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Antioxidant properties of several commercial mushrooms

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Abstract

Winter (strains white and yellow), shiitake (strains 271 and Tainung 1) and oyster mushrooms (abalone and tree oyster mushrooms) were obtained commercially and methanolic extracts were prepared from these mushrooms and their antioxidant properties were studied. The antioxidant activities by the 1,3-diethyl-2-thiobarbituric acid method were moderate to high at 1.2 mg ml^{-1}. Reducing powers were excellent (and higher than 1.28 absorbance) at 40 mg ml^{-1}. Scavenging effects on 1,1-diphenyl-2-picrylhydrazyl radicals were moderate to high (42.9–81.8%) at 6.4 mg ml^{-1}. With regards to the scavenging effect on hydroxyl free radicals, tree oyster mushrooms were the highest (54.3%) at 40 mg ml^{-1} whereas other commercial mushrooms were low. Chelating effects on ferrous ions were 45.6–81.6% at 1.6 mg ml^{-1}. Total phenols were the major naturally occurring antioxidant components found. Overall, tree oyster mushrooms were better in antioxidant activity, reducing power and scavenging abilities and higher in total phenol content.

Keywords: Mushrooms; *Flammulina velutipes*; *Lentinula edodes*; *Pleurotus*; Antioxidant activity; Reducing power; Scavenging effect; Chelating effect; Antioxidant components

1. Introduction

Oxidation is essential to many living organisms for the production of energy to fuel biological processes. However, oxygen-centred free radicals and other reactive oxygen species, that are continuously produced in vivo, result in cell death and tissue damage. Oxidative damage caused by free radicals may be related to aging and diseases, such as atherosclerosis, diabetes, cancer and cirrhosis (Halliwell & Gutteridge, 1984). Although almost all organisms possess antioxidant defence and repair systems that have evolved to protect them against oxidative damage, these systems are insufficient to prevent the damage entirely (Simic, 1988). However, antioxidant supplements, or foods containing antioxidants, may be used to help the human body reduce oxidative damage.

Recently, phytochemicals in food materials and their effects on health, especially the suppression of active oxygen species by natural antioxidants from teas, spices and herbs, have been extensively studied (Ho, Osawa, Huang, & Rosen, 1994). Chinese herbs have been used for diet therapy for several millennia. Some of them are alleged to exhibit significant antioxidant activity (Kim, Kim, Kim, Oh, & Jung. 1994; Su, 1992). Mushrooms are traditional in China and also commonly used as food. In addition to common mushrooms (*Agaricus bisporus*) and paddy straw mushrooms (*Volvariella volvacea*), other commercial mushrooms, including shiitake or forest mushrooms or shiang-ku [fragrant mushrooms; *Lentinula edodes* (Berk.) Pegler], winter or golden mushrooms or enokitake [*Flammulina velutipes* (Curtis: Fries) Sing.], and oyster mushrooms [abalone mushrooms (*Pleurotus cystidiosus* Miller) and tree oyster mushrooms (*P. ostreatus* (Jacquin: Fries) Kummer)] are popular in Taiwan. These mushrooms are highly valued as a centrepiece of Taiwanese cooking.

The nutritional values and taste components of these commercial mushrooms have been thoroughly studied (Yang, Lin, & Mau, 2001). Recently, these commercial mushrooms were found to be medically active in several therapies such as antitumour, antiviral, and immunomodulating treatments (Wasser & Weis, 1999). Although research was focused on the therapeutic effects of these

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commercial mushrooms, little information is available about their antioxidant properties. Our objective was to evaluate the antioxidant properties of these commercial mushrooms, including antioxidant activity, reducing power, scavenging effects on radicals, and chelating effects on ferrous ions. The contents of potential antioxidant components of these commercial mushrooms were also determined.

2. Materials and methods

2.1. Mushrooms

Winter mushrooms (strains white and yellow), shiitake (strains 271 and Tainung 1), abalone mushrooms and tree oyster mushrooms were purchased at a local market in Taichung City, Taiwan. Fresh mushrooms from each species or strain were randomly divided into three samples, ~500 g each. Mushrooms were air-dried in an oven at 40 °C before analysis.

After a fine powder (20 mesh) was obtained using a mill (Restsch Ultra Centrifugal Mill and Sieving Machine, Haan, Germany), dried mushroom sample (10 g) was extracted by stirring with 100 ml of methanol at 25 °C at 150 rpm for 24 h and filtering through Whatman No. 4 filter paper. The residue was then extracted with two additional 100 ml portions of methanol, as described earlier. The combined methanolic extracts were then rotar-evaporated at 40 °C to dryness. The dried extract thus obtained was used directly for analyses of antioxidant components or redissolved in methanol to a concentration of 50 mg ml^{-1} and stored at 4 °C prior to analyses of antioxidant attributes.

2.2. Antioxidant activity

The antioxidant activity was determined by the 1,3-diethyl-2-thiobarbituric acid (DETBA) method (Furuta, Nishiba, & Suda, 1997; Suda, Furuta, & Nishiba, 1994). To various concentrations of methanolic extract from mushrooms (50 μl) were added 50 μl of linoleic acid emulsion (Sigma Chemical Co., St. Louis, MO, 2 mg ml^{-1} 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 the following were sequentially added: 200 μl of 20 mM butylated hydroxytoluene (BHT, Sigma), 200 μl of 8% sodium dodecyl sulphate (SDS, Merck, Darmstadt, Germany), 400 μl of deionised 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 were added, the mixture was mixed and centrifuged at 2000 rpm at 20 °C for 15 min. Ethyl acetate was separated and its absorbance 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 sample being 100%. A higher percentage indicates a lower antioxidant activity.

2.3. Reducing power

The reducing power was determined according to the method of Oyaizu (1986). Various concentrations of methanolic extracts from mushrooms (2.5 ml) were 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) were added, the mixture was centrifuged at 650 rpm for 10 min. The upper layer (5 ml) was mixed with 5 ml of deionised 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 higher absorbance indicates a higher reducing power.

2.4. Scavenging effect on 1,1-diphenyl-2-picrylhydrazyl radicals

Various concentrations of methanolic extracts from mushrooms (4 ml) were mixed with 1 ml of methanolic solution containing 1,1-diphenyl-2-picrylhydrazyl (DPPH, Sigma) radicals, resulting in a final concentration of the DPPH of 0.2 mM. The mixture was shaken vigorously and left to stand for 30 min in the dark, and the absorbance was then measured at 517 nm (Shimada, Fujikawa, Yahara, & Nakamura, 1992).

2.5. Scavenging effect on hydroxyl free radicals

The hydroxyl radical reacted with the nitrone spin trap 5,5-dimethyl pyrroline-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 mixing various concentrations of methanolic extracts from mushrooms (200 μl) 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 set at the following conditions: 3480-G magnetic field, 1.0 G modulation amplitude, 0.5 s time constant, and 200 s scan period (Shi, Dalal, & Jain, 1991).

2.6. Chelating effects on ferrous ions

Chelating effect was determined according to the method of Shimada et al. (1992). To 2 ml of the mixture, consisting of 30 mM hexamine (Wako), 30 mM

potassium chloride (Sigma) and 9 mM ferrous sulphate (Union Chemical Works, Hsinchu, Taiwan), were added various concentrations of methanolic extracts from mushrooms (2 ml) 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.

2.7. Determination of antioxidant components

Ascorbic acid was determined according to the method of Klein and Perry (1982). Methanolic extract from 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 within 15 s at 515 nm. Content of ascorbic acid was calculated on the basis of the calibration curve of authentic L-ascorbic acid (Sigma).

β-Carotene was extracted and analysed as described by Rundhaug, Pung, Read, and Bertram (1988). Methanolic extract from 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, filtered through Whatman No. 4 filter paper and the volume 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 chromato-integrator, 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^{-1} and UV detection at 470 nm. Content of β-carotene was calculated on the basis of the calibration curve of authentic β-carotene (Sigma).

Tocopherols were extracted and analysed according to the method of Carpenter (1979). Methanolic extract from 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. Deionised water (15 ml) was added and the mixture was extracted with 15 ml of *n*-hexane. The organic layer was washed with deionised water to neutral, dried over anhydrous sodium sulphate, 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^{-1} and UV detection at 295 nm. 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, Miller, and Pratt (1984). Methanolic extract from mushrooms (20 mg) was dissolved in a solution of 5 ml of 1.3% HCl in methanol/deionised 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

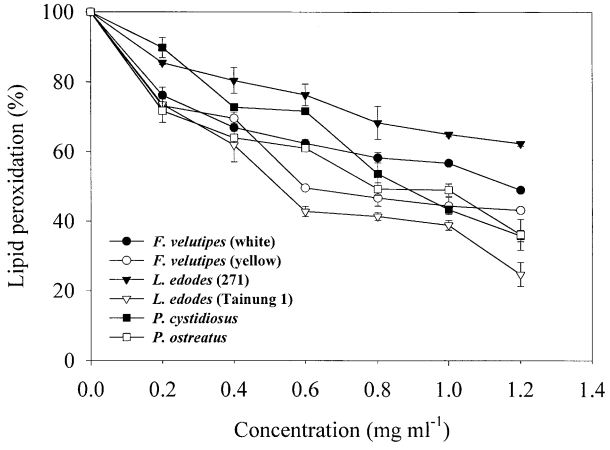

Fig. 1. Antioxidant activity of methanolic extracts from several commercial mushrooms. Each value is expressed as mean±standard deviation ($n=3$).

Table 1
Extraction yield of methanolic extracts from several commercial mushrooms

Mushroom	Yield[a] (g)	Extraction %[b] (w/w)
Winter mushrooms [*Flammulina velutipes* (white)]	3.81±0.11	38.1 B
Winter mushrooms [*Flammulina velutipes* (yellow)]	4.39±0.06	43.9 A
Shiitake [*Lentinula edodes* (271)]	1.88±0.09	18.8 CD
Shiitake [*Lentinula edodes* (Tainung 1)]	1.59±0.09	15.9 D
Abalone mushrooms (*Pleurotus cystidiosus*)	1.95±0.13	19.5 C
Tree oyster mushrooms (*Pleurotus ostreatus*)	1.69±0.11	16.9 D

[a] Extracted from dried mushrooms (10.00 g). Each value is expressed as mean±standard deviation ($n=3$).
[b] Means with different letters within a column are significantly different ($P<0.05$).

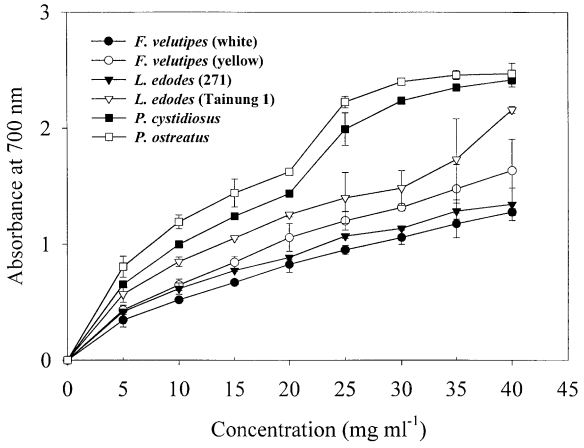

Fig. 2. Reducing power of methanolic extracts from several commercial mushrooms. Each value is expressed as mean ± standard deviation ($n = 3$).

standing, absorbance was measured at 750 nm. Content of total phenols was calculated on the basis of the calibration curve of gallic acid (Sigma).

2.8. 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, Torrie, and Dickey (1997) to determine the least significant difference at the level of 0.05.

3. Results and discussion

3.1. Antioxidant activity

Following the extraction with methanol, two strains of winter mushrooms (strains white and yellow) had the highest yields (38.1 and 43.9%, respectively; Table 1). However, the yields of the rest of the commercial mushrooms ranged from 15.9–19.5%. The higher yields of winter mushrooms were mainly due to the fact that these mushrooms contained higher amounts of soluble sugars and sugar alcohols (32.5 and 29.7% for strains white and yellow, respectively; Yang et al., 2001). Using the DETBA method, all methanolic extracts from commercial mushrooms showed moderate to high antioxidant activities, as evidenced by the moderate to high percentages of lipid peroxidation (24.7–62.3%) at 1.2 mg ml^{-1} (Fig. 1). However, butylated hydroxyanisole (BHA) showed only 66.1% of lipid peroxidation at 10 mg ml^{-1}.

Huang (2000) found that methanolic extracts from a 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^{-1}, respectively. 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^{-1} (Huang, 2000). Lin (1999) found that methanolic extracts from other medicinal mushrooms were extremely effective in inhibiting the lipid peroxidation [6.41% for *Ganoderma lucidum* (Ling-chih), 2.62% for *Ganoderma lucidum* antler, and 2.30% for *Ganoderma tsugae* (Sung-shan-ling-chih) at 0.6 mg ml^{-1}]. However, the methanolic extract from another medicinal mushroom, *Coriolus versicolor* (Yun-chih), showed only 58.6% of lipid peroxidation at 0.6 mg ml^{-1} (Lin, 1999).

Among methanolic extracts from four speciality mushrooms at 1.2 mg ml^{-1}, only *Dictyophora indusiata* (basket stinkhorn) showed an excellent antioxidant activity (2.26% of lipid peroxidation; Lin, 1999). *Grifola frondosa* (maitake) showed a relatively high antioxidant activity (29.8% of lipid peroxidation), whereas *Hericium erinaceus* (lion's mane) and *Tricholoma giganteum* (white matsutake) showed moderate antioxidant activities (48.5% and 67.0% of lipid peroxidation, respectively; Lin, 1999). Chao (2001) reported that methanolic extracts from ear mushrooms, including black, red, jin, snow and sliver ears, showed low to moderate antioxidant activities (57.7–71.5% of lipid peroxidation) at 1.0 mg ml^{-1}. With regard to the antioxidant activities in the DETAB method, methanolic extracts from commercial mushrooms were much lower than those from medicinal mushrooms and basket stinkhorn, and comparable with those from Yun-chih, speciality and ear mushrooms.

3.2. Reducing power

Reducing powers of methanolic extracts from commercial mushrooms were excellent and increased steadily with the increased concentrations (Fig. 2). At 40 mg ml^{-1}, reducing powers were higher than 1.28, and in the order of tree oyster≈abalone mushrooms > strain Tainung 1 > strain yellow≈strain 271≈strain white. At 5 mg ml^{-1}, reducing powers of methanolic extracts from commercial mushrooms were 0.35–0.81. However, reducing powers of BHA and α-tocopherol at 20 mM (3.6 and 8.6 mg ml^{-1}) were 0.12 and 0.13, respectively. The reducing power of ear mushrooms might be due to their hydrogen-donating ability, as described by Shimada et al. (1992). Accordingly, commercial mushrooms might contain higher amounts of reductone, which could react with free radicals to stabilise and terminate radical chain reactions.

Huang (2000) reported that the methanolic extract from Chang-chih showed an excellent reducing power of 0.96–0.97 at 10 mg ml^{-1}, whereas that from Brazilian mushrooms showed a reducing power of 0.86 at 10 mg ml^{-1}. Methanolic extracts from other medicinal

mushrooms, including Ling-chih, antler Ling-chih and Sung-shan-ling-chih, exhibited strong reducing powers of 0.99, 1.25 and 1.26 at 2 mg ml^{-1}, respectively (Lin, 1999). However, a good reducing power of 0.79 was observed with the methanolic extract from another medicinal mushroom Yun-chih at 4 mg ml^{-1} (Lin, 1999).

Among methanolic extracts from four speciality mushrooms, basket stinkhorn showed an excellent reducing power of 1.09 at 3 mg ml^{-1} (Lin, 1999). 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^{-1}, respectively (Lin, 1999).

Chao (2001) reported that methanolic extracts from ear mushrooms, excluding silver ears, showed reducing powers of 0.67–0.82 at 5 mg ml^{-1}. However, the reducing power of the methanolic extracts from ear mushrooms was only determined up to 5 mg ml^{-1}. Since these five ear mushrooms are commercial mushrooms, just like the six commercial mushrooms mentioned earlier, their reducing powers were somewhat comparable to those found in Fig. 2 at 5 mg ml^{-1}.

3.3. Scavenging effect on 1,1-diphenylhydrazyl radicals

Scavenging effects of methanolic extracts from commercial mushrooms on DPPH radicals increased with the increased concentrations and were moderate to high (42.9–81.8%) at 6.4 mg ml^{-1} (Fig. 3). However, the scavenging effects of BHA and α-tocopherol at 20 mM (3.6 and 8.6 mg ml^{-1}) were 0.96 and 0.95, respectively. Excellent scavenging effects (96.3–99.1% and 97.1%) were observed with methanolic extracts from Chang-chih and Brazilian mushrooms at 2.5 mg ml^{-1}, respectively (Huang, 2000). Scavenging effects of methanolic extracts from other medicinal mushrooms were measured at up to 0.64 mg ml^{-1}, and were 24.6, 67.6, 74.4 and 73.5% for Yun-chih, Ling-chih, antler Ling-chih and Sung-shan-ling-chih, respectively (Lin, 1999).

At 6.4 mg ml^{-1}, the methanolic extract from basket stinkhorn scavenged DPPH radicals by 92.1%, whereas scavenging effects of methanolic extracts from other speciality mushrooms were 63.3–67.8% (Lin, 1999). In addition, at 1 mg ml^{-1}, methanolic extracts from black and red ears scavenged DPPH radicals completely (100%) whereas those from snow and jin ears scavenged DPPH radicals by 94.5% at 0.4 mg ml^{-1} and 95.4% at 3 mg ml^{-1}, respectively (Chao, 2001). However, silver ears were not effective in scavenging DPPH radicals (71.5% at 5 mg ml^{-1}; Chao, 2001).

3.4. Scavenging effect on hydroxyl free radicals

The scavenging effect of methanolic extracts from tree oyster mushrooms on hydroxyl free radicals was the highest (54.3%) at 40 mg ml^{-1} (Fig. 4). The rest of the

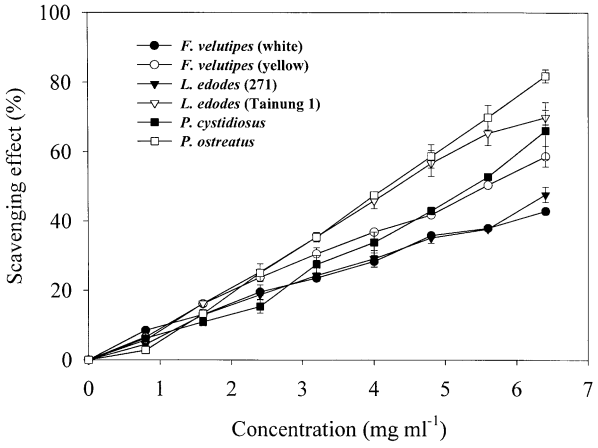

Fig. 3. Scavenging effect of methanolic extracts from several commercial mushrooms on 1,1-diphenyl-2-picrylhydrazyl radical. Each value is expressed as mean ± standard deviation ($n = 3$).

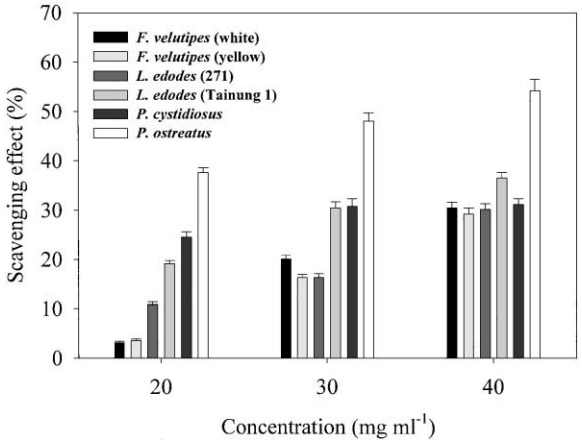

Fig. 4. Scavenging effect of methanolic extracts from several commercial mushrooms on hydroxyl free radical. Each value is expressed as mean ± standard deviation ($n = 3$).

commercial mushrooms scavenged hydroxyl free radicals by 29.2–36.6% at 40 mg ml^{-1}. In addition, the scavenging effect of BHA at 20 mM (3.6 mg ml^{-1}) was 23%, whereas that of α-tocopherol at 20 mM (8.6 mg ml^{-1}) was 34%.

At 5 mg ml^{-1}, scavenging effects were 10.52–14.0% for methanolic extracts from black, snow and silver ears, whereas no scavenging effect was observed with methanolic extracts from red and jin ears (Chao, 2001). Similarly, methanolic extracts from Chang-chih and Brazilian mushrooms did not scavenge hydroxyl free radicals (Huang, 2000). At 40 mg ml^{-1}, methanolic extracts from speciality mushrooms scavenged hydroxyl free radicals by 39.6–75.0% (Lin, 1999). However, at 16 mg ml^{-1}, methanolic extracts from medicinal mushrooms, such as Yun-chih, Ling-chih and Sung-shan-ling-chih, scavenged hydroxyl free radicals by 38.0–52.6% (Lin, 1999). These results indicated that commercial mushrooms are not good scavengers for hydroxyl free radicals.

Table 2
Contents of tocopherols and total phenols of methanolic extracts from several commercial mushrooms

Compound	Content[a] (mg g^{-1})					
	Flammulina velutipes (white)	*Flammulina velutipes* (yellow)	*Lentinula edodes* (271)	*Lentinula edodes* (Tainung 1)	*Pleurotus cystidiosus*	*Pleurotus ostreatus*
Tocopherols	nd	nd	0.12±0.02c (γ)	0.13±0.02c (γ)	0.45±0.02a (γ)	0.24±0.02b (γ)
Total phenols	8.38±0.22d	9.26±0.04c	6.27±0.02e	9.11±0.07c	10.24btc0.04b	15.7±0.10a

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

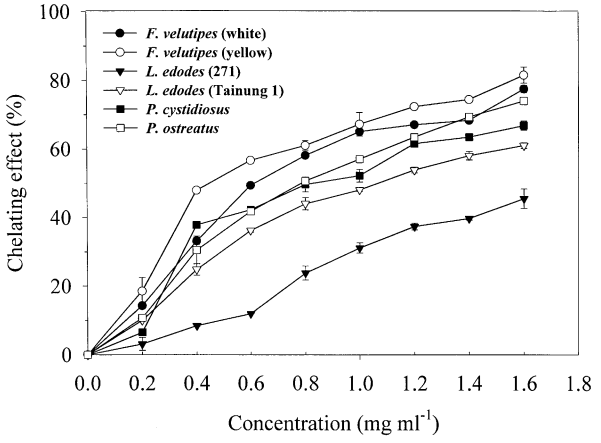

Fig. 5. Chelating effect of methanolic extracts from several commercial mushrooms on ferrous ion. Each value is expressed as mean±standard deviation ($n=3$).

3.5. Chelating effect on ferrous ions

Chelating effects of methanolic extracts from commercial mushrooms on ferrous ions increased with the increased concentrations, and were 45.6–81.6% at 1.6 mg ml^{-1} (Fig. 5). However, at 20 mM (3.6 mg ml^{-1}), the chelating effect of BHA was 36%, whereas that of α-tocopherol at 20 mM (8.6 mg ml^{-1}) was 92%. It is contemplated that a higher chelating effect would be observed at concentrations higher than those used in Fig. 5 (0.2–1.6 mg ml^{-1}). Since ferrous ions are the most effective pro-oxidants in food systems (Yamaguchi, Tatsumi, Kato, & Yoshimitsu, 1988), the higher chelating effects of methanolic extracts from commercial mushrooms would be beneficial.

Methanolic extracts from chang-chih chelated ferrous ions by 64.4–74.5% at 5 mg ml^{-1}, whereas that from Brazilian mushrooms showed an excellent chelating effect of 98.6% at 2.5 mg ml^{-1} (Huang, 2000). The methanolic extract from Yun-chih was not a good ferrous chelator (13.2% at 2.4 mg ml^{-1}), whereas other medicinal mushrooms including Ling-chih, antler Ling-chih and Sung-shan-ling-chih, chelated 44.8–67.7% of ferrous ions at 2.4 mg ml^{-1} (Lin, 1999). Yen and Wu (1999) reported that the methanolic extract of Sung-shan-ling-chih chelated 95.3% of ferrous ions at 600 ppm (0.6 mg ml^{-1}). However, Yen and Wu (1999) used the method of Decker and Welch (1990) to determine the chelating effect instead of the method of Shimada et al. (1992).

The methanolic extract from maitake chelated 70.3% of ferrous ions at 6 mg ml^{-1} whereas, at 24 mg ml^{-1}, methanolic extracts from black stinkhorn, lion's mane and white matsutake chelated ferrous ions by 46.4–52.0% (Lin, 1999). Methanolic extracts from ear mushrooms were good chelators for ferrous ions (85.1–96.5% at 5 mg ml^{-1}; Chao, 2001). As compared with other mushrooms, methanolic extracts from commercial mushrooms would be good chelators for ferrous ions at higher concentrations.

3.6. Antioxidant components

Total phenols were the major naturally occurring antioxidant components found in methanolic extracts from commercial mushrooms (Table 2). However, ascorbic acid and β-carotene were not detected, whereas tocopherols were not found in two strains of winter mushrooms. Only γ-tocopherol was found in small amounts (0.12–0.45 mg g^{-1}) in shiitake, abalone and tree oyster mushrooms. Therefore, total phenols might be responsible for the antioxidant properties studied. Phenols such as BHT and gallate, are known to be effective antioxidants (Madhavi, Singhal, & Kulkarni, 1996). The highest content of total phenols in tree oyster mushrooms might account for the better results found in antioxidant activity, reducing power, scavenging abilities as compared with other commercial mushrooms. Arbitrarily, at 10 mg ml^{-1}, contents of total phenols in methanolic extracts from commercial mushrooms were in the range 83.8–156.5 μg ml^{-1}, which were much lower than BHA and α-tocopherol used at 20 mM (3.6 and 8.6 mg ml^{-1}). Therefore, in addition to these antioxidant components, some other components contributed in part to the antioxidant properties of commercial mushrooms. To study the antioxidant mechanisms by some other potential antioxidant components, the fractionation of the methanolic extract and further identification are in progress.

Overall, for all methanolic extracts from commercial mushrooms, the antioxidant activities were moderate to high at 1.2 mg ml^{-1}. Reducing powers were excellent and increased steadily with the increasing concentrations. Scavenging effects on DPPH radicals increased with the increasing concentrations and were moderate to high at 6.4 mg ml^{-1}. With regard to the scavenging effect on hydroxyl free radicals, tree oyster mushrooms were the highest at 40 mg ml^{-1} whereas the other commercial mushrooms were low. Chelating effects of methanolic extracts from commercial mushrooms on ferrous ions increased with the increasing concentrations, and were 45.6–81.6% at 1.6 mg ml^{-1}. Generally, tree oyster mushrooms were better in antioxidant activity, reducing and scavenging abilities and higher in the content of total phenols. However, shiitake, winter and abalone mushrooms seemed to be comparable in the antioxidant properties assayed.

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