JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Chain-Breaking Antioxidant Activity and Cyclic Voltammetry Characterization of Polyphenols in a Range of Green, Oolong, and Black Teas

VITALY ROGINSKY,† TATYANA BARSUKOVA,† CHYONG F. HSU,‡ AND PAUL A. KILMARTIN*,‡

N.N. Semenov Institute of Chemical Physics, Russian Academy of Sciences, Kosygin Street 4, 117977 Moscow, Russia, and Department of Chemistry, The University of Auckland, Private Bag 92019, Auckland, New Zealand

A series of eight green, eight oolong, and 17 black teas have been analyzed for polyphenol content by absorbance at 272 nm and cyclic voltammetry response at an inert carbon electrode, a new method developed to provide a rapid measure of easily oxidizable polyphenols in beverages. The chain-breaking antioxidant activity of the teas has also been determined during the chain oxidation of methyl linoleate in a pH 7.4 micellar solution, for which realistic kinetic parameters have been derived. While higher mean values were obtained for green teas than for oolong and black teas, the differences were not large, and the spread of values within each type was considerable. The absorbance at 272 nm correlated well with the cyclic voltammetry response only for green teas and black teas taken on their own. The cyclic voltammetry measure and the antioxidant activity correlated well only for the green teas, where the polyphenol content is dominated by epigallocatechin gallate.

KEYWORDS: Antioxidant activity; cyclic voltammetry; tea; polyphenols; catechins

INTRODUCTION

The extraction of leaves of the *Camellia sinensis* plant and its consumption in the form of green, oolong, and black teas are one of the main sources of polyphenols in the diet. The green tea leaf contains very high levels of flavan-3-ols, along with various flavonols. Partial fermentation to oolong tea or a fuller fermentation to black tea produce a range of theaflavins and thearubigins (*1*). These compounds have shown potential antioxidant activity through their abilities to quench reactive oxygen species and chelate metal ions in various in vitro and ex vivo tests (*2*, *3*). Tea polyphenols could thus play a protective role against the development of diseases induced by oxidative stress including cancer and cardiovascular diseases. Despite indications that the bioavailability of tea polyphenols in the blood plasma is limited (*4*), significant antioxidant activity in the gastrointestinal tract has been suggested (*5*). It has also been recognized that in future clinical trials, the type of tea and its preparation need to be determined carefully in order to assess the impact of tea polyphenol intake more effectively (*2*).

The polyphenolic content or antioxidant activity of teas have been examined in several studies. Using a chemiluminescence assay, considerable variation was found in the antioxidant activity of 20 commercially available teas, with higher levels found in the stronger teas (*6*). In other tests involving inhibition of low-density lipoprotein oxidation (*7*) or radical scavenging (*8*), green, oolong, and black teas produced similar values. It has also been shown that 75−90% of the antioxidant activity in green teas for scavenging of the $ABTS^{\bullet+}$ radical cation (*9*), quenching of Fremy's radical and the galvinoxyl radical (*10*), and reduction of Fe^{2+} (*11*) can be accounted for by the flavan-3-ols present. An issue that remains with these antioxidant assays is that different tests rank the antioxidant activity of the individual flavan-3-ols in different orders, while the cooperative effects of the polyphenols remain largely unknown.

The levels of individual polyphenols were seen to span a wide range in further studies involving 40 black teas, where a link was seen between total theaflavin content and tea quality (*12*), and other papers (*13*, *14*), in which the differences were ascribed to variations in growing and storage procedures. A large survey of 95 green and 55 black teas also showed how the variety, growing and manufacturing conditions, tea particle size, and extraction method all affect the level of polyphenols in tea infusions (*15*). While it appears that most of the polyphenols present in green teas can be identified in high-performance liquid chromatography (HPLC) analyses, a large fraction of the polyphenols in black teas remains unknown.

In previous research, we have shown how the chain-breaking antioxidant activity of polyphenols and tea samples can be assessed during the oxidation of methyl linoleate in a micellar solution (*16*, *17*). This lipid-based system with well-derived kinetic parameters provides a more direct and realistic determination of total antioxidant activity of teas than many other

* To whom correspondence should be addressed. Tel: (64-9)3737999 ext. 88324. Fax: (64-9)3737422. E-mail: p.kilmartin@auckland.ac.nz.
† Russian Academy of Sciences.
‡ The University of Auckland.

10.1021/jf030086q CCC: $25.00
Published on Web 08/14/2003

in vitro tests (*17*). We have also shown how cyclic voltammetry of a diluted tea sample can provide a semiquantitative measure of the levels of epigallocatechin gallate in green and oolong teas and the overall level of catechol-containing phenolics in black teas (*18*).

In this study, we compare the chain-breaking antioxidant activity and the cyclic voltammetry estimation of the polyphenol content for eight green, eight partially fermented (oolong), and 17 black commercially available teas. Whereas only representative tea samples were considered in our previous method development studies (*16*−*18*), in this paper, we consider a wider range of teas, as available to the consumer. This allows comparisons to be made between two rapidly obtained polyphenol measures, considered as alternatives to more lengthy HPLC analyses, and the chain-breaking antioxidant activity.

MATERIALS AND METHODS

Chemicals. For the antioxidant assay, methyl linoleate and HPMC were purchased from Sigma. NaH_2PO_4 and Na_2HPO_4 were purchased from Merck. The water soluble initiator, AAPH, was obtained from Polysciences. Aqueous solutions were prepared with doubly distilled water. The testing system was 5−10 mM methyl linoleate, oxidized at 37.0 ± 0.1 °C in a micellar solution of 50 mM Triton X-100 in a phosphate buffer, pH 7.40 ± 0.02, with 2−4 mM AAPH as an initiator. The buffer was prepared by mixing 50 mM solutions of NaH_2PO_4 and Na_2HPO_4 without adding any acid or base. Solutions of the individual phosphates used for the buffer preparation were purged from traces of transition metals by Chelex-100 resin (Bio-Rad) using a batch method. For cyclic voltammetry, NaH_2PO_4 was purchased from Panreac Quimica and Na_2HPO_4 was purchased from Scharlau Chemie.

Teas. A series of eight green, eight oolong, and 17 black teas were purchased from local shops in the Auckland region (**Table 1**). The Darjeeling teas, while usually classified as black teas, have been included in the list of oolong teas, since their fermentation time is limited and the oxidation of flavan-3-ols is also only partially completed (*1*, *14*). The tea extracts were prepared by hanging a tea bag or placing 2.0 g of loose tea (weighed to ±0.0005 g), for 3 min in 200 mL of Milli-Q grade water at 100 °C, which was agitated with a magnetic stirrer at 250 rpm. All of the data obtained with the bagged teas were normalized for brewing 2.0 g of tea in 200 mL of water.

Total Chain-Breaking Antioxidant Capacity. The kinetics of oxygen consumption during the oxidation of methyl linoleate were studied using a 5300 Oxygen Biological Monitor (Yellow Springs Instruments Co., U.S.A.) computerized with a ME-32 Multimeter (Metex, Korea) interface. The protocol for the determination of *N*, the concentration of kinetic chains that can be terminated by a beverage (*16*, *17*), a measure of the total antioxidant capability, is presented in **Figure 1**. A single run included two stages, the determination of the rate of free radical generation (initiation), R_{IN} (trace A), followed by the measurement of *N* proper (trace B). Prior to each stage, the system was bubbled with pure oxygen for a few minutes. R_{IN} was determined at the beginning of the run by the inhibitor method, taking the induction period, t_{IND}, observed when HPMC, a reference antioxidant, was added (trace A). For HPMC, the stoichiometric coefficient of inhibition (the number of active free radicals terminated by one molecule of antioxidant) has been reported to be as high as two (*19*). Thus, R_{IN} may be calculated from t_{IND} using the equation:

$$R_{IN} = 2 \cdot [\text{HPMC}]/t_{IND} \quad (1)$$

The first stage was completed by measuring the rate of the noninhibited oxidation, R_0, which was determined by the slope of the $[O_2]$ trace after the end of the induction period (trace A). Next, the response for the addition of the tea extract was studied (trace B). *N* was calculated from the induction period caused by the tea extract using the equation:

$$N = R_{IN} \cdot t_{IND}/(v/V) \quad (2)$$

Table 1. Chain-Breaking Antioxidant Activity (*N*, mM), Cyclic Voltammetry Response (Q_{400}, μC), and Absorbance at 272 nm (D_{272}) for a Range of Teas

tea	form	brand	*N* (mM)	Q_{400} (μC)	D_{272}
		green			
1	loose	China Young Hyson	10.7 ± 0.4	10.2 ± 0.2	43.8
2	loose	Japanese Green Sechna	9.63 ± 0.56	10.5 ± 0.2	41.1
3	loose	Chinese Gunpowder Special	9.55 ± 0.03	11.5 ± 0.2	41.4
4	loose	Japanese Bancha	4.57 ± 0.37	6.27 ± 0.12	19.5
5	bag	Ever Spring	9.41 ± 0.15	10.5 ± 0.2	34.5
6	bag	Red Seal	15.1 ± 1.3	12.8 ± 0.3	44.8
7	bag	Healtheries	8.78 ± 0.33	8.5 ± 0.2	28.2
8	bag	Dilmah	11.5 ± 0.5	11.1 ± 0.3	44.4
		oolong			
9	loose	Choice Formosa	2.47 ± 0.10	3.87 ± 0.12	22.1
10	loose	Darjeeling Superior Fancy Oolong	4.86 ± 0.23	6.69 ± 0.14	34.4
11	loose	Darjeeling TGFOP[a]	5.03 ± 0.36	9.8 ± 0.2	39.6
12	loose	Darjeeling FTGOP[a]	5.53 ± 0.41	8.74 ± 0.12	46.1
13	loose	Shen Long Chinese	7.38 ± 0.16	9.37 ± 0.05	41.5
14	bag	Ever Spring	9.09 ± 0.12	9.9 ± 0.2	39.1
15	bag	Lipton Ming Han Ching	2.37 ± 0.28	5.92 ± 0.05	23.1
16	bag	Jinshanbo	8.09 ± 0.36	10.3 ± 0.1	43.2
		black			
17	loose	English Leaf Blend	5.59 ± 0.24	8.27 ± 0.13	47.4
18	loose	China Keemun Congou	2.99 ± 0.14	3.64 ± 0.06	42.4
19	loose	Lapsang Souchong Chong	3.05 ± 0.06	3.12 ± 0.06	26.9
20	loose	Assam GBOP Sepon Tea Garden	9.33 ± 0.33	8.51 ± 0.10	66.4
21	loose	Irish Breakfast	7.74 ± 0.17	7.7 ± 0.2	70.5
22	loose	Russian Samovar	2.97 ± 0.23	3.90 ± 0.08	36.1
23	loose	Brooke Bond PG Tips	7.75 ± 0.04	6.24 ± 0.06	49.4
24	bag	Lipton Yellow Label	7.80 ± 0.05	5.32 ± 0.02	49.9
25	bag	Clipper Finest English Blend	7.40 ± 0.28	6.5 ± 0.2	43.6
26	bag	Twinings English Breakfast	8.00 ± 0.27	5.66 ± 0.11	40.8
27	bag	First Choice	5.34 ± 0.39	3.98 ± 0.06	31.1
28	bag	Bell	5.82 ± 0.22	3.79 ± 0.04	34.4
29	bag	Choysa Classic	5.91 ± 0.34	5.86 ± 0.11	46.3
30	bag	Carrefour	7.00 ± 0.61	6.5 ± 0.2	48.6
31	bag	Edglets	6.04 ± 0.35	3.54 ± 0.06	30.9
32	bag	Twinings Irish Breakfast	7.73 ± 0.16	5.59 ± 0.02	50.3
33	bag	Dilmah	9.63 ± 0.10	5.98 ± 0.06	42.0

[a] These Darjeeling teas have been included with the oolong teas, given that their fermentation is also partial.

where v and V are the volume of the added tea extract and that of the testing system (3.6 mL), respectively.

Particular attention was given to calculating t_{IND} from $[O_2]$ traces. Most commonly, t_{IND} is defined as the intersection of the tangent of the kinetic curve during the propagation phase and the time axis. However, this "graphical" procedure has no reliable theoretical basis. More correctly, the value of t_{IND} may be determined by integration:

$$t_{IND} = \int_{\infty}^{0} \{1 - (R/R_0)^2\}\, dt \quad (3)$$

where R is the rate of the inhibited chain oxidation. The deduction of eq 3 was given in ref *20*.

Cyclic Voltammetry. Two milliliters of the tea extracts was diluted 50 times in a pH 7.0 phosphate buffer, consisting of 65% w/v 50 mM Na_2HPO_4 and 35% w/v 50 mM NaH_2PO_4 (*18*). Cyclic voltammograms were recorded from −100 to 400 mV (Ag/AgCl, +207 mV vs she), using a Bioanalytical Systems (BAS) 100A electrochemical analyzer. The working electrode was a 3 mm glass carbon disk electrode (BAS M-2012), which was cleaned by polishing using 3 μm alumina powder (PK-4 polishing kit) for 2 min between runs. The current response due to the phosphate buffer blank was then subtracted away, and the average results for at least three runs were normalized for 2.0 g of tea.

Absorbance at 272 nm. Tea extracts were diluted 100 fold in Milli-Q water and placed in a 1 cm quartz cell. The absorbance at 272

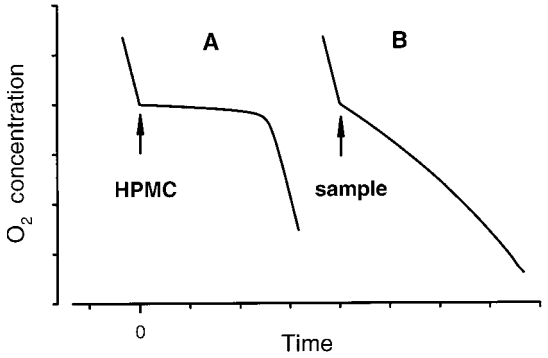

Figure 1. Protocol for the determination of the antioxidant activity of tea extracts (see the text).

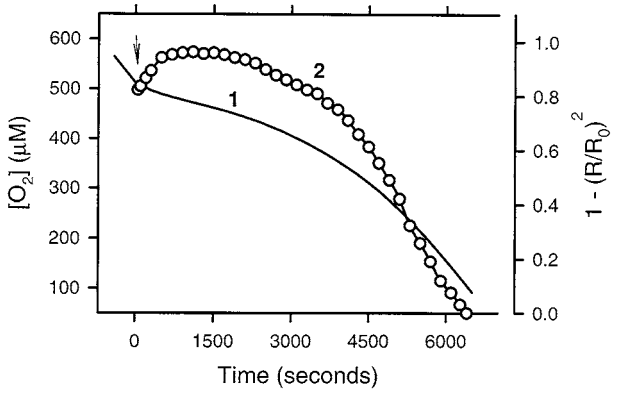

Figure 2. Effect of adding a 6 μL extract of green tea number 6 to 5 mM methyl linoleate oxidized in a 50 mM micellar solution of Triton X-100 in 50 mM phosphate buffer, pH 7.40, induced by 4 mM AAPH at 37.0 °C. 1, $[O_2]$ trace, and 2, $[O_2]$ trace, using the axis of $1 - (R/R_0)^2$ vs time. The arrow shows the moment of tea extract addition.

nm was measured against water using a Schimadzu UV 1240 spectrophotometer. The resulting values (between 0.2 and 0.7) were multiplied by 100 to give the absorbance of the undiluted tea extracts (**Table 1**). However, no attempt was made to subtract away a value due to other compounds present, such as the variable level of proteins and amino acids, which can make up over 10% of the tea solids by weight (*1*).

RESULTS AND DISCUSSION

Chain-Breaking Antioxidant Capacity. All of the tea extracts displayed a pronounced ability to retard methyl linoleate oxidation (see **Figure 2** for a typical green tea and **Figure 3** for a black tea). The key feature of the $[O_2]$ traces observed after adding the tea extract was that during the fast part of induction period, R decreased with time (i.e., an increase in inhibition) and then increased after going through an inflection point. This tendency was more pronounced for the green teas than for the black teas. The inflection point appeared later when a larger volume of tea extract was added (data not shown). After some time, R reached a stationary level, equal to R_0 (**Figures 2** and **3**), due to depletion of tea antioxidants. The following reasons for such an unexpected shape of $[O_2]$ have been suggested earlier (*16*): (i) oxidative transformation of some of the original polyphenols to oligomers with a higher reactivity toward the peroxy radical and (ii) the release of polyphenols from complexes with nonphenolic tea components, such as proteins and polysaccharides.

N values calculated from $[O_2]$ traces using eqs 2 and 3 are presented in **Table 1**. While the average values of N for the green teas, 9.9 ± 1.9 mM, exceeded the average N for the oolong teas, 5.6 ± 1.7 mM, and for the black teas, 6.5 ± 1.7 mM, the

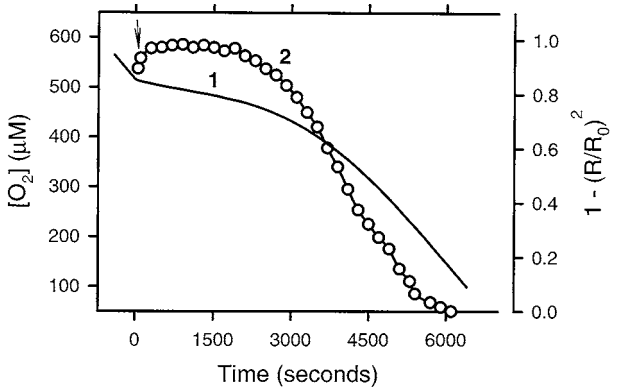

Figure 3. Effect of adding a 6 μL extract of black tea number 33 to 5 mM methyl linoleate oxidized in a 50 mM micellar solution of Triton X-100 in 50 mM phosphate buffer, pH 7.40, induced by 4 mM AAPH at 37.0 °C. 1, $[O_2]$ trace, and 2, $[O_2]$ trace, using the axis of $1 - (R/R_0)^2$ vs time. The arrow shows the moment of tea extract addition.

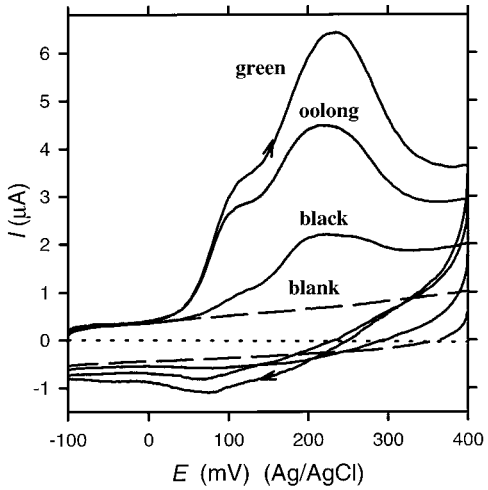

Figure 4. Cyclic voltammograms of green tea number 6, oolong tea number 13, and black tea number 31 diluted 50 times in the pH 7.0 phosphate buffer and a blank run in the buffer itself, measured at a 3 mm glassy carbon electrode at 100 mV s^{-1}.

difference in N between black and oolong teas was not significant and was within the standard errors. At the same time, the spread in N values for each type of tea was rather high, with the most active and least active samples in each group differing by a factor of 3−3.5. On average, the extracts of bagged teas were found to have a higher antioxidant activity than the loose teas, by 30−60%, depending on the tea type (**Table 1**). Most likely, this may be assigned to the fact that the leaves in the tea bags are commonly smaller in size than the loose teas, resulting in a faster extraction of polyphenols (*15*). At the same time, it was possible to find loose teas with rather high N values, for instance green tea number 1 and black tea number 20 (**Table 1**).

Cyclic Voltammetry. The cyclic voltammetry responses of representative green, oolong, and black teas are shown in **Figure 4**. The positive current for oxidation of the diluted green and oolong teas has been shown to be mainly due to epigallocatechin gallate, the major polyphenol and oxidizable component present (*18*). Other flavan-3-ols are also oxidized at potentials less than 400 mV. The peak or shoulder at 120 mV is due to pyrogallol (triphenol) groups on flavonoid B-rings, while the main peak at 200−220 mV is due to gallate or catechol groups (*18*). On the other hand, the pyrogallol group is much diminished in black teas, following the oxidative transformations that occur during

fermentation. The size of the residual current shoulder at 120 mV is an indication of the level of epigallocatechin gallate and epigallocatechin that remains after the fermentation process. In black teas, the various catechol and gallate groups on theaflavins and other polyphenols produce the main peak at around 230 mV (*18*). As a result, the ratio of the current at 200 mV to that at 120 mV went from 1.5−2.0 for the green teas and 1.7−2.6 for the oolong teas to 2.7−4.0 for the black teas.

Figure 4 also includes the response due to the buffer, which is subtracted away when calculating peak currents. The peak current is theoretically directly proportional to concentration, but a nonlinear response is obtained with the flavonoids due to contamination of the electrode by phenolic oxidation products (*21*). Because of the broad nature of the current peaks, an alternative measure of the current response, and hence of concentration, is to take the integral of the current to 400 mV, labeled Q_{400} and measured in μC. A similar measure was used for wine samples and was compared to various total phenol measures (*21*, *22*).

The current response for the 33 teas, as Q_{400} values, is included in **Table 1**. As with the chain-breaking antioxidant activity, a wide variation in values was seen, with some black teas (range 3.1−8.5 μC) showing higher values than some green teas (6.3−15.1 μC). However, the average value for the green teas (10.2 ± 2.0 μC) was higher than the oolong (8.1 ± 2.3 μC) and the black teas (5.5 ± 1.7 μC). On the other hand, the correlation with the chain-breaking antioxidant activity was only moderate ($r^2 = 0.55$) for the 33 teas but was better for the green teas ($r^2 = 0.82$) than for the black teas ($r^2 = 0.44$). The rather poor correlation between Q_{400} and N for the black teas may be due to a relatively high content of phenolics, which only oxidize at potentials greater than 400 mV, which evidently do not contribute to Q_{400}. Generally, the strongest teas showed the largest N and Q_{400} values, and the weakest teas showed the lowest values. However, for teas of similar total polyphenol contents but with different proportions of individual compounds, the more intense response given by particular polyphenols in one test, but not the other, will lead to differences in the overall measures (*16*−*18*).

Absorbance at 272 nm. A comparison was also made with the total phenols present as broadly measured by the peak absorbance at 272 nm. This approach is widely used to provide a measure of total phenols in wines due to the strong absorbance in the 270−280 nm range exhibited by most phenolic compounds (*23*). For each type of tea, reasonable correlations were obtained between the cyclic voltammetry response and the 272 nm absorbance. In **Figure 5**, the currents at 120 mV (near the pyrogallol peak) and 200 mV (near the catechol and gallate peak) are plotted against the absorbance at 272 nm. For a given 272 nm absorbance, a higher current is produced at 120 mV for the green teas as compared to the black teas, corresponding to lower levels of epigallocatechin gallate in the black teas. Between these two groups lie the partially fermented oolong teas. Some of the oolong teas behaved very similar to the green teas, while the results for others were closer to the black teas, reflecting the varying degrees of oxidation to which the oolong teas were subjected.

Concluding Comments. This work confirmed earlier reports (*6*−*8*) that different types of tea (green, oolong, and black) do not differ greatly in antioxidant activity, although green teas tend to show somewhat higher values. Unfortunately, the ability of the main components of black tea, theaflavins and thearubigens, to inhibit lipid peroxidation has never been quantitatively determined. However, a semiquantitative determination for

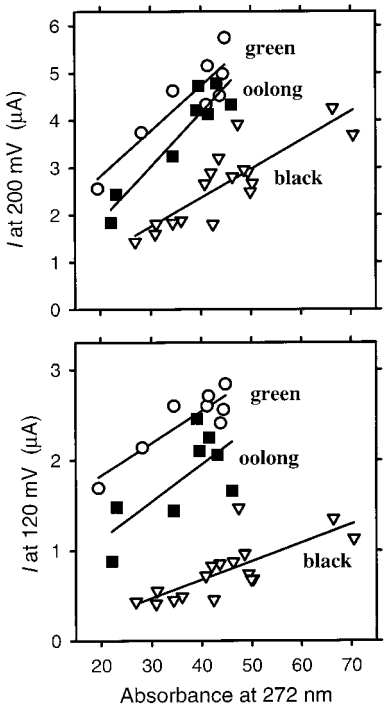

Figure 5. Comparison of the current produced in cyclic voltammograms of diluted tea samples at 120 (near the pyrogallol peak) and 200 mV (near the catechol peak), with the absorbance of the teas at 272 nm ($r^2 = 0.79$ at 120 mV and 0.81 at 200 mV for the green teas; $r^2 = 0.52$ at 120 mV and 0.88 at 200 mV for the oolong teas; and $r^2 = 0.56$ at 120 mV and 0.71 at 200 mV for the black teas).

theaflavins (using the model of low-density lipid oxidation induced by Cu) showed that the antioxidant activity of several theaflavins is comparable with or even superior to that of the flavan-3-ols (*24*). The reported reactivities of theaflavins to $O_2^{\bullet -}$ also exceeded those of epigallocatechin and epigallocatechin gallate (*25*). Thus, the dramatic decrease in flavan-3-ol concentrations, when going from green tea to black tea, is partly compensated for by the rather high antioxidant activities of theaflavins and thearubigens.

Speaking strictly, it is not correct to compare the antioxidant activity of one tea sample with another. This is due to a significant influence of tea leaf size, the properties of tea bag materials, and some other factors of the extraction that occur during brewing. Regarding the level of polyphenols, it is also very difficult to compare different teas using antioxidant tests or other phenolic measures (*14*). What may be compared are the properties of tea extracts, as consumed by a tea drinker. At the same time, we can always compare the data obtained by different methods for the same sample and assess its relevance to the antioxidant activity of the tea.

Of the measures examined in this paper, the absorbance at 272 nm is the easiest to obtain. Plots of the cyclic voltammetry measure of polyphenol content against D_{272} produced two distinct curves for green and black teas. This means that the 272 nm absorbance only works as an indicator of polyphenol concentration, and hence of relative antioxidant activity, for a particular type of tea. In the case of oolong teas, the variable degree of fermentation and tea composition means that D_{272} is a less reliable measure of polyphenol concentration.

More information about polyphenol composition is given by cyclic voltammetry. The total level of components that are oxidized at potentials less than 400 mV, estimated by the Q_{400} value, provides a measure of the reducing strength of the tea

extract. As a measure of total phenols, it is selective for pyrogallol, catechol, and gallate containing polyphenols and does not include polyphenols that only oxidize at more positive potentials. In the case of green teas, the peak current at 120 mV indicates the level of epigallocatechin gallate (*18*), and the value is obtained more rapidly and at less expense than is required using HPLC analysis.

While tea extracts with a higher overall total phenol content are expected to show a higher antioxidant activity, this is not necessarily the case, owing to the variable antioxidant activity of different individual polyphenols. Our approach for determining chain-breaking antioxidant activity, by using the model of controlled chain oxidation in micelles, seems to be the most direct and reliable, if we take into consideration the idea that antioxidant activity is, by definition, the capability of a substance to retard chain oxidation. The majority of other reported methods for antioxidant activity, including the very popular ABTS test (*26*), are not direct and are thus less informative.

It is generally recognized that the beneficial effects of drinking tea are due the antioxidative activity of polyphenols. To increase our understanding of the mechanisms underlying the biological activity of tea, further studies are needed in several directions. First, further efforts should be made to specify the composition of the phenolics contained in teas, including the more minor components. Second, more reliable information is required on the bioavailability of the various components in the tea extracts. Finally, more reliable and reproducible approaches to determining antioxidative activity of tea extracts, and of individual polyphenols, should be developed. The results presented in the current article show that cyclic voltammetry and chain-breaking antioxidant activity measured in micelles can provide important tools to achieve these goals.

ABBREVIATIONS USED

HPMC, 6-hydroxy-2,2,5,7,8-pentamethylbenzochroman; AAPH, 2,2′-azobis(2-amidinopropan)dihydrochloride; ABTS, 2,2′-azino-bis-(3-ethylbenzthiazoline-6-sulfonic acid).

LITERATURE CITED

(1) Harbowy, M. E.; Balentine, D. A. Tea Chemistry. *Crit. Rev. Plant Sci.* **1997**, *16*, 415−480.

(2) McKay, D. L.; Blumberg, J. B. The role of tea in human health: an update. *J. Am. Coll. Nutr.* **2002**, *21*, 1−13.

(3) Dufresne, C. J.; Farnworth, E. R. A review of the latest research findings on health promotion properties of tea. *J. Nutr. Biochem.* **2001**, *12*, 404−421.

(4) Hodgson, J. M.; Puddey, I. B.; Croft, K. D.; Burke, V.; Mori, T. A.; Cacetta, R. A.-A.; Beilin, L. J. Acute effects of ingestion of black and green tea on lipoprotein oxidation. *Am. J. Clin. Nutr.* **2000**, *7*, 1103−1107.

(5) Halliwell, B.; Zhao, K.; Whiterman, M. The gastrointestinal tract: a major site of antioxidant action? *Free Radical Res.* **2000**, *33*, 819−830.

(6) Robinson, E. E.; Maxwell, S. R. J.; Thorpe, G. H. G. An investigation of the antioxidant activity of black tea using enhanced chemiluminescence. *Free Radical Res.* **1997**, *26*, 291−302.

(7) Richelle, M.; Tavazzi, I.; Offord, E. Comparison of the antioxidant activity of commonly consumed polyphenolic beverages (coffee, cocoa, and tea) prepared per cup serving. *J. Agric. Food Chem.* **2001**, *49*, 3438−3442.

(8) Du Toit, R.; Volsteedt, Y.; Apostolides, Z. Comparison of the antioxidant content of fruits, vegetables and teas measured as vitamin C equivalents. *Toxicology* **2001**, *166*, 63−69.

(9) Rice-Evans, C. A.; Miller, N. J.; Paganga, G. Structure-antioxidant activity relationships of flavonoids and phenolic acids. *Free Radical Biol. Med.* **1996**, *20*, 933−956.

(10) Gardner, P. T.; McPhail, D. B.; Duthie, G. G. Electron spin resonance spectroscopic assessment of the antioxidant potential of teas in aqueous and organic media. *J. Sci. Food Agric.* **1998**, *76*, 257−262.

(11) Record, I. R.; Lane, J. M. Simulated intestinal digestion of green and black teas. *Food Chem.* **2001**, *73*, 481−486.

(12) Wright, L. P.; Mphangwe, N. I. K.; Nyirenda, H. E.; Apostolides, Z. Analysis of caffeine and flavan-3-ol composition in the fresh leaf of *Camellia sinesis* for predicting the quality of the black tea produced in Central and Southern Africa. *J. Sci. Food Agric.* **2000**, *80*, 1823−1830.

(13) Arts, I. C. W.; Van de Putte, B.; Hollman, P. C. H. Catechin contents of foods commonly consumed in The Netherlands. 2. Tea, wine, fruit juices, and chocolate milk. *J. Agric. Food Chem.* **2000**, *48*, 1752−1757.

(14) Khokhar, S.; Magnusdottir, S. G. M. Total phenol, catechin, and caffeine contents of teas commonly consumed in the United Kingdom. *J. Agric. Food Chem.* **2002**, *50*, 565−570.

(15) Astill, C.; Birch, M. R.; Dacombe, C.; Humphrey, P. G.; Martin, P. T. Factors affecting the caffeine and polyphenol contents of black and green tea infusions. *J. Agric. Food Chem.* **2001**, *49*, 5340−5347.

(16) Roginsky, V., Baruskova, T. Total chain-breaking antioxidant capability of some beverages as determined by the Clark electrode technique. *J. Med. Food* **2001**, *4*, 219−229.

(17) Roginsky, V. Chain-breaking antioxidant activity of natural polyphenols as determined during the chain oxidation of methyl linoleate in Triton X-100 micelles. *Arch. Biochem. Biophys.* **2003**, *416*, 261−270.

(18) Kilmartin, P. A.; Hsu, C. F. Characterisation of polyphenols in green, oolong, and black teas, and in coffee, using cyclic voltammetry. *Food Chem.* **2003**, *82*, 501−512.

(19) Burton, G. A.; Doba, T.; Gabe, E. J.; Hughes, L.; Lee, F. L.; Prassad, L.; Ingold, K. U. Autoxidation of biological molecules. 4. Maximizing the antioxidant activity of phenols. *J. Am. Chem. Soc.* **1985**, *107*, 7053−7065.

(20) Loshadkin, D.; Roginsky, V.; Pliss, E. Substituted p-hydroquinones as a chain breaking antioxidant during the oxidation of styrene. *Int. J. Chem. Kinet.* **2002**, *34*, 162−171.

(21) Kilmartin, P. A.; Zou, H.; Waterhouse, A. L. A cyclic voltammetric method suitable to characterise antioxidant properties of wine and wine phenolics. *J. Agric. Food Chem.* **2001**, *49*, 1957−1965.

(22) Kilmartin, P. A.; Zou, H.; Waterhouse, A. L. Correlation of wine phenolic composition versus cyclic voltammetry response. *Am. J. Enol. Vitic.* **2002**, *53*, 294−302.

(23) Iland, P.; Ewart, A.; Sitters, J.; Markides, A.; Bruer, N. *Techniques for Chemical Analysis and Quality Monitoring During Winemaking*; Patrick Iland Wine Promotions: South Australia, 2000; p 98.

(24) Leung, L. K.; Su, Y.; Chen, R.; Zhang, Z.; Huang, Y.; Chen, Z. Y. Theaflavins in black tea and catechin in green tea are equally effective antioxidants. *J. Nutr.* **2001**, *131*, 2248−2251.

(25) Jovanovic, S. V.; Hara, Y.; Steenken, S.; Simic, M. G. Antioxidant potential of theaflavins. A pulse radiolysis study. *J. Am. Chem. Soc.* **1997**, *119*, 5337−5343.

(26) Arts, M. J. T. J.; Dallinga, J. S.; Voss, H.-P.; Haenen, G. R. M. M.; Bast, A. A critical appraisal of the use of the antioxidant capacity (TEAC) assay in defining optimal antioxidant structures. *Food Chem.* **2003**, *80*, 409−414.

Received for review February 4, 2003. Accepted June 22, 2003.

JF030086Q