Factors Affecting the Caffeine and Polyphenol Contents of Black and Green Tea Infusions

Conrad Astill,* Mark R. Birch, Clive Dacombe, Philip G. Humphrey, and Philip T. Martin

Unilever Research Colworth, Colworth House, Sharnbrook, Bedford MK44 1LQ, United Kingdom

The effects of product and preparation variables on the in-cup chemical composition of tea extracts is of interest because the appearance and taste characteristics and the possible health effects of a tea liquor arise from the chemical components extracted from the leaf during tea preparation. A comprehensive study was therefore undertaken to determine the contributions of product and preparation variables on the total soluble solids, caffeine, and polyphenol contents of tea extracts. The results of this study show that the variety, growing environment, manufacturing conditions, and grade (particle size) of the tea leaves each influence the tea leaf and final infusion compositions. In addition, the composition of the tea infusion was shown to be influenced by whether the tea was contained in a teabag and, if so, the size and material of construction of the bag. Finally, the preparation method, including the amounts of tea and water used, infusion time, and amount of agitation, was shown to be a major determinant of the component concentrations of tea beverages as consumed. An illustration of the variation introduced by these product and preparation factors is provided by comparing solids, caffeine, and polyphenol contents of green and black tea infusions when commercial products are prepared according to the instructions given on their packaging.

Keywords: *Camellia sinensis; tea; polyphenols; catechins; caffeine; infusions; Folin−Ciocalteu*

INTRODUCTION

Tea is one of the most widely consumed drinks in the world. In the Far East (particularly in China and Japan), tea is consumed mainly as a hot infusion of "unfermented" fresh green shoots (green tea), whereas in most other countries the beverage is prepared from predominantly "fermented" (black) tea. The different methods of manufacture of these products account for the marked difference in the chemical compositions of green and black teas, and even among green and black tea products the effects of plant variety, growth conditions, and processing method would be expected to produce quite wide variations in the chemical compositions of the resulting products.

In addition, worldwide consumer observations and questionnaire studies on tea preparation habits have shown wide variations among countries, and among individuals within countries, in the way they make their tea (e.g., the weight of tea taken, the amount of water added to the leaves, the amount of agitation used to assist infusion, the length of time the leaf is left in contact with the water, and the use of additional ingredients). In general, Western countries drink black tea, made by infusing a quantity of leaf (usually contained in a teabag) in boiling water in a pot or increasingly in a cup/mug. The infusion time is generally short (<3 min), and the beverage is usually consumed hot (either with or without milk and/or sugar). In India, Pakistan, and some Middle Eastern countries an alternative black tea preparation method is widely adopted. Here the drink is largely prepared by boiling the black leaves in a pan for several minutes prior to consumption (often together with water, milk, and sugar). In the Far East (e.g., China and Japan) the drink is normally prepared from green tea by infusing it in hot (but not boiling) water. Generally the first infusion is discarded and it is the second and subsequent infusions that are consumed. More recently, in the West, green teas (often contained in teabags) have gained in popularity.

The effects of these wide product and preparation differences on the in-cup chemical composition of tea infusions is of interest because the quality and health properties of the consumed drink are associated with the chemical components (in particular the polyphenols and caffeine) extracted from the leaf. The health effects of tea consumption have, for example, been the subject of a number of epidemiological and intervention studies, and evidence is emerging of a relationship between tea consumption and a reduced risk of cardiovascular disease and cancer (*1*−*4*). The primary components under study in this respect are the flavonoid polyphenols, which have been demonstrated to have strong antioxidant effects in vitro (*5*, *6*).

Caffeine, because it is widely consumed in a variety of products including tea, continues to be the subject of an intense level of scrutiny. Generally speaking, the extensive studies of caffeine have shown that it is not a harmful compound (*7*), although some epidemiological studies have suggested a link between a high caffeine intake and the risk of spontaneous abortion (*8*, *9*).

In epidemiological and some intervention studies the amount of tea estimated from the number of cups consumed is frequently used as an indicator of caffeine or polyphenol consumption. However, this may provide a misleading measure of intake because of the large variations introduced by product and tea preparation differences. Some of the factors affecting the rate of infusion of tea solubles into aqueous solution have been

* Author to whom correspondence should be addressed [telephone 44 (0)1234 222448; fax 44 (0)1234 222844; e-mail conrad.astill@unilever.com].

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widely studied. One of the earlier experimental studies (*10*) examined the influence of a range of variables, including the nature of the raw materials, the purity and temperature of the water, the infusion time, and the water-to-leaf ratio, on the brewing behavior of tea. This showed that the rate of extraction of selected soluble constituents increased with decreased particle size, increased brewing temperature, and increased water/leaf ratio. Little research then followed until the extensive studies conducted by Spiro and co-workers. Spiro introduced simple kinetic models (*11*, *12*) to explain the observed rates of extraction of individual constituents into aqueous solution. The experimental studies measured the amount of extracted tea constituents as a function of time, when tea is infused in water at a constant elevated temperature. The results yield so-called *infusion curves* in which the liquor concentrations of polyphenols and caffeine (or any other tea soluble) increase with brew time. The rate of increase of concentration with time gradually falls off until an equilibrium level is reached. At this stage the soluble material is partitioned between the tea brew and the hydrated tea leaves.

The rate-determining step in the loose-leaf infusion was determined as the diffusion of the solute through the leaf matrix to the surface. The influence of leaf size, origin, and manufacture on the rate of infusion of caffeine and theaflavin has been reported (*13*). The rate constants increased as leaf size decreased and varied between the different leaf types (i.e., Kenyan CTC infused more rapidly than Indian orthodox teas). Further studies (*14*, *15*) compared the rates of caffeine infusion from green and black teas. These showed that the rate of infusion of caffeine was greater from green tea than from a similarly sized black tea, confirming a possible effect of manufacturing method upon infusion characteristics. A study of the effects of black and green teas on in vitro lipoprotein oxidation (*16*) also looked at the effect of extraction time on the composition of loose-leaf brews prepared at "drinking strength". Differences were observed between the extraction kinetics of the polyphenolics (50−60% extraction after 4 min) and the caffeine (80% extraction after 30 s). No significant difference in antioxidant activity was detected between green and black teas.

Most of the research to date has been concerned with the physicochemical aspects of the dissolution process from *loose-leaf tea* over comparatively *long time periods*. Of increasing interest, however, are the factors that affect the amounts of physiologically active components in a "consumer brew" obtained when a *teabag tea* is infused for a relatively *short time period*. Two recent studies have investigated the composition of consumer strength teabag brews. The polyphenol and caffeine contents of four commercial black tea blends were determined and the compositions of their infusions measured for brew times of up to 2 min (*17*). This confirmed the previous findings on loose-leaf infusions (*16*) and showed caffeine to be more efficiently extracted (up to 90% of the total available in the leaf) than the polyphenols (up to 55% of the total available in the leaf). A second recent study (*18*) looked at the effects of a few preparation variables (brew time, temperature, teabags, and milk addition) on the antioxidant content of green and black tea infusions. Green teas brewed for long time periods and at high temperature gave the highest antioxidant content. The effects of two important domestic preparation variables (leaf/water ratio and agitation) were, however, omitted from the latter study, which was conducted on only a limited number of tea products.

The aim of this study is to evaluate a wider range of product and preparation variables and to determine the contributions they make to the composition and strength of an in-home infusion.

MATERIALS AND METHODS

Preparation of Extracts. Aqueous extracts were prepared by infusing the loose tea (or teabags) in a measured volume of boiling deionized water for a fixed time period (either with or without mechanical agitation). The teabag was then removed (or in the case of loose-leaf infusion the liquor filtered under vacuum through a No.1 sintered glass crucible) and the solution allowed to cool to room temperature. This was repeated three times, and the extracts were combined to form the analytical sample.

The leaf composition was determined by the analysis of "total" extracts. These were prepared by extracting milled leaf samples (0.20 g) with aqueous methanol (70:30 methanol/water). The leaf was extracted twice with aqueous methanol (5 mL at 70 °C for 10 min) and centrifuged, and the combined supernatant liquors were readjusted to volume (10 mL) with 70% aqueous methanol to form the analytical sample. This procedure was used because it gave total polyphenol values comparable to those obtained by aqueous extraction (100:1 water to ground leaf refluxed for 60 min) without degrading the flavonols (*19*).

Determination of Soluble Solids. Soluble solids were determined by transferring a measured volume of aqueous extract (50 mL) into a tared beaker and evaporating to dryness. The residue was finally dried, in an oven at 103 °C, to constant weight.

Determination of Total Polyphenols. Total polyphenols were determined on the extract samples according to the Folin−Ciocalteu method (*20*, *21*), using gallic acid as the calibrant.

Determination of Caffeine and Flavanols (Catechins). The caffeine and flavanol contents of the extracts were determined by high-performance liquid chromatography (HPLC) as described by Kuhr and Engelhardt (*22*).

RESULTS AND DISCUSSION

Effect of the Nature of the Product. *Tea Origin.* Tea is made from the fresh green shoots of the plant *Camellia sinensis*. Two variants, *C. sinensis* var. *sinensis* (China) and *C. sinensis* var. *assamica* (Assam), exist, which differ significantly in caffeine and polyphenol contents. It has been shown (*23*) that fresh green leaves from Assam teas (*C. sinensis* var. *assamica*) are generally higher in caffeine (mean = 4.09%) and polyphenols (mean = 19.42%) than Chinese cultivars (*C. sinensis* var. *sinensis*) (caffeine mean = 3.11%; polyphenol mean = 16.24%).

The two major commercial tea products, black and green teas, derive from the *C. sinensis* plant but are produced by different processes. To explore the differences between these two tea products, a survey was conducted on a range of black and green tea originals. Caffeine, total polyphenols, and flavanols (catechins) were determined on 70% methanol extracts of black and green teas. The different processing routes can be seen to result in a significant compositional difference between processed green and black products (Table 1). These compositional differences are due in part to the different leaf varieties used as raw material, but more important determinants of final composition are the