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Antioxidative Activities of Oolong Tea

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While the antioxidative properties of green and black tea have been extensively studied, less attention has been given to these properties in oolong tea. The reducing powers, the 2,2-diphenyl-1-picrylhydrazyl (DPPH) scavenging activities, the amount of total phenolic compounds, the inhibitory effect on $FeCl_2/H_2O_2$ (Fenton reaction system)-induced DNA damage, and the inhibitory effect on erythrocyte hemolysis of an oolong tea water extract (OTE) were evaluated in the present study. The OTE was found to have strong antioxidative activities in all of the model systems tested. When the OTE was separated into different fractions according to molecular weight, it was found that the fractions with higher amounts of phenolic compounds (lower molecular weight) have stronger antioxidative activities. The present results support the concept that oolong tea contains several low molecular weight antioxidants that may have health promotion activities.

KEYWORDS: Antioxidative activity; oolong tea; reducing power; DPPH scavenging activity; DNA damage; membrane oxidation

INTRODUCTION

Reactive oxygen species (ROS) and reactive nitrogen species (RNS) are forms of activated oxygen and nitrogen which include free radicals such as superoxide ion ($O_2^{\bullet -}$) and hydroxyl ($OH^{\bullet}$) and nitric oxide ($NO^{\bullet}$) radicals, as well as non-free-radical species such as hydrogen peroxide (H_2O_2) and nitrous acid (HNO_2) (*1*−*3*). In living organisms, ROS and RNS form through multiple pathways. Normal aerobic respiration, stimulated polymorphonuclear leukocytes and macrophages, and peroxisomes are thought to be the main endogenous sources of most of the oxidants produced by cells (*4*−*6*). Exogenous sources of free radicals include tobacco smoke, ionizing radiation, certain pollutants, organic solvents, and pesticides (*7*−*10*). Because of their high reactivity, free radicals can damage diverse cellular macromolecules, including proteins, carbohydrates, lipids, and nucleic acids. Free-radical damage to these molecules has been implicated in the causation of some degenerative diseases. For example, destructive effects on proteins may play a role in cataract formation, oxidative damage to DNA may be involved in the development of certain cancers, and lipid oxidative damage can contribute to the occurrence and progression of vascular disease (*11*). The human body has several innate defense systems against ROS. However, the innate oxidant defense system is not sufficient to adequately deal with the amount of ROS/RNS produced, and it is now thought that dietary antioxidants are essential for health and well being (*5*, *12*, *13*). In addition to essential nutrients recognized as antioxidants (such as vitamin C, vitamin E, etc.), several other botanical compounds such as flavonoids are thought to function as antioxidants and have been linked to a reduced incidence of disease (*14*, *15*).

Tea is now one of the most widely consumed beverages in the world, its consumption being similar to that of coffee. The tea plant (*Camellia sinensis*) has been widely used for over 5000 years for its specific aroma, taste, and putative positive physiological functions. Traditionally, tea was drunk to improve blood flow, eliminate toxins, and improve resistance to diseases (*16*). Like grapes, apples, and cocoa, tea can be a rich source of flavonoids and other polyphenols. However, the flavonoid content can be affected by different types of processing. Freshly harvested tea leaves are processed differently to produce specific types of tea (green, oolong, and black tea). Green tea is heated and dried to avoid enzymatic oxidation. Oolong tea is semifermented to permit a moderate level of enzymatic oxidation during processing and then dried. Black tea is the most thoroughly oxidized enzymatically. It is the degree of oxidation that affects the polyphenol profile of the tea.

Many studies have shown that green and black teas can have a wide range of pharmaceutical properties including being antihypertensive (*17*), antioxidative (*18*−*20*), antiarteriolemic (*21*), anticarcinogenic (*22*, *23*), and hypocholesterolemic (*24*−*26*). However, the biological effect of oolong tea remains unclear. Xie et al. (*27*) reported that an oolong tea water extract had a high antioxidative activity and a high lipoxygenase inhibitory activity, as compared to black tea. Yen et al. (*28*) calculated the amount of catechins (catechin, gallocatechin, gallocatechin gallate, epigallocatechin, epicatechin gallate, and epigallocatechin gallate) in various tea extracts and found the

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greatest amount in green tea (26.7%) followed by oolong tea (23.2%) and black tea (4.3%). Yen and his colleagues also observed that oolong tea exhibited a stronger antimutagenic activity than did green or black tea. Recently, Ohe et al. (*29*) reported that catechins are not the major components of oolong tea responsible for their anti-genotoxic effects against nitroarenes. This suggests that other unique polyphenols may be present in oolong tea. In the present study, we tested the antioxidative activities of an oolong tea water extract (OTE) and its fractions for their inhibitory effects on $FeCl_2/H_2O_2$ (Fenton reaction system)-induced DNA damage and erythrocyte hemolysis. In addition, we assessed the reducing power and DPPH radical scavenging activity of OTE and its fractions.

MATERIALS AND METHODS

Preparation of Oolong Tea Extract and Oolong Tea Fractions. The method described by Han et al. (*30*) was used to extract polyphenols from oolong tea. The dry leaves of oolong tea (from FuJian Province in China) (100 g) were extracted with boiling water (1 L) for 1 h under reflux. The water solution was concentrated to give a dark brown extract (33 g) that had strong antioxidative activity. A portion (5 g) of the dark brown aqueous extract was fractionated into fractions of molecular weight (MW) >100 kDa, 30−100 kDa, 10−30 kDa, 3−10 kDa, 1−3 kDa, and <1 kDa, using Centricon centrifugal devices (YM3 = 3000 MW, YM 10 = 10,000 MW, YM 30 = 30,000 MW, YM 100 = 100,000) and Diaflo Ultrafilters (YM1 = 1000 MW cutoff membrane) from Amicon, Inc., Beverly, MA.

Determination of the Amount of Total Phenolic Compounds. Total phenolic concentrations were determined as described previously (*31*). To a 200 μL sample of the OTE, 1 mL of Folin-Denis reagent and 0.8 mL sodium carbonate (7.5%) reagent were added. Subsequently, the mixture was incubated in the dark for 45 min. After incubation, the absorbance was measured at 760 nm. Gallic acid (Sigma Chemical Company, St. Louis, MO) was used as a standard for the calibration curve. The phenolic compound content is reported as gallic acid equivalents (μg) using the following linear equation based on the calibration curve:

$$A = 0.0139\,C + 0.0049,\ R^2 = 0.9995$$

where A is the absorbance and C is gallic acid equivalents (μg).

Reducing Power. Total reducing power was determined as described previously (*32*). Briefly, extracts (50−500 μg) in 1 mL of water were mixed with 2.5 mL of phosphate buffer (0.2 M, pH 6.6) and 2.5 mL of potassium ferricyanide $[K_3Fe(CN)_6]$(1%); the mixture was then incubated at 50 °C for 30 min. Afterward, 2.5 mL of trichloroacetic acid (10%) was added to the mixture, which was then centrifuged at 3000 rpm for 10 min. Finally, 2.5 mL of the upper layer solution was mixed with 2.5 mL of distilled water and 0.5 mL $FeCl_3$ (0.1%), and the absorbance was measured at 700 nm. Increased absorbance of the reaction mixture indicated increased reducing power.

DPPH Radical Scavenging Activity. DPPH radical scavenging activity was determined according to the method of Blois with a slight modification (*33*). Briefly, a 1 mM solution of DPPH radical solution in ethanol was prepared, and then 1 mL of this solution was mixed with 3 mL of extract solution in ethanol containing 50−500 μg of dried extract; the mixture was then vortexed vigorously and left for 30 min at room temperature in the dark. The absorbance was measured at 517 nm. This activity is given as percent DPPH scavenging and is calculated as

$$\%\text{DPPH scavenging} = [(\text{control absorbance} - \text{extract absorbance}) \times 100/\text{control absorbance}]$$

Inhibition of Oxidative DNA Damage. Solutions of DNA were prepared by dissolving 1 mg of calf thymus DNA (Sigma Chemical Company, St. Louis, MO) in 1 mL of 1 mM deferoxamine and 20 mM sodium acetate, pH 5.0. The reaction mixtures (400 μL) containing 200 μg DNA, 25 μM $FeCl_2$, 0.03% H_2O_2, and different concentrations of OTE and individual oolong tea fractions were incubated at 37 °C for 30 min as previously described (*34*). Butylated hydroxyanisole (BHA) was used as a positive control. After incubating at 37 °C for 30 min, the solutions containing DNA were digested with nuclease P1 and then with alkaline phosphatase to obtain the corresponding nucleosides (*35*). The HPLC system consisted of an Agilent 1100 series HPLC with a quartenary pump and diode array detector connected to an ESA Coulochem II electrochemical detector with a 5011 analytical cell (E1: 150 mV; E2: 350 mV) and 5020 guard cell set at 550 mV. Separation was achieved with a Supelco C-18 column (150 × 4.6 mm, 3 μm) and mobile phase consisting of 100 mM sodium acetate buffer, pH 5.2 with 7% methanol at a flow rate of 1 mL/min and column temperature of 25 °C. 8-Hydroxy-2′-deoxyguanosine (8-OHdG) was detected electrochemically at 350 mV and 2′-deoxyguanosine (2′dG) was detected at 248 nm using UV detection. DNA oxidation was evaluated as the formation of 8-OHdG per 10^5 2′-dG molecules according to the method of Shigenaga et al. (*35*).

Erythrocyte Hemolysis. Blood was obtained from the abdominal aorta of rats and collected into heparinized tubes. Erythrocytes were separated from plasma and the buffy coat and washed three times with 5 vols of phosphate buffered saline, pH = 7.4 (PBS, GIBCO BRL, Life Technologies, Grand Island, NY). During every wash, the erythrocytes were centrifuged at 3000 × g for 10 min to obtain a packed cell preparation (*36*). After the last wash, the packed erythrocytes were suspended in 4 volumes of PBS solution.

Erythrocyte oxidative hemolysis was induced by 2, 2′-azo-bis (2-amidinopropane) dihydropropane (AAPH), a peroxyl radical initiator (*36*). Addition of AAPH to the suspension of washed erythrocytes induces the oxidation of membrane lipids and proteins, resulting in hemolysis. Two mL of the erythrocyte suspension was mixed with 2 mL of PBS solution containing varying amounts of oolong tea extracts. Two mL of 200 mM AAPH in PBS was then added to the mixture. The reaction mixture was shaken gently while being incubated at 37 °C for 3 h. After incubation, the reaction mixture was removed, diluted with 8 vols of PBS and centrifuged at 3000 × g for 5 min. The absorbance (A) of the supernatant fraction at 540 nm was recorded in a Beckman DU 640 spectrophotometer. Percent inhibition was calculated by the following equation:

$$\%\ \text{inhibition} = [A_{\text{AAPH}} - A_{\text{oolong}}]/A_{\text{AAPH}} \quad (1)$$

where A_{oolong} is the absorbance of the sample containing OTE and the A_{AAPH} is the absorbance of the sample containing no OTE. L-Ascorbic acid was used as a positive control.

Table 1. Amount of Total Phenolic Compounds in Oolong Tea Extract and Individual Oolong Tea Fractions[a]

sample (200 μg)	absorbance (760 nm)	gallate equivalents (μg)
OTE	0.848	60.6
OF1	0.530	37.9
OF2	0.633	45.2
OF3	0.498	35.6
OF4	0.656	46.9
OF5	0.842	60.9
OF6	0.878	62.7

[a] OTE: oolong tea extract; OF1: oolong tea fraction 1, MW >100kDa; OF2: oolong tea fraction 2, MW = 30–100 kDa; OF3: oolong tea fraction 3, MW = 10–30 kDa; OF4: oolong tea fraction 4, MW = 3–10 kDa; OF5: oolong tea fraction 5, MW = 1–3 kDa; OF6: oolong tea fraction 6, MW < 1 kDa.

RESULTS AND DISCUSSION

Amount of Total Phenolic Compounds. As shown in **Table 1**, the OTE was rich in phenolic compounds. With respect to the six fractions isolated, the greatest amount of phenolic compounds was found in OTE, OF5, and OF6. The molecular weights of OF5 and OF6 are less than 3 kDa. The lowest amount of phenolic compounds was measured in OF1 and OF3, which

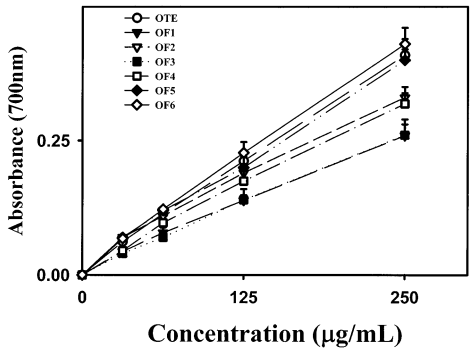

Figure 1. Reducing powers of OTE: oolong tea extract; OF1: oolong tea faction 1, MW > 100 kDa; OF2: oolong tea fraction 2, MW = 30–100 kDa; OF3: oolong tea fraction 3, MW = 10–30 kDa; OF4: oolong tea fraction 4, MW = 3–10 kDa; OF5: oolong tea fraction 5, MW = 1–3 kDa; OF6: oolong tea fraction 6, MW < 1 kDa. Data are expressed as mean ± SD of n = 5–6 samples.

are characterized by higher molecular weights. Preliminary work in our laboratory showed that 100 g of oolong tea leaves produce approximately 4.5 g of tea catechins, which contain 24.1% epigallocatechin (EGC), 3.5% catechin (CT), 6.4% epicatechin (EC), 41.8% epigallocatechin gallate (EGCG), 13.5% gallocatechin gallate (GCG), and epicatechin gallate (ECG) (*unpublished data*). The strong antioxidative activity of OF6 can possibly be attributed to the tea catechins present in oolong tea because their molecular weights are lower than 500. The current work is consistent with other studies which report that oolong tea contains lesser amounts of catechins (low molecular weight polyphenols) than green tea but higher amounts than black tea (*26*, *28*, *37*). The strong antioxidative activity of OF5 is suggestive for the presence of lower molecular weight polymerized tea polyphenols in oolong tea (*38*). Future work is ongoing to identify the active component in OF5.

Reducing Power. Several methods have been developed to measure the efficiency of dietary antioxidants as pure compounds or in food extracts. These methods focus on different mechanisms of the oxidant defense system, that is, scavenging active oxygen species and hydroxyl radicals, reduction of lipid peroxyl radicals, inhibition of lipid peroxidation, or chelation of metal ions. In most cases, irrespective of the stage in the oxidative chain in which the antioxidant action is assessed, most nonenzymatic antioxidative activity (scavenging of free radicals, inhibition of lipid peroxidation, etc.) is mediated by redox reactions. The reducing power of different tea fractions to reduce ferric ions was determined in this study. The highest amount of reducing power was observed in the OTE, OF5, and OF6 fractions, followed by the OF1 and OF3 fractions (**Figure 1**). Interestingly, the OTE, OF5, and OF6 fractions also contained the highest amount of phenolics, with the OF2 and OF4 the second highest, and OF1 and OF3 the lowest in phenolic content (**Table 1**). Given the above, we suggest that there may be a relationship between the amount of total phenolic compounds and reducing power. Consistent with this, there was a statistically significant correlation between these two parameters, $\gamma = 0.98$, $P < 0.01$.

We also measured the reducing power of each fraction using ascorbic acid as a reference compound. As is shown in **Table 2**, the reducing powers of these extracts were markedly lower than that of ascorbic acid. OF5 and OF6, which had the highest reducing power, had reducing powers that were only ~30% that of ascorbic acid.

Table 2. Comparison of Reducing Powers of Ascorbic Acid and Oolong Tea Extracts[a]

sample	absorbance (700 nm)
control	0.0300
ascorbic acid	1.3515
OTE	0.4091
OF1	0.2590
OF2	0.3300
OF3	0.2600
OF4	0.3183
OF5	0.4000
OF6	0.4297

[a] 250 μg oolong tea extract, individual oolong tea fractions (OF1 to OF6), or ascorbic acid was used; in the control there was no extract. High absorbance indicates high reducing power.

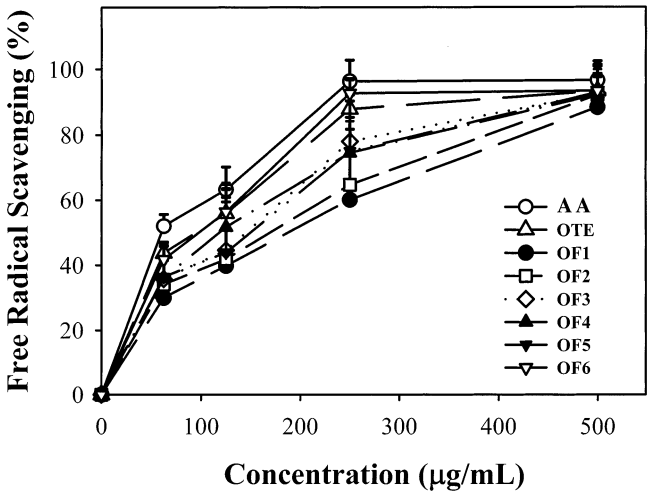

Figure 2. Percent DPPH scavenging activities of oolong tea extract (OTE) and individual oolong tea fractions (OF1 to OF6). Ascorbic acid (AA) was used as a positive control. Data are expressed as mean ± SD of n = 5–6 samples.

The reducing powers determined by the present assay depend on the redox potentials of the compounds present in different tea fractions, characterized by the complexity of their molecules. It is well known that the lower the redox potential, the higher the antioxidant efficiency against free radicals such as the peroxyl or hydroxyl radicals, which have more positive redox potentials (*39*). Therefore, it can be predicted that OTE, OF5, and OF6, which have high reducing power, will keep their high antioxidative activity in other model systems, and thus may also have biological significance for individuals who consume tea.

DPPH Radical Scavenging Activity. This method is based on the reduction of DPPH, a stable free radical. Because of the odd electron of DPPH, it gives a strong absorption maximum at 517 nm by visible spectroscopy (purple color). As the odd electron of the radical becomes paired off in the presence of a hydrogen donor, that is, a free-radical scavenging antioxidant, the absorption strength is decreased, and the resulting decolorization is stoichiometric with respect to the number of electrons captured (*33*). This reaction has been widely used to test the ability of compounds to act as free-radical scavengers or hydrogen donors and to evaluate the antioxidative activity of foods and plant extracts (*40*−*42*). Percent DPPH scavenging activities of OTE, as well as that of all six fractions, were dose-dependent (**Figure 2**). Similar to reducing power, the amount of DPPH scavenging activity appears dependent on the phenolic concentration of the fraction. The highest percent DPPH

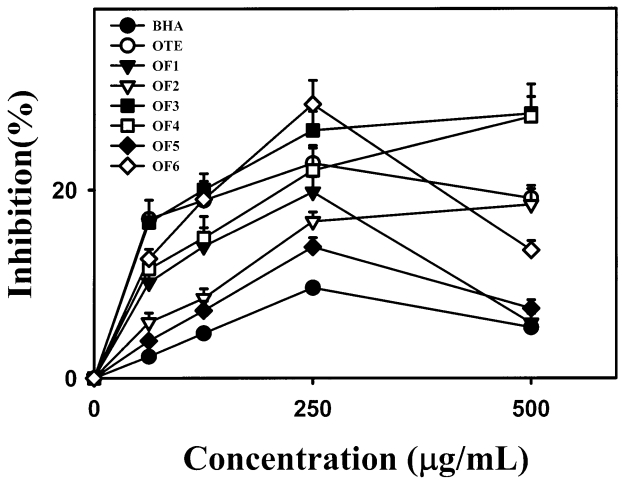

Figure 3. Inhibitory effect of oolong tea extract (OTE) and individual oolong tea fractions (OF1 to OF6) on $FeCl_2/H_2O_2$-induced 8-hydroxy-2′-deoxyguananosine (8-OHdG) formation in calf thymus DNA. Data are expressed as mean ± SD of $n = 5–6$ samples.

scavenging activities were shown by OTE and OF6. The lowest percent DPPH scavenging activities were shown by OF1 and OF2. OF3, OF4, and OF5 demonstrated similar DPPH scavenging activities, although the phenol concentration of OF5 is much higher than that of OF3 and OF4. The strong DPPH scavenging activity of OF6 can be attributed in part to the tea catechins present in oolong tea and probably to some lower molecular weight polymerized polyphenols. The strong DPPH scavenging activities of OF3 and OF4 can probably be attributed to the oxidation procedures of oolong tea manufacturing, as it is suggested that an increase in the antioxidant efficiency of some phenol compounds is observed as a consequence of slight oxidative stress during processing (*40*). The DPPH radical has been widely used to test the free radicals' scavenging ability of various natural products and has been accepted as a model compound for free radicals originating in lipids (*40*). In future work, the assessment of DPPH scavenging activity may be helpful in directing the fractionation, and isolation, of the free-radical scavenging compounds in different oolong tea fractions.

Inhibition of Oxidative DNA Damage. ROS have been known to damage many biological macromolecules, with DNA being a significant target. One of the abundant oxidized DNA bases is 8-OHdG, a well-established biomarker relevant to carcinogenesis and aging (*43*, *44*). Increased levels of 8-OHdG have been found in cancerous tissues (*45*, *46*), and they are thought to contribute to the development of DNA base mutations (*47*, *48*) and activation of certain oncogens, such as H-*ras* and K-*ras* (*49*, *50*). The formation of 8-OHdG can be induced by various environmental factors, such as chemical modification, ionizing irradiation, and UV radiation (*43*, *44*). In view of the potential carcinogenecity of 8-OHdG, it can be argued that dietary habits that reduce this type of DNA damage may reduce the risk for certain cancers and other degenerative disorders. In the present study, we determined the quenching effects of OTE and OF1−OF6 on the formation of 8-OHdG initiated by $FeCl_2/H_2O_2$. Under the conditions used, we observed that the Fenton reaction significantly increased 8-OHdG formation in DNA. As is depicted in **Figure 3**, OTE and all the individual oolong tea fractions (OF1−OF6) had stronger quenching effects on $FeCl_2/H_2O_2$-induced 8-OHdG than BHA. In this system, ascorbic acid demonstrated a very strong pro-oxidant effect (at 62.5 μg/mL, the inhibition is −151.73 ± 6.8%). Although ascorbic acid is a strong antioxidant, a high intake of ascorbic acid may lead to, in some situations, a pro-oxidant activity in the body when free transition metals are available at the same time (*51*). The ranking of the antioxidative activities of the individual oolong tea fractions in this assay, in general, followed the same ranking order as in the other assays, with minor differences. The low molecular weight fraction, OF6, exhibited the strongest inhibition, while the high molecular weight fraction, OF1, exhibited the weakest. At high concentrations, OTE and especially OF6 demonstrated pro-oxidant effects as is depicted in **Figure 3**. It is well known that the same flavonoids can behave as either antioxidants or pro-oxidants, depending on the flavonoid concentration and the free-radical source. In the current work, the flavonoids acted as antioxidants against free radicals at lower concentrations (0−250 μM) but demonstrated pro-oxidant activity at high concentration (500 μM). This is consistent with Cao's report which showed that $Cu^{2+}-H_2O_2$ initiated pro-oxidant activity of a flavonoid makes its antioxidative activity decrease with increasing flavonoid concentration after the maximal hydroxyl radical absorbing value is reached (*52*).

Table 3. Inhibitory Effect (%) of Oolong Tea Extract (OTE) and Individual Oolong Tea Fractions on AAPH-Induced Hemolysis of Rat Erythrocytes in Vitro[a]

	6.25 μg/mL	12.5 μg/mL	25 μg/mL
ascorbic acid	72.68 ± 1.28[c]	80.19 ± 6.96[c]	87.14 ± 6.50[c]
OTE[b]	69.52 ± 3.21[c]	77.10 ± 7.71[c]	83.12 ± 4.25[c]
OF1	45.65 ± 3.60[d]	58.51 ± 6.90[d]	74.38 ± 5.70[d]
OF2	51.98 ± 5.00[e]	71.82 ± 4.30[e]	77.99 ± 3.90[d]
OF3	57.50 ± 3.32[f]	69.00 ± 3.60[e]	78.66 ± 8.65[d]
OF4	63.14 ± 9.67[c]	72.74 ± 3.60[e]	73.11 ± 6.70[d]
OF5	65.41 ± 6.42[c]	76.65 ± 5.52[e]	75.65 ± 0.29[d]
OF6	70.94 ± 5.19[c]	79.04 ± 9.01[c]	82.46 ± 4.54[c]

[a] Data are expressed as mean ± SD of $n = 5–6$. [b] OTE: oolong tea extract; OF1: oolong tea fraction 1, MW >100kDa; OF2: oolong tea fraction 2, MW = 30–100 kDa; OF3: oolong tea fraction 3, MW = 10–30 kDa; OF4: oolong tea fraction 4, MW = 3–10 kDa; OF5: oolong tea fraction 5, MW = 1–3 kDa; OF6: oolong tea fraction 6, MW < 1 kDa. [c,d,e,f] In the same column with different superscripts differ significantly at $p < 0.05$.

Erythrocyte Hemolysis. It is well recognized that the oxidation of polyunsaturated fatty acids in biological membranes can lead to the formation and propagation of lipid radicals, uptake of oxygen, rearrangement of the double bonds in unsaturated lipids, and even destruction of membrane lipids. Many of these biochemical activities can lead to the production of breakdown products that are highly toxic to most cell types. Lipid oxidation of rat erythrocyte membrane mediated by AAPH induces membrane damage and subsequently hemolysis (*36*, *53*). The OTE and oolong tea fractions tested inhibited lipid oxidation in the cell membrane (**Table 3**). OTE, OF6, and OF5 were more effective against lipid oxidation in the erythrocyte membrane than the other fractions. However, six fractions demonstrated dose-dependent inhibition effects toward RBC hemolysis, with the protective effects reaching their maximum at 25 μg/mL. This concentration is much lower than the saturated concentration in the reducing power and DPPH radical scavenging activity systems, indicating that the RBC hemolysis is a more sensitive system for evaluating the antioxidant properties of the tea fractions.

The inhibitory characteristics of the oolong tea fractions were determined by multiple factors including the system used, their hydrophobicity/hydrophilicity, and the total polyphenolic components present in the individual fractions. Among the four systems used, OTE and OF6 demonstrated the highest protective effects and were also highest in their total phenol concentration. Han et al. obtained similar results with OTE and its fractions

corresponding to molecular weights below 1 kDa and 1−3 kDa, which enhanced lipolysis at a concentration of 1000 μg/mL (*30*). Hibasami and Nakahara demonstrated that a polyphenol trimer (molecular weight around 2000), found in an oolong tea fraction corresponding to molecular weight below 3 kDa, can trigger apoptosis and suppress the proliferation of stomach cancer cells (*38*). This fraction has also been reported to strongly inhibit the water-insoluble glucan-synthesizing enzyme, glucosyltransferase I (Gtase-I), of *Streptococcus sobrinus* 6715 (*54*). Interestingly, the inhibitory effect of this fraction on Gtase-I is much stronger than that of typical green tea catechins (catechin, epicatechin, epigallocatechin, epicatechin gallate, and epigallocatechin gallate) (*54*). Further experiments are now in progress to identify the inhibitory substances in oolong tea.

In summary, our observations demonstrate that OTE and its individual fractions have DPPH scavenging activity and reducing power, and they can provide protection against DNA oxidation. In addition, we observed that certain oolong tea fractions can inhibit erythrocyte hemolysis. The inhibitory characteristics of the oolong tea fractions were determined by multiple factors including the assays used, the hydrophobicity/hydrophilicity of the compounds, and the total number and location of hydroxyl groups on the active polyphenolic components present in the individual fractions. Among the six oolong tea fractions, OF5 and OF6 exhibited the strongest antioxidative activities in all of the assays used. Future experiments will be aimed at purifying and characterizing the specific components that are responsible for the relatively high antioxidative activities of these fractions.

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