide*, version 6; SAS Institute: Cary, NC, 1990.

(20) Akhtar, Y.; Isman, M. B. Feeding responses of specialist herbivores to plant extracts and pure allelochemicals: effects of prolonged exposure. *Entomol. Exp. Appl.* **2004**, *111*, 201−208.

(21) Boyer, J. L.; Krum, J. E.; Myers, M. C.; Fazal, A. N.; Wigal, C. T. Synthetic utility and mechanistic implications of the fries rearrangement of hydroquinone diesters in boron trifluoride complexes. *J. Org. Chem.* **2000**, *65*, 4712−4714.

(22) Jeong, E. Y.; Kim, M. G.; Lee, H. S. Acaricidal activity of triketone analogues derived from *Leptospermum scoparium* oil against house-dust and stored-food mites. *Pest Manag. Sci.* **2009**, *65*, 327−331.

(23) Kerr, R. W. Adjuvants for pyrethrins in fly sprays. *Aust. Commonw. Sci. Ind. Res. Organ. Bull.* **1951**, 261.

(24) Pollart, S. M.; Ward, G. W.; Platts-Mills, T. A. E. House dust sensitivity and environmental control. *Immunol. Allergy Clin. North Am.* **1987**, *7*, 447−461.

(25) Bronswijk, J. E.; Sinha, R. N. Pyroglyphid mites (Acari) and house dust allergy. *J. Allergy* **1971**, *47*, 31−52.

(26) Sigma-Aldrich. Material Safety Data Sheet (MSDS): Toxicological Information, Section 11; Sigma-Aldrich: St. Louis, MO, 2010.

(27) Hilton, M. D.; Cain, W. J. Bioconversion of cinnamic acid to acetophenone by a pseudomonad: microbial production of a natural flavor compound. *Appl. Environ. Microbiol.* **1990**, *56*, 623−627.