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Antioxidant Properties of Several Medicinal Mushrooms

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Three species of medicinal mushrooms are commercially available in Taiwan, namely, *Ganoderma lucidum* (Ling-chih), *Ganoderma tsugae* (Sung-shan-ling-chih), and *Coriolus versicolor* (Yun-chih). Methanolic extracts were prepared from these medicinal mushrooms and their antioxidant properties studied. At 0.6 mg/mL, *G. lucidum*, *G. lucidum* antler, and *G. tsugae* showed an excellent antioxidant activity (2.30−6.41% of lipid peroxidation), whereas *C. versicolor* showed only 58.56%. At 4 mg/mL, reducing powers were in the order *G. tsugae* (2.38) ∼ *G. lucidum* antler (2.28) > *G. lucidum* (1.62) > *C. versicolor* (0.79). At 0.64 mg/mL, scavenging effects on the 1,1-diphenyl-2-picrylhydrazyl radical were 67.6−74.4% for *Ganoderma* and 24.6% for *C. versicolor*. The scavenging effect of methanolic extracts from *G. lucidum* and *G. lucidum* antler on hydroxyl radical was the highest (51.2 and 52.6%) at 16 mg/mL, respectively. At 2.4 mg/mL, chelating effects on ferrous ion were in the order *G. lucidum* antler (67.7%) > *G. lucidum* (55.5%) > *G. tsugae* (44.8%) > *C. versicolor* (13.2%). Total phenols were the major naturally occurring antioxidant components found in methanolic extracts from medicinal mushrooms. Overall, *G. lucidum* and *G. tsugae* were higher in antioxidant activity, reducing power, scavenging and chelating abilities, and total phenol content.

KEYWORDS: Medicinal mushrooms; *Coriolus versicolor*; *Ganoderma*; antioxidant activity; reducing power; scavenging effect; chelating effect; antioxidant components

INTRODUCTION

Numerous physiological processes in living organisms occasionally produce oxygen-centered free radicals and other reactive oxygen species as byproducts. Oxidative damage caused by free radicals may be related to aging and diseases, such as atherosclerosis, cancer, and rheumatoid arthritis (*1*). Although humans and other organisms possess antioxidant defense and repair systems that have evolved to protect them against oxidative damage, these systems are insufficient to totally prevent the damage (*2*). However, the antioxidants in human diets are of great interest as possible protective agents to help human body reduce oxidative damage.

Recently, a multitude of natural antioxidants have already been isolated from different kinds of plant materials such as oilseeds, cereal crops, vegetables, fruits, leaves, roots, spices, and herbs (*3*). Chinese herbs have been used for diet therapy for several millennia. Some of them are alleged to exhibit significant antioxidant activity (*4*, *5*). Mushrooms are a traditional Chinese medicine and also commonly used as food. Currently, three species of medicinal mushrooms are commercially available in Taiwan, namely, *Ganoderma lucidum* (Curtis: Fr.) Karsten (Ling-chih or reishi), *Ganoderma tsugae* Murrill (Sung-shan-ling-chih), and *Coriolus versicolor* (Fr.) Quel. (Yun-chih or turkey tail).

These medicinal mushrooms are commonly used for pharmaceutical purposes and as health foods. These medicinal mushrooms were found to be medically active in several therapeutic effects such as antitumor, immunomodulating, and chronic bronchitis (*6*). The nutritional values and taste components of these medicinal mushrooms were clearly studied (*7*). Although research was focused on the therapeutic effects of these medicinal mushrooms, little information is available about their antioxidant properties. Our objective was to evaluate the antioxidant properties of these medicinal mushrooms including antioxidant activity, reducing power, scavenging effects on radical, and chelating effects on ferrous ion. The contents of potential antioxidant components of these medicinal mushrooms were also determined.

MATERIALS AND METHODS

Mushrooms. *C. versicolor*, *G. lucidum*, *G. lucidum* antler (in varnished and forked shape), and *G. tsugae* were purchased, in a dried form, at a local market in Taichung City, Taiwan. For each of four types of mushrooms, three samples (∼50 g each) were randomly selected and prepared for analyses. After a fine powder (20 mesh) was obtained using a mill (Restsch Ultra centrifugal mill and sieving machine, Haan, Germany), a subsample (5 g) was extracted by stirring with 100 mL of methanol at 25 °C at 20*g* for 24 h and filtering through Whatman no. 4 filter paper. The residue was then extracted with two

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additional 100 mL portions of methanol as described above. The combined methanolic extracts were then rotary evaporated at 40 °C to dryness. The dried extract was used directly for analyses of antioxidant components or redissolved in methanol to a concentration of 20 mg/mL and stored at 4 °C for further use.

Antioxidant Activity. The antioxidant activity was determined by the 1,3-diethyl-2-thiobarbituric acid (DETBA) method (*8*, *9*). To 50 μL of each mushroom extract (0.1−0.6 mg/mL) in methanol was added 50 μL of linoleic acid solution (Sigma Chemical Co., St. Louis, MO; 2 mg/mL in 95% ethanol). The mixture was incubated in an oven at 80 °C for 60 min and cooled in an ice bath. To the mixture were sequentially added 200 μL of 20 mM butylated hydroxytoluene (BHT, Sigma), 200 μL of 8% sodium dodecyl sulfate (SDS, Merck, Darmstadt, Germany), 400 μL of deionized water, and 3.2 mL of 12.5 mM DETBA (Aldrich Chemical Co., Milwaukee, WI) in sodium phosphate buffer (pH 3.0). The mixture was mixed thoroughly, placed in an oven at 95 °C for 15 min, and then cooled with an ice bath. After 4 mL of ethyl acetate was added, the mixture was mixed and centrifuged at 1000*g* at 20 °C for 15 min. Ethyl acetate was separated, and its fluorescence intensity was measured in a Hitachi 650-40 spectrofluorometer with fluorescence excitation at 515 nm and emission at 555 nm. The antioxidant activity was expressed as the percentage of lipid peroxidation with a control containing no mushroom extract being 100%. *A higher percentage indicates a lower antioxidant activity.*

Reducing Power. The reducing power was determined according to the method of Oyaizu (*10*). Each mushroom extract (0.5−4 mg/mL) in methanol (2.5 mL) was mixed with 2.5 mL of 200 mM sodium phosphate buffer (pH 6.6; Wako Pure Chemical Co., Osaka, Japan) and 2.5 mL of 1% potassium ferricyanide (Sigma), and the mixture was incubated at 50 °C for 20 min. After 2.5 mL of 10% trichloroacetic acid (w/v; Wako) was added, the mixture was centrifuged at 200*g* for 10 min. The upper layer (5 mL) was mixed with 5 mL of deionized water and 1 mL of 0.1% ferric chloride (Wako), and the absorbance was measured at 700 nm in a Hitachi U-2001 spectrophotometer. A solution with all reagents but the extracts was used as a blank. A higher absorbance indicates a higher reducing power.

Scavenging Effect on 1,1-Diphenyl-2-picrylhydrazyl (DPPH) Radical. Each mushroom extract (0.75−0.65 mg/mL) in methanol (4 mL) was mixed with 1 mL of methanolic solution containing DPPH (Sigma) radical, resulting in a final concentration of 0.2 mM DPPH. The mixture was shaken vigorously and left to stand for 30 min in the dark, and the absorbance was then measured at 517 nm (*11*).

Scavenging Effect on Hydroxyl Radical. The hydroxyl radical reacted with the nitrone spin trap 5,5-dimethylpyrroline-*N*-oxide (DMPO, Sigma), and the resultant DMPO−OH adduct was detected with an electron paramagnetic resonance (EPR) spectrometer. The EPR spectrum was recorded 2.5 min after 200 μL of each mushroom extract (4−16 mg/mL) in methanol was mixed with 200 μL of 10 mM H_2O_2 (Merck), 200 μL of 10 mM Fe^{2+} (Sigma), and 200 μL of 10 mM DMPO using a Bruker EMX-10 EPR spectrometer at the following settings: 3480 G magnetic field; 1.0 G modulation amplitude; 0.5 s time constant; and 200 s scan period (*12*).

Chelating Effects on Ferrous Ion. Chelating effect was determined according to the method of Shimada et al. (*11*). To 2 mL of the mixture consisting of 30 mM hexamine (Wako), 30 mM potassium chloride (Sigma), and 9 mM ferrous sulfate (Union Chemical Works, Hsinchu, Taiwan) was added 2 mL of each mushroom extract (0.3−2.4 mg/mL) in methanol and 200 μL of 1 mM tetramethyl murexide (TMM, Sigma). After 3 min at room temperature, the absorbance of the mixture was determined at 485 nm. *A lower absorbance indicates a higher chelating power.*

Determination of Antioxidant Components. Ascorbic acid was determined according to the method of Klein and Perry (*13*). The dried methanolic extract from medicinal mushrooms (20 mg) was extracted with 10 mL of 1% metaphosphoric acid (Union) for 45 min at room temperature and filtered through Whatman no. 4 filter paper. The filtrate (1 mL) was mixed with 9 mL of 2,6-dichloroindophenol (Sigma), and the absorbance was measured at 515 nm within 15 s. The content of ascorbic acid was calculated on the basis of the calibration curve of authentic L-ascorbic acid (Sigma).

Table 1. Yield of Methanolic Extracts from Several Medicinal Mushrooms

mushroom	amount[a] (g)	extraction yield[b] (%, w/w)
C. versicolor (Yun-chih)	0.46 ± 0.01	9.16a
G. lucidum (Ling-chih)	0.29 ± 0.01	5.61b
G. lucidum antler (Ling-chih)	0.31 ± 0.01	6.15b
G. tsugae (Sung-shan-ling-chih)	0.20 ± 0.01	3.97c

[a] Extracted from dried medicinal mushrooms (5.00 g). Each value is expressed as mean ± standard deviation ($n = 3$). [b] Means with different letters within a column are significantly different ($p < 0.05$).

β-Carotene was extracted and analyzed as described by Rundhaug et al. (*14*). The dried methanolic extract from medicinal mushrooms (20 mg) was extracted with a solution of 1% pyrogallol (Wako) in 10 mL of methanol/dichloromethane (1:1, v/v) for 45 min at room temperature and filtered through Whatman no. 4 filter paper; the volume was adjusted to 10 mL using the same solution. The filtrate was then passed through a filter unit (13 mm, Lida Corp., Kenosha, WI) and filtered using a 0.45-μm CA filter paper prior to injection onto a high-performance liquid chromatograph (HPLC).

The HPLC system consisted of a Hitachi D-6200 pump, a Hitachi L-5000 LC controller, a Rheodyne 7161 injector, a 20-μL sample loop, a Hitachi D-2500 chromatointegrator, a Hitachi L-4000 UV detector, and a Prodigy 5 ODS-2 column (4.6 × 250 mm, 5 μm, Phenomenex Inc., Torrance, CA). The mobile phase was acetone/methanol/acetonitrile, 1:2:2 (v/v/v), at a flow rate of 0.7 mL/min, and UV detection was at 470 nm. The content of β-carotene was calculated on the basis of the calibration curve of authentic β-carotene (Sigma).

Tocopherols were extracted and analyzed according to the method of Carpenter (*15*). The dried methanolic extract from medicinal mushrooms (50 mg) was suspended in 6 mL of pyrogallol (6% in 95% ethanol) and 4 mL of 60% potassium hydroxide aqueous solution, and the resulting mixture was saponified at 70 °C for 20 min. Deionized water (15 mL) was added, and the mixture was extracted with 15 mL of *n*-hexane. The organic layer was washed with deionized water to neutral, dried over anhydrous sodium sulfate, and rotary evaporated to dryness. The residue was redissolved in 5 mL of *n*-hexane and filtered prior to HPLC injection in the same manner as in the β-carotene assay.

The HPLC system was the same as for the β-carotene assay. The mobile phase was acetonitrile/methanol, 85:15 (v/v), at a flow rate of 1.0 mL/min, and UV detection was at 295 nm. The content of each tocopherol was calculated on the basis of the calibration curve of each authentic tocopherol (Sigma).

Total phenols were determined according to the method of Taga et al. (*16*). The dried methanolic extract from medicinal mushrooms (20 mg) was dissolved in a solution of 5 mL of 1.3% HCl in methanol/deionized water (60:40, v/v), and the resulting mixture (100 μL) was added to 2 mL of 2% aqueous sodium carbonate solution. After 3 min, 100 μL of 50% Folin−Ciocalteau reagent (Sigma) was added to the mixture. After 30 min of standing, absorbance was measured at 750 nm. The content of total phenols was calculated on the basis of the calibration curve of gallic acid (Sigma).

Statistical Analysis. For methanolic extracts from mushrooms, three samples were prepared for assays of every antioxidant attribute. The experimental data were subjected to an analysis of variance for a completely random design as described by Steel et al. (*17*) to determine the least significant difference at the level of 0.05.

RESULTS AND DISCUSSION

Antioxidant Activity. Following the extraction with methanol, four medicinal mushrooms had the yields of 3.97−9.16% (**Table 1**). As compared to other specialty and commercial mushrooms (*18*), the lower yields were consistent with their lower amounts of water-soluble components, especially soluble sugars and sugar alcohols (*7*). Using the DETBA method, methanolic extracts from medicinal mushrooms showed two

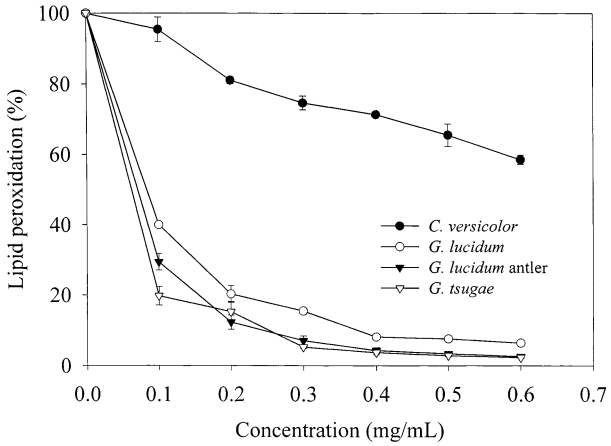

Figure 1. Antioxidant activity of methanolic extracts from several medicinal mushrooms. Each value is expressed as mean ± standard deviation ($n = 3$).

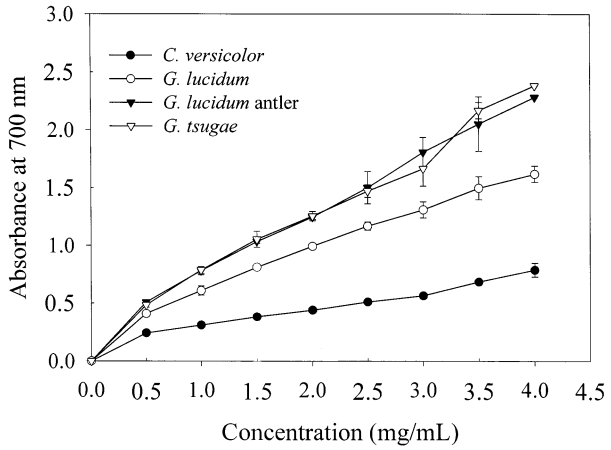

Figure 2. Reducing power of methanolic extracts from several medicinal mushrooms. Each value is expressed as mean ± standard deviation ($n = 3$).

different patterns of antioxidant activities (**Figure 1**). At 0.6 mg/mL, *Ganoderma* spp., including *G. lucidum*, *G. lucidum* antler, and *G. tsugae*, showed an excellent antioxidant activity as evidenced by the relatively low percentages of lipid peroxidation (2.30−6.41%). *C. versicolor* showed only 58.56% of lipid peroxidation at 0.6 mg/mL. It could be contemplated that at concentrations >0.6 mg/mL, *C. versicolor* would show a better antioxidant activity. However, butylated hydroxyanisole (BHA) showed only 66.1% of lipid peroxidation at 10 mg/mL.

Huang (*19*) found that methanolic extracts from the medicinal mushroom *Antrodia camphorata* (Chang-chih) showed excellent antioxidant activities as evidenced by 5.32−5.78% of lipid peroxidation at 1.0 mg/mL. Methanolic extract from another medicinal mushroom, *Agaricus blazei* (Brazilian mushrooms), showed a high antioxidant activity (26.0% of lipid peroxidation) at 1.0 mg/mL (*19*). Among methanolic extracts from four specialty mushrooms at 1.2 mg/mL, only *Dictyophora indusiata* (basket stinkhorn) showed an excellent antioxidant activity (2.26% of lipid peroxidation) (*18*). *Grifola frondosa* (maitake) showed a relatively high antioxidant activity (29.81% of lipid peroxidation), whereas *Hericium erinaceus* (lion's mane) and *Tricholoma giganteum* (white matsutake) showed moderate antioxidant activities (48.45 and 67.02% of lipid peroxidation, respectively) (*18*).

Among methanolic extracts from commercial mushrooms at 1.2 mg/mL, *Flammulina velutipes* (winter mushrooms), *Lentinula edodes* (shiitake), *Pleurotus cystidiosus* (abalone mushrooms), and *Pleurotus ostreatus* (tree oyster mushrooms) showed moderate to high antioxidant activities (24.71−62.30% of lipid peroxidation) (*18*). Mau et al. (*20*) reported that methanolic extracts from ear mushrooms, including black, red, jin, snow, and silver ears, showed low to moderate antioxidant activities (57.7−71.5% of lipid peroxidation) at 1.0 mg/mL.

With regard to the antioxidant activities of methanolic extracts in the DETAB method, *A. camphorata*, *G. lucidum*, *G. lucidum* antler, and *G. tsugae* were excellent, whereas *C. versicolor* was good as compared to other medicinal, specialty, commercial, and ear mushrooms.

Reducing Power. Reducing powers of methanolic extracts from medicinal mushrooms increased readily along with the increased concentrations (**Figure 2**). At 4 mg/mL, reducing powers were in the order *G. tsugae* (2.38) ∼ *G. lucidum* antler (2.28) > *G. lucidum* (1.62) > *C. versicolor* (0.79). Similar to antioxidant activity, *C. versicolor* showed a lower reducing power than *Ganoderma* spp. However, reducing powers of BHA and α-tocopherol at 20 mM (3.6 and 8.6 mg/mL) were 0.12 and 0.13, respectively. The reducing power of medicinal mushrooms might be due to their hydrogen-donating ability as described by Shimada et al. (*11*). Accordingly, medicinal mushrooms might contain a higher amount of reductone, which could react with radicals to stabilize and terminate radical chain reactions.

Huang (*19*) reported that the methanolic extract from *A. camphorata* showed an excellent reducing power of 0.96−0.97 at 10 mg/mL, whereas that from Brazilian mushrooms showed a reducing power of 0.86 at 10 mg/mL. Among methanolic extracts from four specialty mushrooms, basket stinkhorn showed an excellent reducing power of 1.96 at 6 mg/mL (*18*). Reducing powers of methanolic extracts from maitake, lion's mane, and white matsutake were 1.18, 1.01, and 0.63 at 9 mg/mL, respectively (*18*). Among methanolic extracts from commercial mushrooms, abalone and tree oyster mushrooms exhibited excellent reducing powers of 1.00 and 1.19 at 10 mg/mL, respectively (*18*). Reducing powers of methanolic extracts from two strains of winter mushrooms were 0.52 and 0.65 at 10 mg/mL, whereas reducing powers of 0.62 and 0.85 were observed with extracts from two strains of shiitake at 10 mg/mL (*18*). Mau et al. (*20*) reported that methanolic extracts from ear mushrooms excluding silver ears showed reducing powers of 0.67−0.82 at 5 mg/mL. The reducing power of that from silver ears was 0.32 at 5 mg/mL (*20*).

Apparently, with regard to reducing powers of methanolic extracts, *A. camphorata*, *G. lucidum*, *G. lucidum* antler, and *G. tsugae* were excellent, whereas *C. versicolor* was good as compared to other medicinal, specialty, commercial, and ear mushrooms.

Scavenging Effect on 1,1-Diphenyl-2-picrylhydrazyl Radical. Scavenging effects of methanolic extracts from medicinal mushrooms on DPPH radical increased with the increased concentrations (**Figure 3**). At 0.64 mg/mL, scavenging effects were 67.6−74.4% for *Ganoderma* and 24.6% for *C. versicolor*. It was anticipated that scavenging effects would be excellent for *Ganoderma* and higher for *C. versicolor* at concentrations >0.64 mg/mL. However, the scavenging effects of BHA and α-tocopherol at 20 mM (3.6 and 8.6 mg/mL) were 96 and 95%, respectively.

Excellent scavenging effects (96.3−99.1 and 97.1%) were observed with methanolic extracts from *A. camphorata* and Brazilian mushrooms at 2.5 mg/mL, respectively (*19*). At 6.4 mg/mL, the methanolic extract from basket stinkhorn scavenged

Table 2. Contents of Ascorbic Acid, β-Carotene, Tocopherols, and Total Phenols of Methanolic Extracts from Several Medicinal Mushrooms

	content[a] (mg/g)			
compound	*C. versicolor*	*G. lucidum*	*G. lucidum* (antler)	*G. tsugae*
ascorbic acid	nd[b]	nd	nd	nd
β-carotene	nd	nd	nd	nd
tocopherols	nd	1.19 ± 0.03a (γ)[c]	0.13 ± 0.02c (γ)	1.07 ± 0.07b (γ)
total phenols	23.28 ± 0.90d	47.25 ± 0.20c	55.96 ± 0.58a	51.28 ± 0.51b

[a] Each value is expressed as mean ± standard deviation ($n = 3$). Means with different letters within a row are significantly different ($p < 0.05$). [b] nd, not detected. [c] γ, γ-tocopherol.

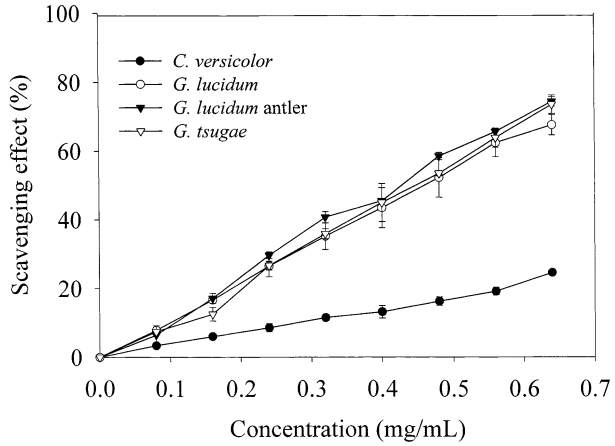

Figure 3. Scavenging effect of methanolic extracts from several medicinal mushrooms on 1,1-diphenyl-2-picrylhydrazyl radical. Each value is expressed as mean ± standard deviation ($n = 3$).

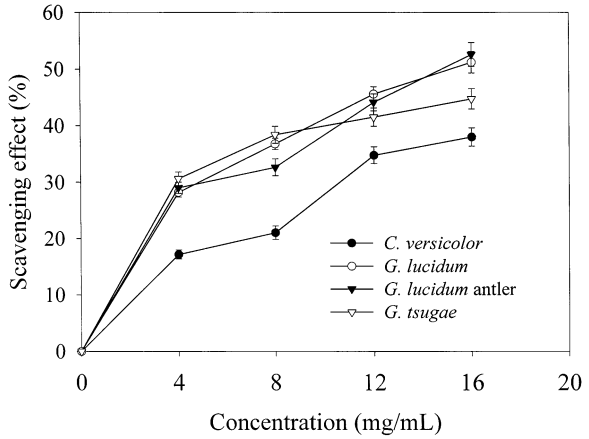

Figure 4. Scavenging effect of methanolic extracts from several medicinal mushrooms on hydroxyl free radical. Each value is expressed as mean ± standard deviation ($n = 3$).

DPPH radical by 92.1%, whereas scavenging effects of methanolic extracts from other specialty mushrooms were 63.3−67.8% (*18*). At 6.4 mg/mL, the methanolic extract from tree oyster mushrooms scavenged DPPH radical by 81.8%, whereas scavenging effects of extracts from other commercial mushrooms were 42.9−69.9% *(18)*. In addition, at 1 mg/mL, methanolic extracts from black and red ear mushrooms scavenged DPPH radical completely (100%), whereas those from snow and jin ear mushrooms scavenged DPPH radical by 94.5% at 0.4 mg/mL and 95.4% at 3 mg/mL, respectively (*20*). However, silver ear mushrooms were not effective in scavenging DPPH radical (71.5% at 5 mg/mL) (*20*).

These results revealed that medicinal mushrooms were free radical inhibitors or scavengers, acting possibly as primary antioxidants. Their methanolic extracts might react with free radicals, particularly of the peroxy radicals, which are the major propagator of the autoxidation chain of fat, thereby terminating the chain reaction (*21*−*23*). The antioxidant activity of natural antioxidants has been shown to be involved in termination of free radical reactions and reducing power (*11*, *24*).

Scavenging Effect on Hydroxyl Radical. The scavenging effects of methanolic extracts from *G. lucidum* and *G. lucidum* antler were highest (51.2 and 52.6%) at 16 mg/mL, respectively (**Figure 4**). *G. tsugae* and *C. versicolor* scavenged hydroxyl radical by 44.7 and 38.0%, respectively. However, the scavenging effect of BHA at 20 mM (3.6 mg/mL) was 23%, whereas that of α-tocopherol at 20 mM (8.6 mg/mL) was 34%.

At 40 mg/mL, scavenging effects were 75.0 and 69.4% for basket stinkhorn and lion's mane and 39.6 and 47.4% for maitake and white matsutake, respectively (*18*). In addition, at 40 mg/mL, the scavenging effect of methanolic extracts from tree oyster mushrooms on hydroxyl radical was 54.3%, whereas other commercial mushrooms scavenged hydroxyl radical by 29.2−36.6% (*18*). At 5 mg/mL, scavenging effects were 10.52−14.01% for methanolic extracts from black, snow, and silver ear musrhooms, whereas no scavenging effect was observed with methanolic extracts from red and jin ear mushrooms (*20*). Similarly, methanolic extracts from *A. camphorata* and Brazilian mushrooms did not scavenge hydroxyl radical (*19*).

These results indicated that many mushrooms are not good scavengers for hydroxyl radical. In addition, Shi et al. (*12*) reported scavenging activity of hydroxyl radicals of caffeine and attributed the alleged anticarcinogenic properties of caffeine to this activity. Accordingly, it was anticipated that the moderate to high scavenging effects of medicinal mushrooms might be associated with some antimutagenic properties.

Chelating Effect on Ferrous Ion. Chelating effects of methanolic extracts from medicinal mushrooms on ferrous ion increased with the increased concentrations (**Figure 5**). At 2.4 mg/mL, chelating effects were in the order *G. lucidum* antler (67.7%) > *G. lucidum* (55.5%) > *G. tsugae* (44.8%) > *C. versicolor* (13.2%). Evidently, *C. versicolor* was not a good ferrous chelator. However, at 20 mM (3.6 mg/mL), the chelating effect of BHA was 36%, whereas that of α-tocopherol at 20 mM (8.6 mg/mL) was 92%. It is contemplated that a higher chelating effect would be observed with the concentration >2.4 mg/mL. Because ferrous ions are the most effective pro-oxidants in the food system (*25*), the higher chelating effects of methanolic extracts from medicinal mushrooms would be beneficial.

Methanolic extracts from *A. camphorata* chelated ferrous ions by 64.4−74.5% at 5 mg/mL, whereas that from Brazilian mushrooms showed an excellent chelating effect of 98.6% at 2.5 mg/mL (*19*). Yen and Wu (*26*) reported that the methanolic extract of *G. tsugae* chelated 95.3% of ferrous ion at 600 ppm (0.6 mg/mL). However, Yen and Wu (*26*) used the method of Decker and Welch (*27*) to determine the chelating effect instead of the method of Shimada et al. (*11*).

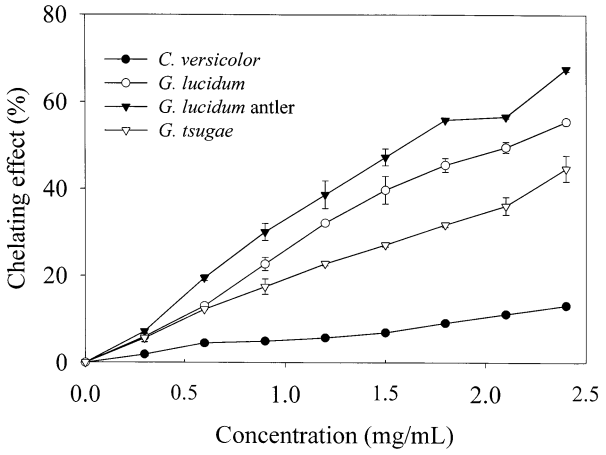

Figure 5. Chelating effect of methanolic extracts from several medicinal mushrooms on ferrous ion. Each value is expressed as mean ± standard deviation ($n = 3$).

The methanolic extract from maitake chelated 70.3% of ferrous ion at 6 mg/mL, whereas at 24 mg/mL, methanolic extracts from black stinkhorn, lion's mane, and white matsutake chelated ferrous ion by 46.4−52.0% (*18*). For commercial mushrooms including winter, abalone, and tree oyster mushrooms and shiitake, their methanolic extracts chelated 45.6−81.6% of ferrous ion at 1.6 mg/mL (*18*). Methanolic extracts from ear mushrooms were good chelators for ferrous ion (85.1−96.5% at 5 mg/mL) (*20*). Summarily, *Ganoderma* mushrooms were good chelators for ferrous ion but *C. versicolor* was not.

Antioxidant Components. Total phenols were the major naturally occurring antioxidant components found in methanolic extracts from medicinal mushrooms (**Table 2**). However, ascorbic acid and β-carotene were not detected, whereas tocopherols were not found in *C. versicolor*. Only γ-tocopherol was found in small amounts (0.13−1.19 mg/g) in *Ganoderma* spp. However, *Ganoderma* mushrooms contained much higher amounts of total phenols (47.25−55.96 mg/g) than *C. versicolor* (23.28 mg/g). Therefore, total phenols might be responsible for the antioxidant properties studied. Phenols such as BHT and gallate were known to be effective antioxidants (*28*).

As compared to the contents of total phenols in methanolic extracts from specialty mushrooms (7.61−16.28 mg/g) (*18*), commercial mushrooms (6.27−15.65 mg/g) (*18*), and ear mushrooms (3.20−8.72 mg/g) (*20*), the highest contents of total phenols in *Ganoderma* (47.25−55.96 mg/g) might be the key components accounting for their better results found in antioxidant activity, reducing power, and scavenging and chelating abilities.

Arbitrarily at 10 mg/mL, contents of total phenols in methanolic extracts from medicinal mushrooms were in the range of 232.8−559.6 μg/mL, much less than BHA and α-tocopherol used at 20 mM (3.6 and 8.6 mg/mL, respectively). Therefore, in addition to these antioxidant components, some other components also existed and contributed in part to the antioxidant properties of medicinal mushrooms. To study the antioxidant mechanisms by some other potential antioxidant components, the fractionation of the methanolic extract and further identification are in progress.

ABBREVIATIONS USED

BHA, butylated hydroxyanisole; BHT, butylated hydroxytoluene; DETBA, 1,3-diethyl-2-thiobarbituric acid; DMPO, 5,5-dimethylpyrroline-*N*-oxide; DPPH, 1,1-diphenyl-2-picrylhydrazyl; EPR, electron paramagnetic resonance; TMM, tetramethyl murexide.

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