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Original Article

Major flavonoids in dry tea

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Abstract

Data in the food composition analytical literature were reviewed. They were then aggregated and assembled into a provisional database for the major flavonoids in brewed tea. As levels of fermentation increased from green to oolong to black tea, the major flavan-3-ol profiles changed. Total catechins were 13.6 g/100 g in green and 4.2 g/100 g dry weight in black tea. A discussion of methods to calculate the flavonoid content in tea is presented.

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1. Introduction

Teas are used as beverages worldwide although consumers vary in their preferences for the degree of fermentation, taste and color (Balentine et al., 1997). Green tea, the non-fermented tea, is widely consumed in China and Japan, and health benefits such as cancer risk reduction have been suggested (Ahmad and Mukhtar, 1999; Bushman, 1998; Fujiki et al., 1996; Liao et al., 2001;

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Mitscher et al., 1997; Mukhtar and Ahmad, 1999). Oolong, which is partially fermented by endogenous enzymes in the tealeaf, is drunk in many countries. Black tea, the most highly fermented, is popular in Western countries and dominates the market economically (Balentine et al., 1997). Pu'er, a rare tea fermented by anaerobic bacteria rather than the enzymatic fermentation processes that characterize the other teas, is consumed almost exclusively in Asia (Balentine et al., 1997; Bokuchava and Skobeleva, 1980).

The flavonoids are largely responsible for the distinctive taste and color of tea, and are therefore of commercial interest (Fig. 1). There is also growing interest in the positive health effects of tea, which are thought to be associated with the presence of tea flavonoids (Kohlmeier et al., 1997; Kuroda and Hara, 1999; Middleton et al., 2000; Moline et al., 2000; Mukhtar and Ahmad, 2000; Nijveldt et al., 2001; Riemersma et al., 2001; Stoner and Mukhtar, 1995; Wang, 2000; Weisburger, 1999; Wiseman et al., 1997; Yang et al., 1997; Yang and Landau, 2000).

The most common flavonoids in tea are the flavan-3-ols (flavanols or flavans), which are present in relatively large amounts in tea compared to their levels in other foods. The flavan-3-ols provide the “signature” flavonoid pattern that is distinctive in tea. The flavan-3-ol subclasses are ranked by degree of polymerization. The catechins are monomers (catechin, epicatechin, epicatechin gallate, epigallocatechin, and epigallocatechin gallate), the theaflavins are dimers (theaflavin, theaflavin 3-gallate, theaflavin 3'-gallate, theaflavin 3,3'-digallate), and the derived tannins the arubigins are oligomers of unknown structure. Other flavonoids, including the flavonols (quercetin, kaempferol, myricetin) and flavones (apigenin and luteolin), are also present but in lesser amounts than the flavan-3-ols.

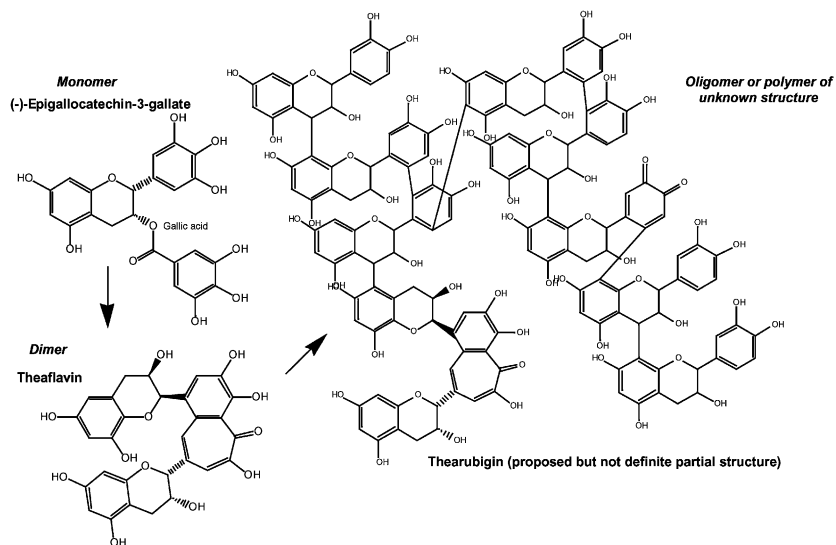


Fig. 1. Structures of significant flavan-3-ols in tea. The arubigins are oligomers of unknown structure. Other flavonoids, including the flavonols (quercetin, kaempferol, myricetin) and flavones (apigenin and luteolin), are also present but in lesser amounts than the flavan-3-ols.

This article provides provisional estimates of the content of 10 flavan-3-ols, 3 flavonol, and 2 flavone compounds in dry tea, which were derived from a systematic and selective review of the available food composition analytical literature.

2. Materials and methods

The literature search strategy for relevant research is described in detail in an earlier article (Peterson and Dwyer, 2000). Table 1 provides specific details pertaining to the searches for tea. Nineteen articles met the inclusion criteria for this study.

The flavonoid classes (and compounds) included were: flavan-3-ols (catechin, epicatechin, epigallocatechin, epicatechin gallate, epigallocatechin gallate, theaflavin, theaflavin 3-gallate, theaflavin 3'-gallate, theaflavin 3,3'-digallate, and the arubigins), flavonols (kaempferol, myricetin, and quercetin) and flavones (apigenin and luteolin).

Table 1
Article search and disposition

Number of articles	Description of criteria for acceptance/elimination
594	Food Science Technology Abstracts (FSTA) chosen AGRICOLA (United States Department of Agriculture) and CAB (Commonwealth Agricultural Bureau) would increase the horticultural (breeding, genetic, pesticide, etc) citations; Medline (Index Medicus) dropped many food science journals in the mid 1980s. FSTA search results using Germplasm Resources Information Network (GRIN) botanical, Latin, and common names and synonyms Flavonoid terms 6177 citations Tea genus terms 172 CAMELLIA 38 THEA Tea terms 4578 TEA 909 TEAS Collection of tea terms 4691 Cross flavonoid and tea terms 594 articles/citations
+ 14	14 additional articles obtained subsequent to search
608	All citation abstracts were read for possible quantitative data
- 500	That did not appear to have quantitative data
108	With possible quantitative data or other useful information
- 17	Unavailable, 12 foreign articles and 5 US articles requested but not received
91	Obtained
- 12	On physiology, epidemiology, and the general subject “antioxidants and flavonoids”
79	Data on flavonoid content of teas.
- 49	Without quantitative data were 2 botanical, 4 characterization of compounds, 7 experimental, 8 isolation of compounds, 1 lecture, 20 method, and 7 review
30	Articles had quantitative data
- 11	On isoflavonoids (1), on lignans (1), methodological problems (2), non-commercial tea not caught earlier (1), weight of tea used not given (1), uncommon samples and did not dry the tea (1), on rutin which is not the only source of quercetin in tea (1), on theaflavins using the Flavognost method (3).
19	Articles met criteria for inclusion in this study

The quality of food composition analytic data in each study was reviewed using an approach originally developed and used in compiling other food composition tables (Holden et al., 1987, 2002) by the United States Department of Agriculture (USDA). The basic ideas of this system were used to make decisions about the inclusion or exclusion of the data sources after consultation with collaborators familiar with the chemistry and food science of tea. Five quality assessment criteria were used: number of samples, sample handling, sampling plan, analytical method, and analytical quality control. These criteria were applied to all data in the food composition analytical literature that could be identified for the tea flavonoids of interest (Mangels et al., 1993).

With respect to number of samples, each analytical value reported in the articles was regarded as a tea “datapoint”. Each datapoint was treated conservatively as an *N* of 1 and no weighting factor was applied to adjust for the number of samples included. Datapoints described as “not detected” were assigned a value of zero. “Trace” values were assigned to be 71% of the limit of quantification for the method used based on previous statistical work (Mangels et al., 1993).

Only teas sold commercially were used. Data were categorized by the type of tea (eg., black, green, oolong, and Pu'er) and evaluated by brand (Lipton, Twining, etc.), origin (India, Kenya, etc.), product (Earl Gray, English Breakfast, etc.), and variety (Assam, Keemun, etc.).

Although not presented in this paper, our database also included details about amount of water, extraction time, and tea weight. Only tea samples with water infusions of known strength were included because comparisons between aqueous and non-aqueous extraction of dry tea were not available in the literature. It is thought that water extracts 15% less flavonoids from dry tea than non-aqueous (usually methanolic) solvents (Beecher, 2000). In addition, for food composition data, we needed to use data as tea would be prepared for human consumption (i.e., brewed with water). For comparative purposes (because the strength of tea infusions often varies), flavonoid amounts were standardized to mg/100 g dry tea from mg/L, mg/kg, percent, or other measures provided in the original articles. In general, flavonoids are stable; an exception is catechins in dilute aqueous solutions. Therefore, protection from oxidation and ultraviolet light were not a major concern in tea analysis. Also, teas are usually analyzed shortly after brewing or freeze-dried and frozen for later analysis.

The analytic methods used varied from article to article, but the most common was high-performance liquid chromatography (HPLC). Spectrophotometric methods for total theaflavins and the arubigins and column chromatographic (CC), gas chromatographic (GC), and capillary electrophoretic (CE) separation for individual compounds were also used. Since the use of different analytic methods might introduce considerable variability, before aggregating the flavan-3-ol data, we ascertained if the values from different methods were similar to values obtained using the currently preferred analytical method (HPLC). No significant differences by analytic method were evident between HPLC and CC or CE data for 8 flavan-3-ol compounds that were examined using the Mann–Whitney or Kruskal–Wallis tests (non-parametric). Only 2 compounds in black tea (catechin and epicatechin by GC and HPLC) and 2 compounds in green tea (epicatechin gallate and epigallocatechin gallate by CE and HPLC) were significantly different by analytical method. None of the datapoints from CC, GC, or CE were outliers (e.g., greater than 1.5 times the interquartile range) when compared to the HPLC data, and so all of these data were retained.

Total theaflavin data comprised four methods—HPLC, CE, CC, and Flavognost (an older spectrophotometric method). The Flavognost data were significantly different from HPLC and column chromatography data. In addition, the molecular weight used for this method could not be confirmed. As a result the Flavognost data were not used for this study. One article (Lee and Ong, 2000) provided both CE and HPLC data on black, green, oolong, and Pu'er tea. However, the brewing method was 30 min at 90°C in accordance with local custom. This is exceptionally long compared to most brewing methods. As a result we did not use these data.

There is currently no HPLC method for the arubigins. Although there is a newer method of calculating the arubigin content (Wiseman et al., 2001), no the arubigin data utilizing that method and no comparisons between this new method and the current spectrophotometric method have been found in the literature. Since the arubigins contribute significantly to the color and body of tea and are a major portion of black tea flavonoids, currently available spectrophotometric data have been included in this article. Eventually, these the arubigin data will be superseded by data utilizing newer and more accurate methods.

Analytical quality control was also reviewed. Older articles often did not mention these details although it may be that such controls were employed; newer articles were more detailed.

Before statistical analysis, the lowest and highest datapoints for each compound and for each type of tea were reviewed to verify amounts recorded in the database. Box plots were calculated for each flavonoid in black, green, oolong and Pu'er tea, on which sufficient data (at least three datapoints) were available to detect possible outliers.

For our research 19 studies were acceptable. Eleven unacceptable studies were archived or discarded (three were for analyses of compounds not included in this study and eight were for methodologically unusable analyses, Table 1). Fourteen articles with acceptable data on flavan-3-ols were available. Nine articles had data on catechins, four on theaflavins, and three on the arubigins. Eight of the nine articles on catechins employed HPLC.

Of a total of 892 flavan-3-ol datapoints, most of the analyses were of the flavan-3-ol compounds in black (51%, 454 datapoints) and green (33%, 298 points) tea with fewer analyses for oolong (9%, 83 points) and Pu'er (7%, 57 points) teas.

The data for individual flavan-3-ols were variable. There were 31 “near” (1.5–3.0 times the interquartile range) outliers and 11 “far” (>3.0 times interquartile range) outliers. Only two outliers were below the median. Twenty-three per cent ($\frac{3}{13}$) of the black, 57% ($\frac{4}{7}$) of the green, 57% ($\frac{4}{7}$) of the oolong, and 86% ($\frac{6}{7}$) of the Pu'er tea box plots had no “near” or “far” outliers. Compounds in black tea with four outliers were epicatechin and epigallocatechin. In black tea, the variety with the most outliers was Ceylon Uva Highland, which had outlying values in four catechin compounds, suggesting that growing conditions contributed to a greater than usual production of catechins. However, total catechins had only one outlier each for black and oolong tea. This may suggest that the individual catechin compounds are intermediates of each other and possibly of other compounds such as theaflavins and the arubigins.

Seven published studies and one unpublished report, all using HPLC methodology, were available on the flavonol content of teas. Four studies were discarded, one (Justesen et al., 1998) because the strength of the infusion could not be determined and the others because an alcoholic rather than a water extraction was used initially (Engelhardt et al., 1992; Finger et al., 1991; Finger and Engelhardt, 1991). When published studies determine what the differences are between

aqueous and non-aqueous extractions for each class (and compound), these tea studies will be incorporated.

Of the remaining four studies, three (de Vries, 1994; Hertog et al., 1993; Toyoda et al., 1997) hydrolyzed the sugars before quantifying the flavonols and one (Price et al., 1998) did not. Out of 105 flavonol datapoints available, 90 datapoints were for black (86%), 9 datapoints for green (8%), and 3 each for oolong (3%) and Pu'er (3%) teas.

Using the black tea flavonol data, for quercetin and kaempferol there was no significant difference between hydrolyzed aglycones and summed aglycones of the glycosides, but for myricetin a difference was present (P 0.01 two-tailed, Mann–Whitney test), probably due to the many teas in which myricetin was not detected. One study (Price et al., 1998), which measured flavonol glycosides, reported no myricetin glycosides for nine teas out of 13, perhaps due to the tea-processing methods used (Lakenbrink et al., 2000). No 'trace' amounts and no outlying values were found for flavonols.

Only three studies measured flavones, and all used HPLC methodology. Two performed acid hydrolysis before measuring the flavone content of brewed tea (de Vries, 1994; Hertog et al., 1993). Neither of these studies detected flavones for three possible reasons: co-elution of the flavones with flavonols, hydrolysis may have destroyed some of the flavone aglycones or not hydrolyzed the C-glycosides of flavones, and what little was present was below the limit of detection. The third study (Engelhardt et al., 1993), which measured the presence of flavone glycosides, was specific and thorough. There were 39 flavone datapoints, 32 for black (80%), four for green (10%), two for oolong (5%), and two for Pu'er teas (5%). No 'trace' amounts and no outliers in the data for flavones were found.

The flavones are a minor class of flavonoids in tea and are all C-glycosides (sugar attached to the carbon on the aglycone). The aglycone apigenin has at least five glycosides and the aglycone luteolin has at least two glycosides that may be present (Engelhardt et al., 1993).

Descriptive statistics were calculated for each compound, the distributions were tested for normality, and statistical tests were performed to compare the different types of tea. Outliers were included in the data as they are indicative of the variability encountered with these compounds. Chemical compound names were also standardized, and aglycones were calculated as needed for the flavonols and flavones.

3. Results

3.1. Flavan-3-ols

Provisional estimates for the major flavan-3-ols in the different types of tea are provided in Table 2. Substantial data were available for black and green but less for oolong and Pu'er teas.

Flavan-3-ols constituted the major flavonoid class in tea from the quantitative standpoint. The most common flavan-3-ols were the catechins (catechin, epicatechin, epicatechin gallate, epigallocatechin, epigallocatechin gallate), and the theaflavins (theaflavin, theaflavin 3-gallate, theaflavin 3'-gallate, theaflavin 3,3'-digallate) and the the arubigins.

The different types of tea varied considerably in total amount of flavan-3-ols and individual flavan-3-ol compounds. Total flavan-3-ols were approximately 17 g per 100 g dry black tea and

Table 2

Flavan-3-ols, flavonols, and flavones in black, green, oolong, and Pu'er teas (mg/100 g dry tea, water extraction data only)

Teas	Black						Green					
	Mean	S.D.	Median	Range	N	Ref	Mean	S.D.	Median	Range	N	Ref
<i>Flavan-3-ols</i>												
Catechin	167	105	130	35–480	41	1–4	24	17	20	0–80	30	2,8
Epicatechin	316	262	212	60–1100	44	1–6	793	411	696	190–2000	50	2,5,6,8,9
Epicatechin 3-gallate	923	480	791	280–2380	44	1–6	1755	1056	1600	328–4630	50	2,5,6,8,9
Epigallocatechin	1257	850	1110	29–3870	44	1–6	1712	1466	940	100–5440	50	2,5,6,8,9
Epigallocatechin 3-gallate	1393	1098	1085	140–5090	44	1–6	8975	5923	9435	1182–18 810	50	2,5,6,8,9
Galocatechin	126	90	89	56–278	8	4						
Galocatechin 3-gallate							316	234	240	160–1380	28	8
Total catechins by sum of means	4182						13 575					
<i>Total catechins^a</i>	3937	2139	4000	1019–12480	39	2,3,4,7	15 667	4884	16 290	7820–24 110	38	2,8,9
Theaflavin	162	98	148	40–527	38	3,10,11						No data
Theaflavin 3-gallate	105	81	93	7–260	28	10,11						No data
Theaflavin 3'-gallate	178	1005	166	13–413	28	10,11						No data
Theaflavin 3,3'-digallate	144	123	97	7–496	38	3,10,11						No data
Total theaflavins by sum of means	589											No data
<i>Total theaflavins^a</i>	568	364	462	67–1448	38	3,10,11						No data
The arubigins	12 490	2471	12 945	4530–16 933	20	12,13,14						No data
Total flavan-3-ols	17 262						13 576					
<i>Flavonols</i>												
Kaempferol	132	47	133	44–213	30	15,16,17	130	34	149	91–150	3	15,18
Myricetin	25	21	25	0–80	30	15,16,17	101	43	120	52–131	3	15,18
Quercetin	210	76	213	89–416	30	15,16,17	175	48	156	140–230	3	15,18
Total flavonols	367						406					
<i>Flavones</i>												
Apigenin	54	19	53	21–91	16	19	77	73	77	25–128	2	19
Luteolin	5	2	5	3–9	16	19	8	9	8	2–14	2	19
Total flavones	59						85					
<i>Total Flavonoids</i>	17 687						14 066					

Table 2 (continued)

Teas	Oolong						Pu'er					
<i>Flavan-3-ols</i>												
Catechin	17	21	10	0–70	11	2,8	0	0	0	0	7	8
Epicatechin	259	101	240	120–450	13	2,6,8	81	90	49	0–250	9	6,8
Epicatechin 3-gallate	707	298	690	170–1210	13	2,6,8	40	54	10	0–120	9	6,8
Epigallocatechin	510	387	390	180–1640	13	2,6,8	157	126	100	12–400	9	6,8
Epigallocatechin 3-gallate	3861	2031	4120	736–7110	13	2,6,8	120	126	90	0–370	9	6,8
Gallocatechin												
Gallocatechin 3-gallate	93	42	90	40–160	9	8	13	22	0	0–50	7	8
total catechins by sum of means	5447						411					
<i>Total catechins^d</i>	6037	1812	5700	2100–8870	11	2,8	493	298	510	140–940	7	8
Theaflavin						No data						No data
Theaflavin 3-gallate						No data						No data
Theaflavin 3'-gallate						No data						No data
Theaflavin 3,3'-digallate						No data						No data
Total theaflavins by sum of means												
<i>Total theaflavins^a</i>						No data						No data
The arubigins						No data						No data
Total flavan-3-ols	5447						411					
<i>Flavonols</i>												
Kaempferol			90		1	15				23	1	18
Myricetin			49		1	15				40	1	18
Quercetin			130		1	15				52	1	18
Total flavonols	269						115					
<i>Flavones</i>												
Apigenin			109		1	19				0	1	18
Luteolin			9		1	19				5	1	18
Total flavones	119						5					
<i>Total Flavonoids</i>	5834						531					

¹Collier and Malloes (1971), ²Kuhr and Engelhardt (1991), ³Ding et al. (1992), ⁴Arts et al. (2000), ⁵Bronner and Beecher (1998), ⁶Lee and Ong (2000), ⁷Arts et al. (1999), ⁸Lin et al. (1998), ⁹Price and Spitzer (1993), ¹⁰Takeo (1974), ¹¹Steinhaus and Englehardt (1989), ¹²Brown et al. (1969), ¹³Owuor et al. (1986), ¹⁴Owuor and Orbanda (1995), ¹⁵Hertog et al. (1993), ¹⁶de Vries (1994), ¹⁷Price et al. (1998), ¹⁸Toyoda et al. (1997), ¹⁹Engelhardt et al. (1993).

^aData from datapoints presented in articles as sums of individual catechin and theaflavins.

Table 3

Differences between green, oolong, black, Pu'er teas illustrating the effect of increasing fermentation on the presence of flavonoid compounds

Flavan-3-ols	(mg/100 g dry tea water extraction only)			
	Green	Oolong	Black	Pu'er ^d
Catechin	24	17 ^{a,*} ,b,***	167 ^{a,***}	0 ^{a,***} ,b,***,c,**
Epicatechin	793	259 ^{a,***}	316 ^{a,***}	81 ^{a,***} ,b,***,c,***
Epicatechin 3-gallate	1755	707 ^{a,**}	923 ^{a,***}	40 ^{a,***} ,b,***,c,***
Epigallocatechin	1712	510 ^{a,***} ,b,**	1,257	157 ^{a,***} ,b,***,c,**
Epigallocatechin 3-gallate	8975	3861 ^{a,*} ,b,***	1,393 ^{a,***}	120 ^{a,***} ,b,***,c,***
Gallocatechin 3-gallate	316	93 ^{a,***}	126 ^{a,**}	13 ^{a,***} ,b,***,c,**
Total catechins by sum of means	13,576	5447	4,183	411
Total catechins	15,667	6037 ^{a,***} ,b,***	3,937 ^{a,***}	493 ^{a,***} ,b,***,c,***
Flavonols				
Kaempferol	130	90	132	23
Myricetin	101	49	25 ^{a,**}	40
Quercetin	175	130	210	52
Flavones				
Apigenin	77	110	54	0
Luteolin	8	9	5	5

^a Significantly different from green tea.

^b Significantly different from black tea.

^c Significantly different from oolong tea.

* $P \leq 0.05$.

** $P \leq 0.01$.

*** $P \leq 0.001$ by Kruskal Wallis ANOVA (chi-square approximation, corrected for ties).

^d Increasing order of enzymatic fermentation from green to oolong to black: Pu'er is an anaerobic bacterial fermentation process.

14 g per 100 g dry green tea. Catechins—which include catechin, epicatechin, epicatechin 3-gallate, epigallocatechin, epigallocatechin 3-gallate, and gallocatechin—were present in all teas, but the amounts present varied. Epigallocatechin 3-gallate, the “signature” compound in green tea, decreased in amount with increasing fermentation from green to oolong to black tea as did total catechins. The other individual catechins were consistently higher in green tea than in oolong or black tea but not consistently higher in oolong than in black tea. It was not possible to determine if total theaflavins and the arubigins are useful as quantitative indicators of fermentation because acceptable data were not available for oolong and green teas.

Pu'er tea was lowest in all the flavan-3-ol compounds studied. However, the anaerobic bacterial fermentation methods producing Pu'er tea may metabolize these compounds or form other flavonoids that were not measured in these analyses.

Table 3 shows that the variability between the different kinds of tea was greater than that within the types of teas, for the tea “signature” flavonoid compound epigallocatechin 3-gallate as well as for total catechins. As degrees of fermentation varied from least (green) to most (black), total catechin and epigallocatechin 3-gallate content decreased.

3.2. *Flavonols and flavones*

The flavonols were the second major class of flavonoids in dry tea by weight (Tables 2 and 3). In contrast to the flavan-3-ols, the flavonols were approximately 0.4 g and the flavones 0.06 g per 100 g dry black tea and 0.4 g flavonols and 0.08 g flavones per 100 g dry green tea. The most common flavonol aglycones were quercetin, kaempferol, and myricetin. However, these rarely occur 'free' (without sugars); 7 quercetin, 6 kaempferol, and 3 myricetin O-glycosides (sugar attached to an oxygen on the aglycone) may be present in tea (Price et al., 1998). Provisional estimates for flavonols and flavones are provided in Table 2.

The differences between the flavonol compounds in green and black teas were not significant, except for myricetin, which was higher in green than in black tea, possibly decreasing with increasing levels of fermentation (Lakenbrink et al., 2000). There are chemical structural similarities between myricetin and epigallocatechin 3-gallate, a compound that also decreases with increasing fermentation.

4. Discussion

The flavan-3-ols were the major flavonoid class in tea. The predominant flavan-3-ol compounds were the the arubigins (derived tannins) in black tea and the catechins in green tea. Many flavan-3-ol analyses were available for black and green teas, the data quality was good, and therefore we consider these provisional estimates of flavan-3-ols in teas to be relatively robust. The flavonol and flavone data quality was very good, but the majority of data were on black tea. Analytic data for oolong and Pu'er teas on catechins were sparse and more data are needed. More analyses are also needed on the theaflavins, the arubigins, flavonols and flavones in green, oolong, and Pu'er teas.

Tea preparation techniques vary substantially from one country to another and by individual habit. Therefore food composition tables that provide data only on "tea as served" are based on a number of assumptions that may be inappropriate for special populations. Tables on the composition of dry teas permit more precise calculations to be made for research purposes.

The flavonoid content of a cup of tea depends on two factors: (1) the composition of the tea itself, and (2) the brewing characteristics. The tea itself is influenced by tea type and processing method (blend, green versus black, etc.). The brewing characteristics are affected by the strength and type of infusion (tea particle size and weight), the flavonoid extraction rate and efficiency (e.g., there are smaller tea particles in teabags than in the loose tea used in tea balls or pots) and brewing conditions (such as tea/water ratio, time and temperature, see Table 4). Of all these influences, the type and weight of tea have the greatest effects on the flavonoid content in a cup of tea (Lakenbrink et al., 2000).

Food composition tables presenting data using a standard percent infusion and brewing time of 4 min are satisfactory for most purposes. The USDA Database for the Flavonoid Content of Selected Foods (US Department of Agriculture, Agricultural Research Service, USDA, 2003) data for brewed tea were compiled using a standard 1% infusion (1 g of tea per 100 mL water). However, for specialized research purposes, calculations that account for possible differences in extraction efficiency may be useful, especially for teabags and short brewing times (2 min or less).

Table 4
Factors influencing flavonoid content in a serving of black tea prepared with a tea bag^a

Factor	Category	Probable effect	Comment
Tea type	Tea (blended or unblended), grade (plant parts—buds, leaf), processing (black, green, etc.)	Amount and kind of flavonoid compounds and capacity for extraction	Most commercial teas are a blend of teas from different tea producers made up of buds, leaves and stems. There are 2 primary methods for processing tea. The orthodox method and the CTC (cut, tear, curl) method, which is more extensively fermented.
Brewing characteristics Tea	Tea weight, particle size, method for infusing tea	Influence strength and type of infusion and ease of extraction	<ol style="list-style-type: none"> 1. Weight affects amount of compounds available and ease of tea–water contact. 2. Particle size affects surface area available for extraction. 3. Tea form (loose tea, tea ball and teabag) affects tea–water contact.
Conditions	Tea/water ratio, time, Temperature	Some but less significant effects.	Amount of the water determines the infusion strength. At brewing times up to 2 min, elution of compounds is independent when tea bags are used. With extended (brewing time > 4 min) the tea–water ratio effect on extraction diminishes. Most tea is made with boiling water.

^aTable based on Lakenbrink et al. (2000). Flavonoids and other polyphenols in consumer brews of tea and other caffeinated beverages. *Journal of Agricultural and Food Chemistry* 48, 2848–2852 and Arts et al. (2000). Catechin contents of foods commonly consumed in the Netherlands. 2. Tea, wine, fruit juices, and chocolate milk. *Journal of Agricultural and Food Chemistry* 48, 1752–1757.

Since most flavonoid compounds are extracted quite rapidly, the amount of water used and brewing times are not important when infusion time is greater than 4 min (Lakenbrink et al., 2000; Arts et al., 2000). However, some populations use shorter brewing times, and the effects on flavonoid content have not yet been quantified definitively. Using the differences between the total flavonoid content (by methanolic extraction) and aqueous infusions of flavonoids in the same

teabags, infusion times of less than 4 min have recently been examined (Lakenbrink et al., 2000) and extraction efficiencies were calculated. The flavonoid content in a serving was found to vary by time, teabag, tea (particle size, bag type, and tea weight) and flavonoid compound. For example, extraction efficiencies vary from 34% to 63% depending on flavonoid class (catechins 0.45, theaflavins 0.34, the arubigins 0.49, flavonols 0.63, and flavones 0.59). To determine flavonoid content in a serving with a short brewing time (less than 4 min) using teabags, extraction efficiencies need to be redetermined using water extraction throughout the analysis.

For individuals who are heavy tea drinkers, tea is probably the major dietary source of catechins, and for many purposes, the data on tea presented here may suffice to obtain relatively complete estimates of these flavonoids in their diets. However, some flavonoids, such as the catechins, are also present in many other foods in the diet, including apples, peaches, pears, and other members of the Rosaceae family. For individuals who consume these other foods in large amounts, or who do not drink tea, considerable amounts of catechins in their diets may be derived both from them and from dietary supplements containing flavonoids. For such persons, complete exposure estimates for flavonoid intake must include foods, supplements, and beverages other than teas. Without obtaining complete data on all foods containing catechins, the health effects due to catechins in the diet might be attenuated and less apparent than with more complete data. The USDA Database for the Flavonoid Content of Selected Food (US Department of Agriculture, Agricultural Research Service, (USDA), 2003) and the earlier isoflavonoid database (US Department of Agriculture, Agricultural Research Service, (USDA), 2002) provide the most extensive data on these compounds to date.

Our data have certain limitations. Additional analyses are constantly being conducted. For example, the US Department of Agriculture's flavonoid database (US Department of Agriculture, Agricultural Research Service, (USDA), 2003) includes new unpublished analytical values for tea, which were not available to be included in calculations of means for this article. Our estimates were generally slightly higher than USDA values for dry tea but the differences were trivial.

Areas for future research include measurement of the flavonoid content of brewed teas of different strengths and infusion times. Also, the associations between water and methanolic flavonoid extracts of dry tea need to be determined to ascertain how best to correlate data with alcoholic extraction to a water basis. Finally, data on the flavonoid content of herbal teas are needed.

5. Conclusions

The flavonoid composition data on black tea are consistent and relatively robust for the major flavan-3-ols. However, the paucity of available data on oolong and Pu'er teas indicates that more studies are needed. Theaflavins need to be measured in green, oolong, and Pu'er teas. The arubigins must be isolated and characterized as well as measured. More data are required on the flavonols and flavones that are also present in all types of teas although in much lower amounts than the flavan-3-ols. The actual intake of tea flavonoids by consumers and the kinetics of brewing tea also merit additional study.

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