Flavonoids and Other Polyphenols in Consumer Brews of Tea and Other Caffeinated Beverages†

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The polyphenolic, flavonoid, and caffeine compositions of four commercial tea bag products (typical of those used in the UK, US, continental Europe, and the Middle East) and beverages prepared from them under a range of typical consumer use conditions have been studied. Leaf composition was determined by extraction with aqueous methanol: the absolute compositions of all four products were remarkably similar in terms of most phenolic compounds. The flavonoids comprised the major proportion (93−94%) of the total phenolics estimated by the Folin−Ciocalteu method. At brew times up to 2 min the composition of the brew solids was for each product practically independent of brew time, with flavonoids again comprising the major proportion (86−88%) of the total phenolics. The efficiency of extraction in brewing of total phenolics, total flavonoids, catechins, and theaflavins was up to 35−55% of the total available in the leaf, whereas the flavonol and flavone glycosides and caffeine were more efficiently extracted (up to 55−90%). The contribution of tea to the UK adult average total dietary intake of flavonols and flavones was calculated to be up to 80% depending on brewing conditions.

Keywords: *Camellia sinensis; tea; coffee; polyphenols; flavonoids; consumer brews*

INTRODUCTION

Tea polyphenols, especially the flavonoids, have attracted considerable interest recently because of their associated health properties, and these have been summarized in a series of reviews (Balentine et al., 1997; Hollman et al., 1997; Wiseman et al., 1997; Blot and McLaughlin, 1997). There is growing interest in dietary antioxidants, among which flavonoids feature in a wide variety of commonplace foods. Green tea leaves contain a high level of catechins (flavanols and flavanol gallates), which are transformed by enzymic oxidation during black tea manufacture into more complex flavonoids (the theaflavins and thearubigins) (Robertson, 1992). It is generally accepted that flavonoids are formed at the confluence of the shikimic and acetate−malonate biosynthetic pathways and have a C6−C3−C6 skeletal structure corresponding to phenyl-substituted benzpyrans and pyrones (Harborne, 1994). In terms of this definition, the catechins, theaflavins, flavonol, and flavone glycosides found in tea are all flavonoids. The situation in the case of the thearubigins is less well-defined. No individual thearubigins have as yet been isolated in pure form with fully elucidated structures. Nevertheless, it has been established that the thearubigins are formed during black tea processing from known flavonoids such as the catechins and theaflavins (and possibly others such as the proanthocyanidins) by enzymic oxidation mediated by leaf polyphenol oxidase and peroxidase. It seems unlikely that the flavonoid carbon skeleton is affected during such enzymic processes, and indeed NMR evidence for retention of the flavonoid C6−C3−C6 structure in isolated thearubigin fractions is now emerging (Davis, 1999). The aim of the current study was to determine what the consumer gets in normal brewed tea in a range of consumer preparation conditions.

MATERIALS AND METHODS

Samples. UK-type tea bags (heat sealed bags, load 3.125 g), US-type tea bags (double chamber bags, load 2.25 g), and two tea bag products commonly used in continental Europe and the Middle East (load 2.0 g) were used. Coffee samples: samples from the UK market, one light roast, one medium roast, and one dark roast sample each. Cocoa powders: two cocoa powders (20% fat content) from different manufacturers.

Leaf composition was determined by analysis of extracts prepared from loose leaf (0.2 g from 10 bags, mixed and ground before extraction) with aqueous (70%) methanol (2 × 5 mL) at 70 °C and 20 min (2 × 10 min) extraction time. In the case of coffee and cocoa similar extractions were carried out to get the overall composition.

Extraction and Isolation Procedures. Consumer beverages were prepared from individual tea bags using initially boiling water under typical consumer conditions at a range of infusion times (25, 40, 60, 90, 120, and 240 s) as follows: UK product, 235 mL of water per 3.125 g bag (water/leaf ratio, 75/1); US product, 180 mL of water per 2.25 g bag (water/leaf ratio, 80/1); international products, 180 mL of water per 2.0 g bag (water/leaf ratio, 90/1).

Six replicate extractions were performed, the extracts combined, and the combined extracts analyzed.

Coffee was extracted using a percolator at concentrations of 20 g/L of water; in the case of the medium roast sample a brew of 40 g/L was also prepared.

Total soluble solids were determined only on aqueous (consumer) extracts. Fifty milliliter tea samples were trans-

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ferred to suitable tared beakers and dried at 103 °C for 6 h and thereafter until a constant weight was reached. The procedure for coffee brews was practically the same.

Total polyphenols were determined using the Folin−Ciocalteu assay (Singleton and Rossi, 1965).

Flavanols. Catechins, gallic acid, theogallin, and alkaloids were determined by RP-HPLC as described by Kuhr and Engelhardt (1991) with slight modifications.

Theaflavins. The total extracts for the theaflavin analysis were prepared as follows: 2.5 g of tea was transferred to the perforation unit of a rotary perforator, and 800 mL of boiling water was added. The perforation unit was placed on a preheated magnetic stirrer and connected to a flask containing 800 mL of boiling ethyl acetate. Extraction time was 3.5 h. After that time, the organic layer was separated, and the ethyl acetate was removed (60 °C, reduced pressure). The residue was dissolved in 10 mL of the mobile phase (Lapczynski and Engelhardt, 1999). The mobile phase was acetic acid (1.5%, aq) and acetonitrile (77/23, v/v).

Tea brews: 800 mL of the hot infusion were transferred to the perforation unit and treated as above. The HPLC separation was conducted on a 5 μm Nucleosil 100 C 18, 250 × 4.6 mm i.d., at 50 °C, 1 mL/min, connected with a diode array detector (set at 380 nm); mobile phase: A = 1.5% acetic acid (aq); B = 100% acetonitrile; 77% A/23% B.

Chlorogenic acids were determined by RP-HPLC after a cleanup using solid-phase extraction (SPE) on RP-18 cartridges. One milliliter of the aqueous tea extracts was applied to the conditioned column, and the polar compound was eluted with 9 mL of water. This procedure was repeated four times (4 mL in total) and the eluate concentrated to a final volume of 2 mL (Engelhardt et al., 1989, Kiehne and Engelhardt, 1996a,b). Five milliliters of the total extracts of the tea samples were concentrated and made up to 10 mL; 4 × 1 mL of this solution were used for SPE.

Flavonol Glycosides. The method of Engelhardt et al. (1992) was followed with slight modifications. Briefly, the brews were applied to a polyamide column and eluted with methanol, concentrated, and made up to volume with DMF/water. In case of the tea and coffee samples, 50 mL of the aqueous extracts was used for the column chromatography. HPLC conditions cf. Engelhardt et al. (1992).

Flavone C-Glycosides. The method of Engelhardt et al. (1993) was followed with the exception of the enzyme used for hydrolysis of the interfering flavonol glycosides (now: pectinol 10 L, an enzyme preparation for the fruit juice industry with glucosidase activity was purchased from Roehm, Darmstadt, FRG).

Calculations. Total flavonoid levels are not directly accessible, since there is no specific method of analysis for thearubigins. Flavonoid levels were therefore calculated from the expression

$$\text{total flavonoids} = \text{total phenolics} - (\text{gallic acid} + \text{theogallin} + \text{chlorogenic acids})$$

This is based on the assumption that the principal NONflavonoid phenolics of tea are gallic acid, theogallin and the chlorogenic acids.

Thearubigin levels were calculated from the expression

$$\text{thearubigins} = \text{total flavonoids} - (\text{catechins} + \text{theaflavins} + \text{flavonols} + \text{flavones})$$

Please note that the data for the flavonols and flavones were those for the aglycons.

RESULTS AND DISCUSSION

Data on levels of total phenolics were obtained by the relatively nonspecific Folin−Ciocalteu method calibrated on gallic acid. Other calibrants (e.g., catechins such as EGCG) may be more relevant to tea, but in comparison with gallic acid they are less freely available in pure form, are of a variable hydration state, and are less stable than gallic acid. The Folin−Ciocalteu response factors for the catechins and theaflavins are under investigation, and it is clear that gallic acid calibration may underestimate the true total phenolics and total flavonoid levels (to be reported).

The gallic acid-derived data reported here nevertheless serve adequately to compare brew compositions at different infusion times, while giving minimum levels of total phenolics, total flavonoids and thearubigins.

Table 1. Composition of Blends (by Analysis of Aqueous MeOH Extracts) (Data Are Reported in % by Weight on Leaf)

	UK	int A	int B	US
caffeine	3.85	3.36	3.77	3.21
theobromine	0.21	0.15	0.24	0.16
total phenolics[a]	16.00	15.49	16.12	16.29
catechins	1.34	0.92	0.68	3.65
theaflavins	1.54	1.15	1.47	1.17
flavonol glycosides	0.86	0.78	0.71	0.88
flavone C glycosides	0.09	0.19	0.10	0.14
thearubigins[b]	11.09	11.56	12.18	9.45
gallic acid + theogallin	0.63	0.57	0.56	0.67
chlorogenic acids	0.45	0.31	0.42	0.33
total flavonoids	14.92	14.61	15.14	15.29
total NONflavonoids	1.08	0.88	0.97	1.00
% flavonoids/ total phenolics	93.30	94.30	94.00	93.90

[a] Estimated by Folin Ciocalteu method calibrated on gallic acid [b] Calculated as total flavonoids − (catechins + theaflavins + flavonols and flavones); see text. Int, international.

Table 2. Composition of Total Phenolics Fraction (Expressed as % of Total Phenolics)

	UK	int A	int B	US
catechins	8.4	5.4	4.2	22.4
theaflavins	9.6	7.4	9.1	7.2
thearubigins	69.3	74.6	75.6	58.0
flavonol/flavone glycosides	5.95	6.3	5.1	6.3
total flavonoids	93.3	94.3	94.0	93.9
gallic acid + theogallin	3.9	3.7	3.4	4.1
chlorogenic acids	2.8	2.0	2.6	2.0
total NONflavonoids	6.7	5.7	6.0	6.1

Product Compositions. The absolute compositions of the tea bag products are remarkably similar in terms of most phenolics (Table 1 - this gives the sums of 14 distinct flavonol glycosides, seven distinct flavone glycosides, the four principal catechins, and the four main theaflavins). The minor theaflavins constitute only up to 10% of the total theaflavins (Lapczynski and Engelhardt, 1999).

On the basis of the caveats mentioned above, the flavonoid fraction dominates compositionally, representing 14.6−15.3 wt % of the leaf and 93.3−94.3% of the total leaf phenolics. The NONflavonoids constitute only up to 1 wt % of the leaf and represent only 5.7−6.7% of its total phenolics (Table 2).

The thearubigins are the most abundant flavonoids, representing 9.5−12.2 wt % of the leaf and 58−75% of the leaf phenolics. The relatively low level of thearubigins (9.5% on leaf, 58% of total phenolics) and high level of catechins (3.65% on leaf, 22.4% of leaf phenolics) in the US product arise from its high level of orthodox Ceylon and Indonesian teas. The UK and international products are largely CTC teas that have undergone more extensive fermentation, resulting in lower levels of catechins (0.7−1.3% on leaf, 4−8% of total phenolics) and higher levels of thearubigins (11.6−12.2% on leaf, 70−75% of total phenolics). CTC (from the words curl, tear, crush) stands for a machine used in black tea

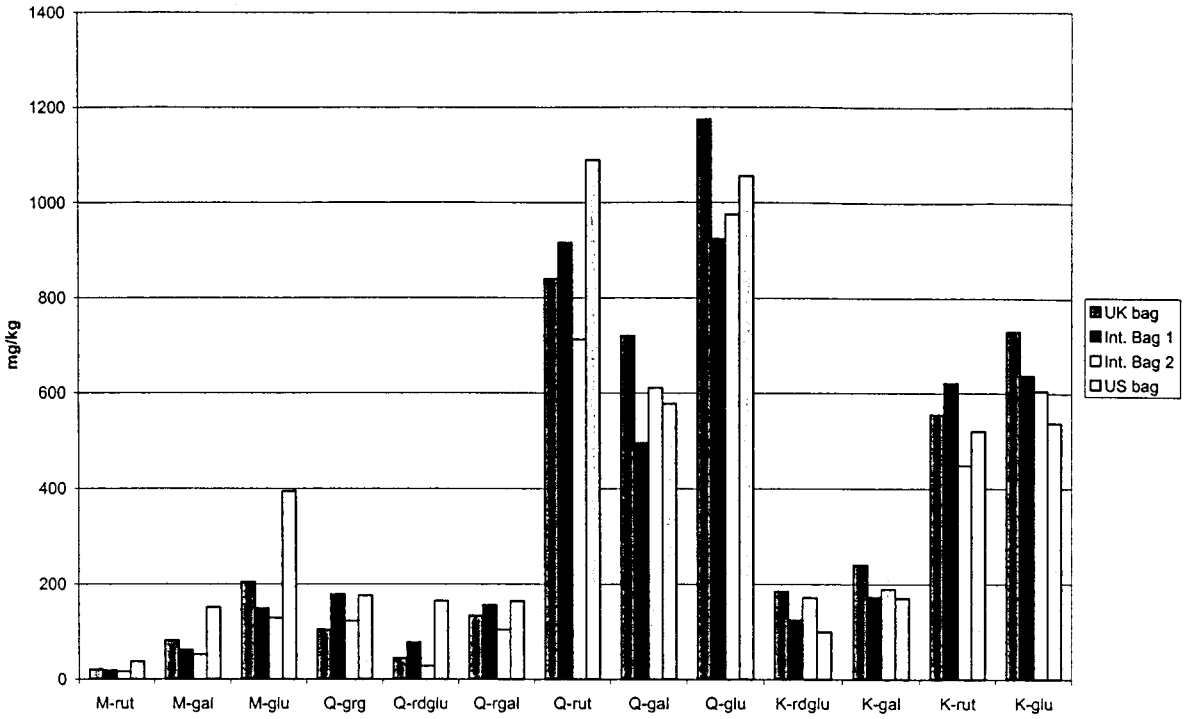

Figure 1. Flavonol glycosides (calculated as aglycons) in the tea bag products: m, myricetin; q, quercetin; k, kaempferol; rut, rutin; glu, glucose; gal, galactose; rdglu, rhamnodiglucoside; grg, glucorhamnogalactoside; rgal, rhamnogalactoside.

Table 3. Compositional Data on Coffee Samples (Methanol Extracts)[a]

	total phenolics (%)[b]	chlorogenic acids (mg/kg)	FOG (agl.)	catechins (sum)	caffeine (%)
instant coffee 1	15.14	23845	n.det	n.det	2.36
instant coffee 2	14.58	22989	n.det	n.det	2.99
GCLR, MeOH	5.41	16915	n.det	n.det	1.37
GCMR, MeOH	5.25	19176	n.det	n.det	1.62
GCDR, MeOH	5.70	15545	n.det	n.det	1.48

[a] Key: GCLR, ground coffee low roast; GCMR, ground coffee medium roast; GCDR, ground coffee dark roast. [b] Calculated as gallic acid equivalents; n.det, not detected; agl., aglycon.

production that consists of two toothed rollers that move in opposite directions with the leaf between. This machine produces smaller pieces than the orthodox rollers and usually the fermentation is more intense (Hampton, 1992).

As mentioned above, we determined not only the sum of the flavonoids in question but also the individual compounds. As can be seen from Figure 1 the flavonol glycosides in the four different tea bags are slightly different. Please note that Q-rut includes probably a small amount of K-glucorhamnogalactoside. The US bag contains some more of the myricetin glycosides which may be due to the different leaf blend composition. This is in tune with the observation that this tea blend consists of orthodox teas as it is known that there is a possible decrease in myricetin with the degree of fermentation (Finger, 1994).

The data on coffee are compiled in Table 3. The total polyphenols are lower compared with tea. Chlorogenic acids are the main polyphenolic constituents. As expected, none of the individual flavonoids analyzed could be detected. In the case of cocoa only exhaustive extracts were analyzed as it is consumed without aqueous extraction as a beverage or in the form of chocolate (Richelle et al., 1999). The total polyphenols of the cocoa samples were 5.96 and 5.98%. The samples also had ca. 1.9% of theobromine, catechins (0.15 and 0.14%), and small amounts of chlorogenic acids and flavonol glycosides (around 100 mg/kg).

Composition of Brew Solids. The flavonoids dominate the composition of the brew solids (ca 25%) and constitute a practically constant 86% of the total phenolics (Table 4). This is lower than the corresponding figure for leaf composition (94%), indicating that some flavonoids in the leaf are relatively poorly extracted by hot water—possibly highly polymerized thearubigins bound to denatured leaf protein—and consequently not amenable to aqueous extraction at brew times up to 2 min. (In this connection, note that at an extended brew time of 4 min, extraction efficiency of thearubigins is similar for all four products (ca 50%)). The theaflavins—efficient protein denaturants—are even less efficiently extracted (27−37%). These results stand in need of further test.

The contribution of the flavonoids to the total phenolics (assayed by the Folin−Ciocalteu method) in leaf (flavonoids 94% of total phenolics) and in brews (flavonoids: 86% of total phenolics, independent of brew time) is very similar. This might suggest that total flavonoid levels in tea may be derived from Folin−Ciocalteu figures for total phenolics. The viability of the phenolics conversion factor approach to estimation of total flavonoids is to be further investigated (see above).

Recently, a study was published on the flavonol glycosides content of tea infusions, which varied from 36.5 to 88.3 mg/L (Price et al., 1998). These results are very similar to ours, which varied from 24.7 to 37.5 mg/L. The difference might be due at least in part to the shorter brewing time we used in our study.

For comparison purposes, some measurements have been carried out with coffee brews (Table 5). As expected, chlorogenic acids were the main group of polyphenols in coffee brew (up to 0.9 g/L in the 40 g/L brews).

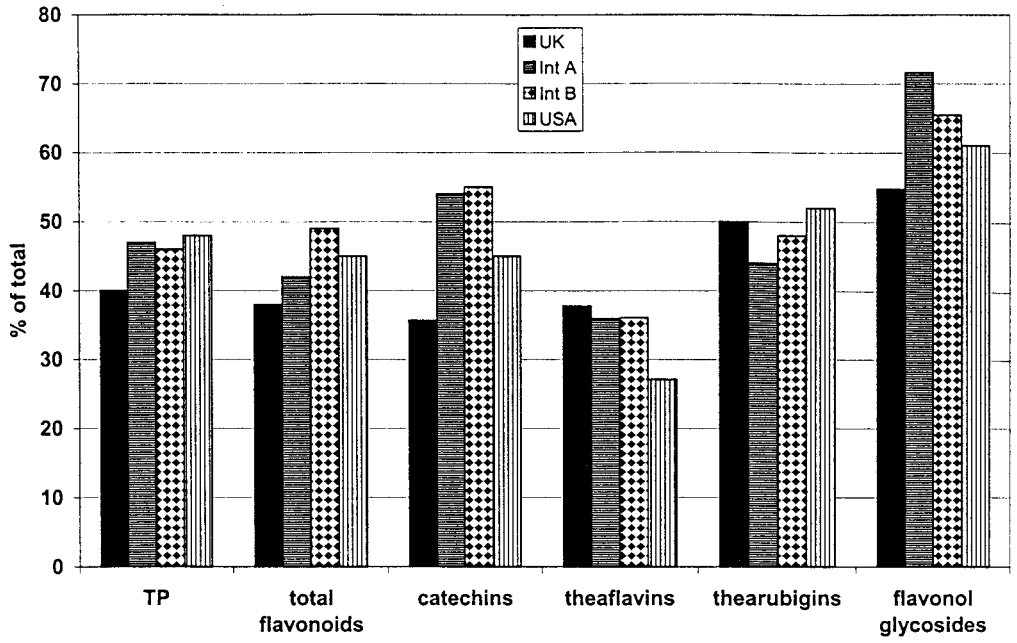

Figure 2. Extraction efficiency of selected polyphenols in 2 min brews. TP, total phenolics.

Table 4. Composition of Brew Solids (Results Given in % on Total Brew Solids)[a]

	TP	total flavonoids	TR	TF	catechins	FOG	FCG	caffeine
UK	26−30	22−24	17−18	2.2−2.8	1.3−2.6	2.0−2.2	0.21−0.22	9.2−9.8
int A	25−28	22−23	17−18	1.4−1.7	1.2−1.9	1.9−2.1	0.45−0.46	9.4−11.2
int B	27−30	23−24	18−19	1.9−2.0	0.9−1.4	1.6−1.8	0.19−0.43	9.8−10.1
US	27−31	23−27	15−17	1.2−1.3	5.3−6.5	2.0−2.1	0.30−0.31	8.6−8.7

[a] Abbreviations: TP, total phenolics; TR, thearubigins; TF, theaflavins; FOG, flavonol glycosides; FCG, flavone C glycosides.

Table 5. Composition of Coffee Brews[a]

	soluble solids (g/L)	total polyphenols (g/L)	chlorogenic acids (mg/L)	caffeine (mg/L)
GCLR (20 g/L)	5.64	0.96	375	310
GCMR (20 g/L)	5.74	1.06	403	410
GCMR (40 g/L)	12.11	2.27	858	760
GCDR (20 g/L)	6.08	1.06	342	340

[a] For abbreviations, see Table 3.

Again, in coffee brews none of the individual catechins or flavonol and flavone glycosides could be detected. This it not too surprising as coffee is a seed where the presence of these compounds is not expected.

Efficiencies of Extraction. At brew times up to 2 min, extraction of total phenolics, total flavonoids, catechins, and theaflavins is relatively inefficient (up to 35−55%), whereas the glycosylated flavonols and flavones and caffeine (data not shown) are more efficiently extracted (up to 55−90%). The order of extraction efficiencies for the international and US products is catechins > thearubigins > theaflavins. Figure 2 shows an example of selected compounds.

The data for catechin extraction efficiency for the UK product seems anomalously low: this will be followed up in further product monitoring work. The lower bag charges (2−2.25 g) and higher water/leaf extraction ratios (90/1−80/1) associated with the international and American products result in higher extraction efficiencies than those found for the UK product, with its higher bag charge (3.125 g) and lower water/leaf extraction ratio (75/1). However at an extended brew time of 4 min these differences diminish: all four products behave similarly, with 52−59% of phenolics and 44−52% thearubigins extracted from the leaf (data not shown).

Table 6. Selected Polyphenols in Consumer Brews[a]

	brew time	
	40−60 s	120 s
total phenolics		
UK	139−163	198
int A	92−117	145
int B	100	150
US	96	174
total flavonoids		
UK	137−141	172
int A	99−103	124
int B	86	129
US	82	152
catechins		
UK	9.2−11.3	13.2
int A	5.9−6.3	9.9
int B	3.4	7.4
US	18.5	37.1
theaflavins		
UK	13−16.6	18.2
int A	6.2−6.7	8.2
int B	7.3	10.6
US	4.4	7.1
flavonols (flavonol glycosides, calcd as aglycons)		
UK	11.8−12.7	14.8
int A	8.5−8.8	11.2
int B	5.9	9.4
US	6.8	12.1

[a] Data are given in mg/serving (235 mL in case of the UK product, 180 mL for the US and international products).

Dietary Intakes of Flavonoids. The time course and the delivery/serving of relevant groups of polyphenols is shown in Table 6. At a typical UK consumer brew time of 40−60 s, for cup/mug brewing, the UK product delivers 137−141 mg total flavonoids (including the thearubigins) per average serving (235 mL mug),

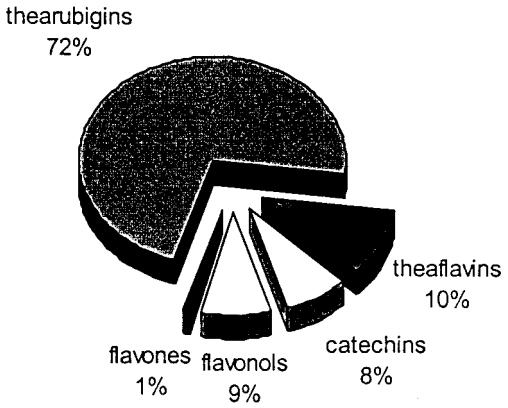

Figure 3. Flavonoids per serving (235 mL) from UK type tea bag (2 min brew time). Data are given as percent of the total flavonoids.

of which the structurally identified flavonoids (catechins, theaflavins, and flavonol/flavone glycosides) together constitute 34.0−40.6 mg. The thearubigins make the dominant contribution to flavonoid delivery (100−102 mg). Figure 3 gives an example of the contributions made by the individual groups of flavonoids. At an average UK adult daily tea consumption of 600 mL (MAFF, 1987), the UK product contributes (as aglycons) 19.7−25.6 mg flavonols and flavones to the average UK total daily intake of 29.8 mg (Sheperd and Ibe, 1995). Tea is the dominant dietary source of these flavonoids (66−85% depending on brew time): vegetables contribute 12%, fruit products 6%, and other beverages (including fruit juices) up to 16%.

Diet studies in The Netherlands indicate that there, too, tea makes a dominant contribution to total dietary intake of flavonol and flavone glycosides (albeit lower, 48%, due to the lower per caput consumption of tea in The Netherlands) (Hertog et al., 1993a,b). In Finland, the intake of flavonols from tea was estimated to be 21% of the total (Häkkinen et al., 1999).

The flavonols constitute 85% of the dietary intake from the phenolic glycosides of tea (flavones 15%), with quercetin the main contributor (59%), followed by kaempferol (21%) and myricetin (5%).

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LITERATURE CITED

Balentine, D. A.; Wiseman, S. A.; Bouwens, L. C. M. The Chemistry of Tea Flavonoids. *Crit. Rev. Food Sci. Nutr.* **1997**, *37*(8), 693−704.

Blot, W. J.; McLaughlin, J. K. Cancer rates among drinkers of black tea. *Crit. Rev. Food Sci. Nutr.* **1997**, *37*(8), 739−760.

Davis, A. Personal communication (to be published), 1999.

Engelhardt, U. H.; Finger, A.; Herzig, B. Untersuchungen über Polyphenole im Tee mittels HPLC: Hydroxyzimtsäurederivate. *Lebensmittelchem. Gerichtl. Chem.* **1989**, *43*, 58−59.

Engelhardt, U. H.; Finger, A. L.; Kuhr, S. Determination of flavone-C-glycosides in tea. *Z. Lebensm. Unters. Forsch.* **1993**, *197*, 239−244.

Engelhardt, U. H.; Finger, A.; Herzig, B.; Kuhr, S. Determination of flavonol glycosides in black tea. *Deutsche Lebensmittel-Rundschau* **1992**, *88*, 69−73.

Finger, A. In-Vitro Studies on the effect of polyphenol oxidase and peroxidase on the formation of polyphenolic black tea constituents. *J. Sci. Food Agric.* **1994**, *66*, 293−305.

Häkkinen, S. H.; Kärenlampi, S. O.; Heinonen, I. M.; Mykkänen, H. M.; Törrönen, A. R. Content of the flavanols quercetin, myricetin, and kaempferol in 25 edible berries. *J. Agric. Food Chem.* **1999**, *47*, 2274−2279.

Hampton, M. G. Production of black tea. In Tea − cultivation to consumption; Willson, K. C., Clifford, M. N., Eds.; Chapman & Hall: London, 1992; pp 459−512.

Harborne, J. *The flavonoids−Advances in research since 1986*; Chapman and Hall: London 1994.

Hertog, M. G. L.; Hollman, P. C. H.; van de Putte, B. Content of potentially anticarcinogenic flavonoids in tea infusions, wines, and fruit juices. *J. Agric. Food Chem.* **1993a**, *41*, 1242−1246.

Hertog, M. G. L.; Hollman, P. C. H.; Katan, M. B.; Kromhout, D. Intake of potentially anticarcinogenic flavonoids and their determinants in adults in The Netherlands. *Nutr. Cancer* **1993b**, *20*, 21−29.

Hollman, P. C. H.; Tijburg, L. B. M.; Yang, C. S. Bioavailability of flavonoids from tea. *Crit. Rev. Food Sci. Nutr.* **1997**, *37*(8), 719−738.

Kiehne, A.; Engelhardt, U. H. TSP-LC-MS analysis of various groups of polyphenols in tea. Part 1: Catechins, flavonol O-glycosides and flavone C-glycosides. *Z. Lebensm. Unters. Forsch.* **1996a**, *202*, 48−54.

Kiehne, A.; Engelhardt, U. H. TSP-LC-MS analysis of various groups of polyphenols in tea. Part 2: Chlorogenic acids, theaflavins and thearubigins. *Z. Lebensm. Unters. Forsch.* **1996b**, *202*, 299−302.

Kuhr, S.; Engelhardt, U. H. Determination of flavanols, theogallin, gallic acid and caffeine in tea using HPLC. *Z. Lebensm. Unters. Forsch.* **1991**, *192*, 526−529.

Lapczynski, S.; Engelhardt, U. H. Unpublished results, 1999.

MAFF (Ministry of Agriculture Fisheries and Food). UK Total Diet and Nutrition Report, 1987.

Price, K. R.; Rhodes, M. J. C.; Barnes, K. A. Flavonol glycoside content and composition of tea infusions made from commercially available teas and tea products. *J. Agric. Food Chem.* **1998**, *46*, 2517−2522.

Richelle, M.; Tavazzi, I.; Enslen, M.; Offord, E. A. Plasma kinetics in man of epicatechin from black chocolate. *Eur. J. Clin. Nutr.* **1999**, *53*(1), 22−6.

Robertson, A. The chemistry and biochemistry of black tea production − the nonvolatiles. In *Tea − cultivation to consumption*; Willson, K. C., Clifford, M. N., Eds.; Chapman & Hall: London, 1992; pp 555−601.

Shepherd, M. J.; Ibe, F. Flavonoids in UK total Diet samples. Ministry of Agriculture Fisheries and Food, CSL report No. FD95/2, 1995.

Singleton, V. L.; Rossi, J. A. J. Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. *Am. J. Enol. Vitic.* **1965**, *16*, 144−158.

Wiseman, S.; Balentine, D. A.; Frei, B. Antioxidants in tea. *Crit. Rev. Food Sci. Nutr.* **1997**, *37*(8), 705−718.

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