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Effects of cosolvents on the decaffeination of green tea by supercritical carbon dioxide

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Abstract

Due to the adverse effects of the caffeine in a variety of plant products, many methods have been explored for decaffeination, in efforts to remove or reduce the caffeine contained in plant materials. In this study, in order to remove caffeine from green tea (*Camellia sinensis*) leaves, we have employed supercritical carbon dioxide (SC–CO_2), which is known to be an ideal solvent, coupled with a cosolvent, such as ethanol or water. By varying the extraction conditions, changes not only in the amount of caffeine, but also in the quantities of the principal bioactive components of green tea, including catechins, such as epigallocatechin gallate (EGCG), epigallocatechin (EGC), epicatechin gallate (ECG) and epicatechin (EC), were determined. The extraction conditions, including temperature, pressure and the cosolvent used, were determined to affect the efficacy of caffeine and catechin extraction. In particular, the type and concentration of a cosolvent used constituted critical factors for the caffeine removal, combined with minimal loss of catechins, especially EGCG. When the dry green tea leaves were extracted with SC–CO_2 modified with 95% (v/v) ethanol at 7.0 g per 100 g of CO_2 at 300 bar and 70 °C for 120 min, the caffeine content in the decaffeinated green tea leaves was reduced to 2.6% of the initial content. However, after the SC–CO_2 extraction, a substantial loss of EGCG, as much as 37.8% of original content, proved unavoidable.

Keywords: Supercritical carbon dioxide; Green tea; Caffeine; Catechins; Decaffeination

1. Introduction

Green tea (*Camellia sinensis*) leaves have a long tradition of being used as a drink in Asian countries, and green tea has become one of the most popular drinks in the world, owing both to its unique taste and to a variety of positive health benefits associated with the plant. Green tea is prepared via processing of the tea leaves, such as by steaming or roasting, without fermentation. When the leaves are semi-fermented or fully fermented, the products can be differentiated from green tea, and are referred to as oolong tea and black tea, respectively. The components of green tea include the catechins, caffeine, and essential oil components, and the composition of green tea varies significantly with the strain of tea tree, the harvest time, and the manufacturing process employed (Chu & Juneja, 1997).

The primary bioactive components of green tea are catechins, which are composed primarily of epigallocatechin gallate (EGCG), epigallocatechin (EGC), epicatechin gallate (ECG) and epicatechin (EC). These catechins are generally recognized to exert desirable biological and physiological effects, including antioxidative (Cai et al., 2002; Joshi, Hasan, Chandra, Husain, & Srivastava, 2004; Liu, Ma, Zhou, Yang, & Liu, 2000), anticancer (Moyers & Kumar, 2004), anti-inflammation (Tedeschi, Suzuki, & Menegazzi, 2002; Trompezinski, Denis, Schmitt,

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& Viac, 2003), anti-aging (Cooper, Morre, & Morre, 2005), antibiotic, and antiviral effects (Fassina et al., 2002; Stapleton et al., 2004; Yanagawa, Yamamoto, Hara, & Shimamura, 2003). In particular, the most profound antioxidative activity among catechins is attributed to EGCG, generally the most abundant catechin in green tea. As a result of such health-positive effects from the catechins in green tea, the consumption of green tea is rapidly increasing, as is the consumption of a variety of green tea byproducts, including drinks, ice cream, beauty foods and cosmetics.

Meanwhile, another important component in green tea is caffeine, which is known to exert relatively adverse effects in humans, including sleep deprivation (Hindmarch et al., 2000), abortions and miscarriages (Giannelli, Doyle, Roman, Pelerin, & Hermon, 2003; Rasch, 2003), and hypersensitivity (Bernhisel-Broadbent, 1999). Therefore, the intake of caffeine by vulnerable consumers, including pregnant women, infants, and children, has become a major cause of concern. According to the information available by the American Beverage Association (2006) and the International Food Information Council (2003), a daily intake of less than around 300 mg of caffeine a day is considered as the safe level. Since one tea bag of green tea (2 g) normally contains 70–100 mg of caffeine (Chu et al., 1997; Perva-Uzunalic et al., 2006; Wright, Mphangwe, Nyirenda, & Apostolides, 2000), less than four cups of green tea daily are determined to be a safe consumption level. Due to the increasing concern regarding caffeine, for example, the European Union has recently decided to mandate warning labels for drinks with a caffeine content above 150 mg/l (Food production daily Co, 2006; University of Reading, 2002).

Considering the negative effects of caffeine, many efforts have been made to remove caffeine from several caffeine-containing foods, including coffee (Brunner, 1988; Peker, Srinvasan, Smith, & McCoy, 1992), guarana (Mehr, Biswal, Collins, & Cochran, 1996), and black tea (Vitzthum & Hubert, 1979). Current research into decaffeination follows a growing dissatisfaction with traditional decaffeination techniques, which utilize organic solvents such as trichloroethylene and dichloromethane. These solvents have been banned in techniques for the decaffeination of foods, due to the likelihood that these solvents cause cancer in humans, as warned by National Cancer Institute in the US. Currently, ethyl acetate and supercritical carbon dioxide have been commercially employed as alternatives for the removal of caffeine from coffee and black tea (Coffee Research Institute, 2007; Johnston, 2007).

To avoid a large loss of other beneficial components during the decaffeination process (Farah, De Paulis, Moreira, Trugo, & Martin, 2006), there have been several attempts at developing more selective caffeine removal methods, by using hot water (Liang et al., 2007), microbial degradation of caffeine (Gokulakrishnan, Chandraraj, & Gummadi, 2005), or even by developing genetically-modified (GM) coffee plants without synthesizing of caffeine (Ashihara & Crozier, 2001; Ogita, Uefuji, Yamaguchi, Koizumi, & Sano, 2003). However, those methods have some limitations since the hot water treatment is applicable to green tea leaves only in fresh form, and the microbial degradation of caffeine and the caffeine-free GM plants are too early to apply to food products.

Meanwhile, pressurized carbon dioxide in its supercritical state, in which the carbon dioxide is pushed beyond its critical point (73.8 bar and 31.1 °C), has been recognized as an ideal non-polar extraction solvent, because supercritical carbon dioxide (SC–CO_2) offers several positive features, including high dissolving power, high diffusivity, and low viscosity. Also, more importantly, SC–CO_2 can be readily recovered after extraction, and is considered to be very safe for humans.

Despite the advantages of SC–CO_2, only a few attempts have been made to apply SC–CO_2 to the processing of green tea or green tea components. As the equilibrium solubilities of catechin and epicatechin SC–CO_2 have been assessed (Berna, Chafer, Monton, & Subirats, 2001; Chafer, Berna, Monton, & Munoz, 2002), it has also been determined that the solubility of catechins in SC–CO_2 is too low to extract catechins from green tea, as a consequence of the low polarity of SC–CO_2. In an attempt to acquire low caffeine-containing polyphenol oleoresin oil from green tea and oolong tea (Chang, Chiu, Chen, & Yang, 2001), SC–CO_2 proved not to be feasible since the removal of caffeine was less effective when using SC–CO_2 than when using the Soxhlet method.

To the best of our knowledge, there has been no report, thus far, on the production of decaffeinated green tea using SC–CO_2. In this study, we focussed primarily on the development of a supercritical CO_2 extraction process, using a polar cosolvent, which resulted in the effective acquisition of decaffeinated green tea leaves with low caffeine contents.

2. Materials and methods

2.1. Materials

Dry-processed green tea leaves (*C. sinensis*) were provided by the Boseong Tea Experimental Station (Boseong, Jeonnam, Korea) in July 2004, and were stored at −70 °C until used. All solvents used in this work were of HPLC grade, and were obtained from TEDIA (Fairfield, USA). Authentic standards, including caffeine (100%), epicatechin (EC, 97%), epigallocatechin (EGC, 98.3%), epigallocatechin gallate (EGCG, 94.4%) and epicatechin gallate (ECG, 99.5%), were all obtained from the Sigma Chemical Company (St. Louis, USA). High-purity CO_2 (99.5%, Daehan Specialty Gases, Seoul, Korea) was employed throughout the supercritical fluid extraction in this study.

2.2. Supercritical CO_2 extraction

Supercritical fluid extraction (SFE) was conducted using the laboratory-scale SFE system (Ilshin Autoclave Co.,

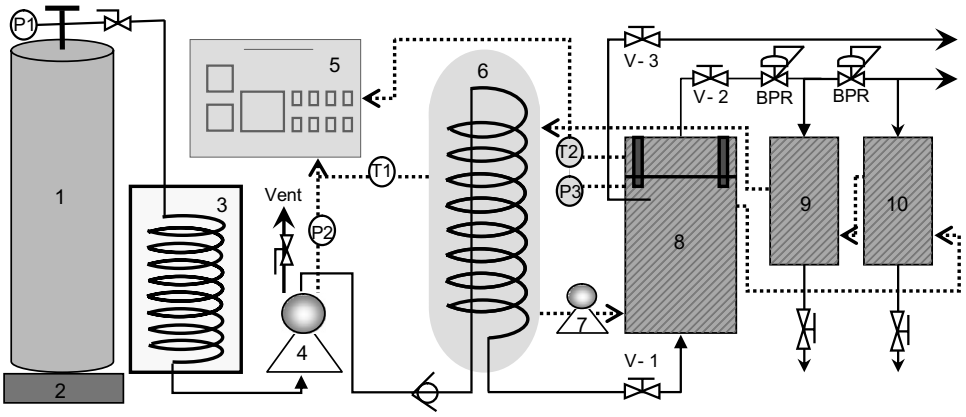

Fig. 1. Schematic diagram of supercritical CO_2 extraction system: 1, CO_2 cylinder; 2, electronic balance; 3, chiller; 4, CO_2 pump; 5, controller; 6, heating bath; 7, circulation pump; 8, extraction vessel; 9, separator 1; 10, separator 2; V-1, valve 1; V-2, valve 2; BPR, back pressure regulator; dotted lines, water; solid lines, CO_2.

Daejeon, Korea) shown in Fig. 1. Green tea was ground via the use of a cutting mill (IKA, Staufen, Germany) and sieved, by using sieves (Chung Gye Sang Gong Sa, Seoul, Korea), in order to obtain particle sizes from 425 to 710 μm. When the temperature of the SFE system reached the designated temperature, 10 g of ground sample were soaked in a cosolvent and loaded into the extraction vessel with an internal volume of 100 ml. After the vessel had been tightly closed, the connecting valves (V-1 and V-2 in Fig. 1) were opened carefully, and the decompression valve (V-3) was closed. Then, the liquid CO_2 contained in the siphoned cylinder was pumped through the water bath, the temperature of which was kept constant, into the SC–CO_2 vessel. The vessel was generally fully pressurized to a designated pressure within 2 min. Then, the valves connecting to the treatment vessel (V-1 and V-2) were maintained in the "open" position throughout the entirety of the extraction time.

The extraction pressure was monitored continuously and controlled using a back pressure regulator (BPR) connected to the extraction vessel. To determine the mass flow rate of CO_2 during extraction, the weight of the CO_2 cylinder was monitored by using an electronic balance (CAS, Seoul, Korea) with an accuracy of 0.01 g and was recorded every 2 min after the extraction system reached the set pressure. The mass flow rate of CO_2 generally remained constant within ±5% deviation from the average flow rate of 8.5 g/min. The total mass of CO_2 used for the entire extraction was determined by integrating the mass flow rate. After each 40 min extraction batch was completed, the extraction vessel was depressurized by slowly opening the decompression valve (V-3), after which depressurization took approximately one min. Since the addition of a cosolvent took place off line, to replenish the cosolvent after 40-min extraction, the extracted green tea leaf sample was then removed and soaked with the same amount of cosolvent as used initially. Then, the green tea leaves were reloaded into the extractor for further SC–CO_2 extraction. After the end of the extraction cycle, the extracted sample was subjected to composition analysis by HPLC.

2.3. Composition analysis of green tea

To determine the contents of caffeine and catechins in the green tea, before and after SC–CO_2 extraction, the green tea samples were extracted with an aqueous ethanol solution, and the obtained extract was then analyzed by high-performance liquid chromatography (HPLC). The green tea sample was dried overnight in a convective drying oven at a temperature of 105 °C, in order to remove any moisture and residual cosolvent. The dried green tea sample was ground with a cutting mill (IKA, Staufen, Germany), sufficiently finely to pass through the sieve, with an opening of 425 μm, and 200 mg of the samples were extracted in triplicate by using 20 ml of 30% (v/v) aqueous ethanol solution in a shaking water bath (Biofree, Seoul, Korea) for 30 min at 35 °C and 100 rpm. The extracted slurry was then filtered by using filter paper (110 mm, No. 2, Whatman, Brentford, UK), and the filtrate was centrifuged for 10 min at 13,000 rpm by using a microcentrifuge (Hanil, Seoul, Korea). The centrifuged filtrate was then further filtered by using a 0.45 μm syringe filter (Hydrophilic PTFE, Advantec, Dublin, USA) prior to HPLC analysis. The above analytical procedures were optimized and validated in a preliminary study by comparing with similar protocols found in other reports (Wang, Provan, & Helliwell, 2003; Wright et al., 2000).

The prepared sample was analyzed for caffeine and catechin contents using a HPLC system (Agilent 1100, Agilent Technologies, Waldbronn, Germany). Twenty microlitres of the sample were injected into the HPLC system with a Hypersil ODS column (Hypersil ODS, 5 μm, 4.6 × 100 mm, Thermo Electron Corporation, Bellefonte, USA) operated at 20 °C with a gradient elution, as described elsewhere (Copeland, Clifford, & Williams, 1998), and peaks in the eluent were detected at 280 nm.

3. Results and discussion

3.1. Composition of green tea before SFE

The caffeine and catechin contents in the green tea used in the study were analyzed, as is shown in Table 1. Our analysis indicated that the caffeine content in the green tea was as high as 41.4 mg/g on a dry weight basis, which is considered to be comparable to the caffeine contents of green teas reported by others, e.g., 35.0 (Chu et al., 1997), 36.0 (Perva-Uzunalic et al., 2006), and 51.9 mg/g of dry green tea (Wright et al., 2000). The most abundant catechin was EGCG, and the catechin contents were determined to be in the following order: EGCG > EGC > ECG > EC. The contents of EGCG and EGC in the green tea were relatively higher than most reported ones, but the high values such as those of EGCG, 210–145 mg/g and EGC, 37–81 mg/g, can also be found elsewhere (Chu et al., 1997; Perva-Uzunalic et al., 2006; Wright et al., 2000).

After the SC–CO_2 extraction step, it is necessary to apply a drying process to the extracted green sample in order to prepare dry samples for the quantitative analysis of caffeine and catechins, especially to remove any residual cosolvent and moisture from the extracted green tea. To expedite the drying process, the feasibility of the oven-drying procedure at 105 °C overnight was tested by comparing the changes in caffeine and catechins of dry green tea before and after 105 °C drying, as shown in Table 1. With the exception of EC, which is present at only minimal concentrations among the green tea components, the differences between the caffeine and catechin contents of the samples prior to and after drying were within 5%. Therefore, the 105 °C oven-drying method was utilized for the rapid analysis of extracted green tea samples in the present study.

3.2. Effect of water as a cosolvent

Supercritical CO_2 is a non-polar or low-polar solvent, owing to the fact that carbon dioxide possesses two oxygen atoms attached to a carbon atom in perfect symmetry. Therefore, SC–CO_2 is generally utilized in the extraction of solutes possessing little or no polarity. Due to the high polarity of caffeine, it is desirable to add a polar solvent to SC–CO_2 as a cosolvent in order to increase the polarity of SC–CO_2, so that the modified SC–CO_2 can be used to extract the caffeine from the green tea. In this study, water and ethanol (95%, v/v) were evaluated as a cosolvent for SC–CO_2, as water and grain ethanol (190 proof) can be used for green tea without the concern of food safety.

Table 2 shows the effects of water as a cosolvent on the extraction yields of the components of SC–CO_2 extraction conducted with the addition of differing amounts of water at a CO_2 flow rate of 8.5 g/min, a pressure of 300 bar, and a temperature of 70 °C for 120 min. When 8.8 g of water per 100 g of CO_2 was utilized for the extraction, 31.1 mg of caffeine and 100 mg of EGCG were extracted per g of dry green tea, and their extraction yields, on the basis of the total quantity of CO_2 used, were 289 mg and 931 mg per kg of CO_2 used, respectively.

As the water concentration was increased from 0.0 to 8.8 g per 100 g of CO_2, both the caffeine and catechin contents in the extracted green tea were significantly reduced following SC–CO_2 extraction. These results imply that the increased polarity of the modified SC–CO_2, due to

Table 1
Effect of drying on the composition analysis of green tea

Component (mg/g)[a]	Caffeine	EGCG	EGC	ECG	EC
As received (before drying at 105 °C)	41.4 ± 0.41	145 ± 0.89	81.5 ± 3.72	27.3 ± 0.49	6.01 ± 0.14
After drying at 105 °C	40.2 ± 0.38	143 ± 1.51	85.6 ± 3.42	26.3 ± 0.25	4.89 ± 0.57

Each value is expressed as mean ± standard deviation from triplicate data.
[a] Concentration of each component is mg of component per g of green tea on a dry weight basis.

Table 2
Supercritical CO_2 extraction yields of caffeine and catechins from green tea leaves with particle sizes of 425–710 μm when operated at a CO_2 flow rate of 8.5 g/min, a pressure of 300 bar, and a temperature of 70 °C for 120 min with cosolvents

Cosolvent (g per 100 g of CO_2)	Extraction yield									
	(mg component/g dry green tea)					(mg component/kg total CO_2 used)				
	Caffeine	EGCG	EGC	ECG	EC	Caffeine	EGCG	EGC	ECG	EC
No Cosolvent	3.7 ± 0.5	19.2 ± 2.1	11.9 ± 0.2	3.4 ± 0.6	1.0 ± 1.1	34.8 ± 0.5	178.3 ± 2.1	111 ± 0.2	32.0 ± 0.6	9.9 ± 1.1
Water, 2.9 g	6.2 ± 0.4	46.2 ± 2.5	37.9 ± 2.2	6.1 ± 0.9	0.6 ± 0.4	57.5 ± 0.4	429.1 ± 2.5	352 ± 2.2	57.3 ± 0.9	5.5 ± 0.4
Water, 5.8 g	16.9 ± 0.1	72.1 ± 0.4	54.1 ± 1.0	11.9 ± 0.0	3.5 ± 0.0	157 ± 0.1	669.4 ± 0.4	502 ± 1.0	111 ± 0.0	32.5 ± 0.0
Water, 8.8 g	31.1 ± 0.3	100 ± 1.8	74.6 ± 0.2	15.7 ± 0.2	5.4 ± 0.3	289 ± 0.3	930.6 ± 1.8	693 ± 0.2	146 ± 0.2	50.1 ± 0.3
EtOH, 2.3 g	28.7 ± 0.3	63.1 ± 5.2	55.0 ± 1.1	12.0 ± 0.7	1.7 ± 0.1	267 ± 0.3	585.7 ± 5.2	511 ± 1.1	112 ± 0.7	16.3 ± 0.1
EtOH, 4.6 g	35.0 ± 0.1	77.6 ± 1.7	55.4 ± 0.9	15.5 ± 0.1	1.0 ± 0.1	326 ± 0.1	720.8 ± 1.7	514 ± 0.9	144 ± 0.1	9.3 ± 0.1
EtOH, 7.0 g	40.3 ± 0.2	90.4 ± 0.1	65.0 ± 2.2	19.1 ± 0.2	2.6 ± 0.6	374 ± 0.2	838.8 ± 0.1	604 ± 2.2	177 ± 0.2	24.8 ± 0.6

Each green tea sample was extracted in triplicate, each extract was analyzed by HPLC in duplicate, and the calculated extraction yields were expressed as means ± standard deviations.

the use of water as a cosolvent, also enhanced the extraction of caffeine and catechins with relatively high polarity. Besides, the extraction may have been enhanced by the facilitated release of solutes through the swollen channels of the solid matrix of the green tea, as previously found in the decaffeination of coffee beans (Peker et al., 1992).

3.3. Effect of ethanol as a cosolvent

SC–CO_2 extraction was also conducted using aqueous ethanol (95%, v/v) as a cosolvent at a CO_2 flow rate of 8.5 g/min, a pressure of 300 bar, and an extraction temperature of 70 °C for 120 min, as shown in Table 2. When 7.0 g of ethanol per 100 g of CO_2 were used in the SC–CO_2 extraction, 40.3 mg of caffeine and 90.4 mg of EGCG were extracted from each gramme of dry green tea. In this case, 97.4% and 62.2% of the initial contents of caffeine and EGCG were extracted, respectively. Also, the extraction efficiencies, and the yield of component on the basis of the total mass of CO_2 used, were 374 mg of caffeine and 839 mg of EGCG per kg of CO_2.

As the extraction yields of caffeine and catechins increased, in parallel with increases in the ethanol content, these results were similar to those observed when water was used as a cosolvent. However, by comparison of the results of the added cosolvents, water and ethanol in Table 2, caffeine was much more effectively removed when using ethanol, but the losses of EGCG by extraction were similar for both water and ethanol. This may be attributable to the fact that ethanol, which is less polar than water, is more effective in caffeine extraction than in catechin extraction, as caffeine is less polar than catechins. Besides, the higher solubility of ethanol than water in SC–CO_2 (Delaossa, Brandani, Delre, Digiacomo, & Ferri, 1990; Yao, Guan, & Zhu, 1994) may have contributed to the enhanced extraction of caffeine when using ethanol rather water as a cosolvent. Therefore, it was considered that CO_2/ethanol was the superior solvent/cosolvent system since it gave a higher yield of caffeine extraction but still preserved EGCG at a similar level compared to CO_2/water.

Taken together in Table 2, the extraction efficiency of caffeine per unit weight of used CO_2 from the green tea leaves ranged from 57.5 to 374 mg/kg of CO_2 at 70 °C, 300 bar, and a CO_2 flow rate of 8.5 g/min for 120 min when water or 95% aqueous ethanol was used as a cosolvent. These results can be compared to the extraction efficiencies of caffeine from several other plants based on the amount of used CO_2. For example, coffee beans soaked with water showed an extraction efficiency of 45.2–121 mg of caffeine/kg of CO_2 at 50 °C, 103–193 bar, and a CO_2 flow rate of 1.51 g/min for 200 min (Peker et al., 1992). When coffee beans were acidified and wetted with citric acid solution prior to SC–CO_2 extraction, the extraction efficiency of 48.1 mg of caffeine/kg of CO_2 was achieved at 90 °C, 280 bar, and a CO_2 flow rate of 106 g/min for 8 h (Kazlas, Novak, & Robey, 1994). In the case of black tea, the extraction efficiency was 60 mg of caffeine/kg of CO_2 at 63 °C, 260 bar, and a CO_2 flow rate of 3.3 kg/min for 120 min (Hurberturs, Erwin, & Heinz-Riidiger, 1990) In the extraction of guarana seeds, the extraction efficiency was 74.3–119 mg caffeine/kg of CO_2 for with at 40–70 °C, 400 bar and a CO_2 flow rate of 5.7 g/min for 210 min (Saldana, Zetzl, Mohamed, & Brunner, 2002). Mate leaves exhibited the extraction efficiency of 7.2–5.3 mg of caffeine/kg of CO_2 when extracted at 40–70 °C, 400 bar, and a CO_2 flow rate of 5.7 g/min for 400 min (Saldana et al., 2002). Among these plant sources, the extraction efficiency of mate leaves was much lower than those of green tea leaves, coffee beans or guarana seeds.

3.4. Effect of temperature

SC–CO_2 extractions were conducted at 4 different temperatures (50, 60, 70, and 80 °C) at a CO_2 flow rate of 8.5 g/min and a pressure of 300 bar for 120 min. As shown in Fig. 2, the extraction yield of caffeine increased with increases in temperature. The most pronounced removal (over 90% of its initial content) of caffeine was seen at the highest temperature, 80 °C. At equilibrium, the solubility curves of solute in the supercritical phase at different temperatures evidenced crossovers that were balanced by density and vapour pressure effects. When the density effect is predominant, at a higher temperature, a decrease in the solvent density results in the reduction of dissolving power of the supercritical fluid. In other words, the solubility of the solute in the supercritical phase decreases at a higher temperature. In event that the vapour pressure effect is overwhelming, the solubility of the solute rather increases at a higher temperature as it is strongly influenced by an increase in the vapour pressure of the solute. In this experiment, the enhanced caffeine extraction observed at higher temperatures was probably attributable to the predominance of the vapour pressure effect over the density effect.

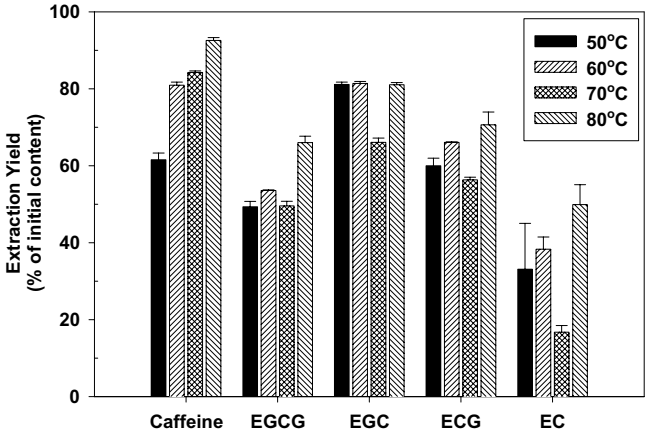

Fig. 2. Effects of extraction temperature on the extraction yields of green tea components in dry green tea (wt.% of their initial contents) extracted with supercritical CO_2 at 300 bar and at a CO_2 flow rate of 8.5 g/min for 120 min. The particle size of the green tea was 425–710 μm, and ethanol (95%, v/v) was utilized as a cosolvent at a concentration of 4.6 g per 100 g of CO_2. Control indicates the green tea before extraction.

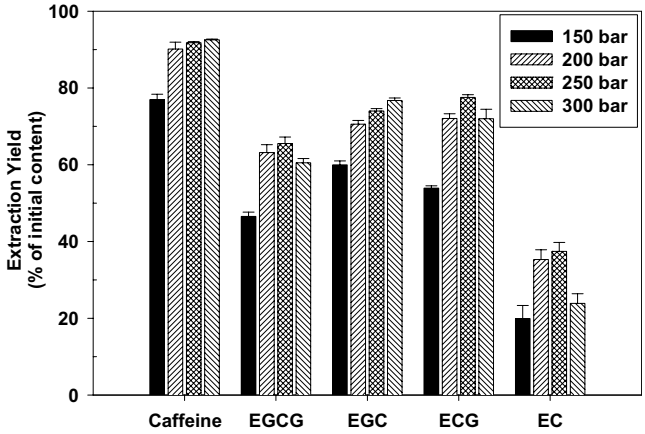

Fig. 3. Effects of extraction pressure on the extraction yields of green tea components in dry green tea (wt.% of their initial contents) extracted with supercritical CO_2 at 70 °C and at a CO_2 flow rate of 8.5 g/min for 120 min. The particle size of the green tea was 425–710 μm, and ethanol (95%, v/v) was used as a cosolvent at a concentration of 4.6 g per 100 g of CO_2. Control indicates the green tea before extraction.

EGCG, the most important catechin, was removed to a far less pronounced degree at 50, 60, and 70 °C than at 80 °C. Therefore, in order to minimize the loss of EGCG, and also considering the upper temperature limit of the system at 80 °C, the temperature 70 °C was recommended for extraction.

3.5. Effect of pressure

In order to determine the effects of pressure on caffeine extraction, extraction was conducted at four different pressures in the range of 150–300 bar at 70 °C, and at a CO_2 flow rate of 8.5 g/min for 120 min, as shown in Fig. 3. The extraction yields of caffeine and catechins generally increased with increasing extraction pressure except for EGCG, ECG, and EC at 300 bar. These results may be attributed to the increased dissolving power of SC–CO_2 as a result of the increased density induced by an increase in pressure at the same temperature. When the extraction pressure was 300 bar, over 90% of the caffeine was removed, as well as 60% of the EGCG.

4. Conclusion

In a study to investigate the effects of extraction conditions, including temperature, pressure and a cosolvent, on the removal of caffeine from dry green tea leaves, the caffeine content in the decaffeinated green tea was reduced to 2.6% of its initial content after extraction with SC–CO_2 modified with 95% aqueous ethanol at 7.0 g per 100 g of CO_2 at 300 bar and 70 °C and at a CO_2 mass flow rate of 8.5 g/min for 120 min. However, a substantial quantity of EGCG was also lost during the decaffeination process, by as much as approximately 37.8% of the initial content under the above extraction conditions. Therefore, further improvements, such as the more selective extraction of caffeine than catechins, the recovery of EGCG and other catechins from the outlet stream, as well as the intentional reconstitution of original green tea flavour in decaffeinated green tea will be necessary topics for future studies.

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