AGRICULTURAL AND FOOD CHEMISTRY

Application of Microwave Energy in the Manufacture of Enhanced-Quality Green Tea

ASHU GULATI,* RENU RAWAT, BRAJINDER SINGH, AND S. D. RAVINDRANATH

Hill Area Tea Science Division, Institute of Himalayan Bioresource Technology, Palampur 176 061 (H.P.), India

Green tea manufacture was standardized with respect to the inactivation of polyphenol oxidase (PPO), rolling, and drying for quality manufacture. Inactivation of PPO by parching, steaming, microwave heating, and oven heating was monitored in tea shoots. The inactivated shoots were rolled under regimens of high and low pressures and dried by microwave heating, oven heating, or sun-drying; total phenols and catechins were estimated. Parched and sun-dried teas contained the lowest levels of total phenols and catechins, and their infusions were dull in color with a slightly burnt odor. Microwave-inactivated and-dried teas showed the highest levels of total phenols and catechins, and their infusions were bright in color and sweet in taste with a subtle pleasant odor. In steam-inactivated and oven/microwave-dried teas, total phenol and catechin contents were intermediate between parched and sun-dried teas and microwave-inactivated and microwave-dried teas, and their infusions were bright with a umami taste.

KEYWORDS: Tea; *Camellia sinensis*; catechins; polyphenols; polyphenol oxidase; microwave inactivation; sun-drying

INTRODUCTION

Tea is the most popular nonalcoholic beverage in the world. Tea is manufactured either following fermentation (CTC/ Orthodox tea) or nonfermention methods of manufacture (green tea). Green tea is popular in Eastern Asian countries (Japan, China, Taiwan, Korea, etc.) and is also slowly becoming popular in the Western countries. Recent studies have shown that the quality determinants of green tea, namely, total polyphenols and total catechins, are associated with medicinal properties such as antidiabetic, antimicrobial, anticancer, antioxidant, and antiaging activities (1-4).

Fresh shoots of tea are very rich in polyphenols, especially the flavan-3-ol group (catechin and its derivatives), which are present at up to 30% on a dry weight basis (5). In processing younger tea shoots for green tea manufacture, polyphenol oxidase (PPO) inactivation is necessary to avoid the oxidation of catechins to form theaflavins and thearubigins. These catechins are responsible for the astringency and taste of green teas (6-8).

Orthodox Kangra black tea from Himachal Pradesh, based on China hybrid, is well-known for its characteristic flavor in the first flush. Studies on seasonal variations have revealed loss in the first-flush quality through the main (rains) flush, with slight improvement in the backend flush. The loss in quality could be attributed to relatively fast growth, accompanied by dilution of quality-related biochemicals, mainly catechins and

PPO activity, monoterpene alcohol content, and lowered dry matter accumulation during rains flush (9). Lowered levels of catechins and small variations in amino acids through rains flush make the harvest suitable for green tea manufacture during this growth flush. China hybrid tea shoots contain relatively low levels of catechins and caffeine and high amino acids as compared to Assam hybrids and hence are better suited for green tea manufacture (10). Moreover, in the Kangra region 700000 kg of green tea is manufactured every year by following the traditional method of inactivation of tea shoots by roasting, rolling, and sun-drying. The quality of the product is highly variable. To obtain a consistent gree tea product, there is a need to standardize the conditions of processing and to compare green teas made by different known methods to arrive at an improvement in the quality of the product that could even be accepted by the international consumer.

With a view to developing a process for manufacturing quality green tea, the present study was, therefore, undertaken to follow/ monitor the changes in quality parameters, that is, total polyphenols and total catechins, during green tea manufacture. Major emphasis has been given to the process of enzyme inactivation and to drying, which are crucial for tea quality.

MATERIALS AND METHODS

Tea Shoot Sampling. Tea shoots comprising anapical bud and subtending three leaves were harvested at monthly intervals for two consecutive years (March 1999–October 2000) from plots under regular

10.1021/jf026227q CCC: \$25.00 © 2003 American Chemical Society Published on Web 07/03/2003

^{*} Corresponding author (e-mail ashugulati2000@yahoo.co.in).

7-day plucking at the Banoori Tea Experimental Farm of the Institute of Himalayan Bioresource Technology, Palampur, India. The plucked shoots were divided into 28 parts of 2 kg each.

Tea Manufacture. The plucked shoots were immediately processed for PPO inactivation by one of the following treatments:

(a) Shoots were exposed in a covered glass container to microwave energy for different times from 2 to 6 min at power level 7 in a BPL-Sanyo microwave—convection domestic microwave oven model BMC 900T (34 L capacity, microwave 1.35 kW, frequency 2450 MHz, with a loading capacity of 1.5 kg of green leaves).

(b) Shoots were exposed to microwave energy with no cover for different times from 2 to 6 min at power level 7 in a BPL-Sanyo microwave-convection domestic microwave oven (same as above).

(c) Shoots were heated using a tray dryer with a temperature regulator (maximum = 300 °C) and air circulator at different temperatures with the air vent open for different times from 2 to 10 min (Narang Scientific Equipment, India), with a loading capacity of 25 kg of green leaves in 24 trays with 1-in. thickness.

(d) Shoots were heated using a tray dryer with a temperature regulator and an air circulator with the air vent closed for different times from 2 to 8 min, with a loading capacity of 25 kg green leaves in 24 trays with 1-in. thickness.

(e) Shoots were inactivated in a steamer for 2-8 min. [The steamer is locally fabricated, consisting of a boiler to generate steam, and the steam (at atmospheric pressure) is passed into a rotating drum consisting of a steam pipe and a shaft (the leaves are indirectly heated by steam), with loading capacity of 20 kg of green leaves.]

(f) Shoots were parched in a roaster for 2-8 min. (The roaster is locally fabricated and consists of a drum with a shaft and a hearth placed below it, and the drum was directly heated to dry the leaves; the roaster has a loading capacity of 20 kg of green leaves.)

The tea shoots were cooled for 20-30 min by passing air at ambient temperature over them after inactivation to stop further reactions such as cooking and browning of the leaves, thus conditioning the leaves for rolling.

The shoots were rolled at different pressures (high and low) for different time regimens varying from 5 to 20 min in a piezy roller in the mini tea factory at IHBT, Palampur, following inactivation of PPO. (The roller was manufactured locally and standardized for performance, data not presented.) The shootes were partially dried in a conventional tea dryer.

The shoots were then processed for a second roll and dried. The final drying was done either by spreading leaves directly under the sun on the floor at ambient conditions (sun-drying), by microwave-drying carried out at power level 7, or by conventional oven-drying at 100 $^{\circ}$ C, at ambient humidity conditions.

PPO activity and total polyphenol and catechin contents were estimated after each treatment.

Polyphenol Oxidase Activity Estimations. Acetone powder (0.2 g) of freshly plucked shoots with moisture contents of 75–78% at the time plucking, prepared by homogenizing samples in chilled acetone (-20 °C) and dried under vacuum, was extracted with a 0.2 M Na₂-SO₄ solution (15 mL) and assayed spectrophotometrically with (+)-D-catechin (5 mM) as substrate (*11*) with necessary enzyme and substrate controls. The enzyme activity was expressed as units per gram of acetone powder, with unit activity equivalent to 0.001 absorbance change per minute at 380 nm. Absorbance/optical density (OD) of test solutions was measured with a Hitachi 150-20 spectrophotometer.

Catechin and Total Phenol Estimations. Shoot samples were dried to constant weight in a microwave oven. Catechins, extracted from 100 mg of dried leaf powder in 50% acetone, were estimated by measuring absorbance of yellow color developed with diazotized sulfanilamide at 425 nm (12) and expressed as (+)-D-catechin equivalents. Total phenols were quantitatively estimated from 100 mg of dried leaf powder using Folin–Ciocalteu reagent in the presence of Na₂CO₃, and absorbance of blue color developed was read at 760 nm (13). Absorbance/optical density (OD) of test solutions was measured with a Hitachi 150-20 spectrophotometer. Total phenols and catechins were expressed on a dry weight basis.

Tasters' Infusions. Green tea (3 g) was infused with 125 mL of water at 80 and 100 °C for 3 min in a taster's cup and filtered. The filtered teas were tasted by a group of tea tasters. The leaf style was noted, and total polyphenols and total catechins in the infusions were estimated. The values (**Table 5**) represent milligrams of total polyphenol and catechins extracted as soluble solids by infusion in a cup.

RESULTS AND DISCUSSION

The inactivation of PPO in fresh tea shoots was achieved by treating the shoots for different times either by microwave energy, oven heating, parching, or steaming (**Table 1**). The standardization of inactivation conditions under microwave treatment is shown in **Table 2**.

The shoots were heated under microwave power at level level 7 for different times and were kept either under a glass cover (treatment under cover) or with no glass cover (treatment no cover) analogous to steam inactivation and roasting, respectively. Microwave heating shoots under cover accomplished the inactivation of PPO in 4 min. The inactivated shoots were sticky/ gummy and did not brown even when kept for a longer time (6 min) (**Table 1**). Cover provides a closed environment where heat is trapped and generates more steam, which prevents the leaf from drying out and helps to more quickly inactive the enzyme. The temperature of the inactivated leaf at the end of treatment by microwave heating under cover was ~ 80 °C.

In the case of microwave oven treatment of shoots without a cover, the inactivation was achieved in less time (3 min), but the shoots were slightly dry and withered and the edges of the leaves browned if kept for a longer time (6 min). In the uncovered microwave treatment, there is air circulation in the system and moisture escapes from the surface more rapdily, leaving the edges slightly dried and brown. As a consequence, it took 1 min less to inactivate the enzyme. The temperature of the shoots at the end of treatment by microwave without cover was 70 °C.

The inactivation of PPO by oven took a longer time compared to microwave inactivation of shoots (**Table 1**). The inactivation of shoots by oven showed different time optima for inactivation under different temperatures. The shoots were greener and sticky when inactivated with the air vent closed (explanation similar to microwave covered treatment) than with the air vent open (similar to microwave uncovered treatment), but a longer time was needed to inactivate the enzyme with the air vent open because in the air vent open treatment the moisture loss is higher and the relative humidity is low (procedure similar to drying). The inactivated shoots had drier edges when inactivated at higher temperatures in the oven irrespective of closed/open air vent (100 °C). The temperature of the inactivated shoots was 75–80 °C.

The shoots inactivated by employing steam (95–102 °C) were greener in color compared to the shoots parched in traditional roasters but were quite wet and needed a flow of air to remove the extra moisture associated with them. The greenness in the shoots inactivated by steam could be related to the steam protection/cover to the shoots absent in the roasted shoots. The final temperature of the inactivated shoots by roasting and steaming was between 75 and 80 °C.

However, the contents of polyphenols and catechins did not show much variation within the different sets of treatments. Inactivation at lower temperatures took longer with the tea leaves turning brown and their infusion being dark and reddish in color, which indicated the oxidation of catechins to theaflavins and thearubigins (data not shown). This also lowered the quality of green tea.

Table 1. Changes in Chemical Composition of Tea Shoots Following Different Methods of Polyphenol Oxidase Inactivation

			total	total	
PPO inactivation conditions		PPO ^a	phenol %	catechin %	remarks on leaf appearance after treatment ^b
untreated		1804	12.58	12.04	
parching	2 min	1235	12.72	12.13	
1 5	3 min	580	12.56	11.78	
	4 min	IA	12.16	11.54	shoots slightly sticky and browned if treated longer
steaming	2 min	1301	12.66	12.42	5 5 5 5
3	3 min	650	12.54	12.11	
	4 min	IA	12.24	11.71	shoots wet, did not brown if treated longer
covered shoots microwaved for	2 min	1257	12.54	12.19	
	3 min	459	12.67	12.18	
	4 min	IA	12.84	12.56	shoots gummy/sticky, did not brown if treated longer
	6 min	IA	12.76	12.19	6 5 5
uncovered shoots microwaved for	2 min	1156	12.81	12.43	
	3 min	IA	12.82	12.24	shoots gummy/sticky, but edges browned if treated longer
	4 min	IA	13.09	12.24	
	6 min	IA	12.56	11.59	
oven-inactivated, covered at 60 °C for	4 min	1456	13.52	11.45	
	8 min	851	13.25	10.73	
	10 min	IA	12.55	11.05	shoots gummy/sticky, but edges browned if treated longer
oven-inactivated, covered at 80 °C for	4 min	1156	12.97	12.09	
	6 min	357	12.95	12.04	
	8 min	IA	12.85	11.81	shoots less gummy/sticky, but edges browned if treated longer
oven-inactivated, covered at 100 °C for	2 min	1195	12.23	12.07	
	4 min	457	12.62	11.83	
	6 min	IA	12.79	11.89	shoots less gummy/sticky, but browned if treated longer
	8 min	IA	12.63	11.89	
oven-inactivated, uncovered at 100 °C for	4 min	776	12.23	11.75	
	6 min	454	12.52	11.23	
	8 min	IA	12.05	10.83	shoots slight gummy/sticky, but browned if treated longer
SEM		0.133	0.535	0.542	
CD 5%		0.559	2.115	2.452	
CV		1.45	2.11	2.85	

^a Enzyme activity is expressed as units g^{-1} of FW tissue, with unit activity equivalent to 0.001 absorbance change min⁻¹ at 380 nm. IA = inactive. ^b Treatment conditions that gave inactivation were evaluated.

 Table 2. Changes in Chemical Composition of Tea Shoots Following

 Polyphenol Oxidase Inactivation by Microwave Oven at Different

 Power Levels

microwave power level ^a	PPO	total phenol %	total catechin %
untreated	1828	13.25	12.22
2	1655	9.56	7.55
4	995	9.88	8.05
6	548	10.55	8.93
8	IA ^b	12.67	12.18
10	IA	12.76	12.19
SEM	0.145	0.245	0.232
CD 5%	0.585	1.093	0.942
CV	1.21	1.62	1.85

^{*a*} Treatment was given for 4 min, and shoots were kept covered with a glass lid. $^{$ *b* $}$ IA = inactive.

The inactivated tea shoots were rolled under different pressures (high and low) for different times to optimize the catechin and polyphenol contents and then half-dried using tea dryers and microwave ovens (**Table 3**). The shoots half-dried by microwave showed higher values of catechins and polyphenols compared to those dried in a conventional tea dryer.

The shoots were processed for a second roll at a single pressure (low) only and finally dried using microwave ovens, conventional tea dryers, or the sun. The tea dried by microwave ovens showed higher contents of polyphenols and catechins compared to the other two methods of drying (**Table 4**). The sun-dried samples had the lowest polyphenol and catechin contents and required being kept in conventional ovens or microwave ovens for complete drying as they still had higher moisture contents (6%). The sun-dried teas were dull in color, and their infusions were comparatively dark and possessed an unpleasant odor.

Total polyphenols and total catechin contents were extracted in the infusions following the Japanese (infusing leaves with water at 80 °C) and Chinese methods of preparation (infusing at 100 °C) (**Table 5**). The total polyphenols and total catechins were higher in microwave-dried treatments perhaps because microwave inactivation might have prevented the binding of polyphenol and catechin to the leaf matrix, thereby increasing extractability with water. Tea tasters suggested that the infusion of teas, manufactured by either microwave oven or oven inactivation, when prepared at 80 °C were light and sweet, having a umami taste, whereas the infusions prepared at 100 °C were stronger and slightly bitter. The tasters were of the opinion that these teas would serve the green tea drinking population with myriad taste by selecting the infusion temperature.

Teas made from microwave oven- and oven-inactivated shoots were bright green, and their infusions had a subtle pleasant odor, whereas the teas made from roasted shoots had a burnt odor and the teas as well as the infusions were dull in color.

Use of a microwave oven for inactivation and drying in green tea manufacture compared well to use of conventional steaming and roasting of shoots for the inactivation of PPO and sundrying.

These studies were conducted at laboratory scale with the largest available domestic microwave oven; studies on large sample sizes may be required for utilization of microwave energy for green tea manufacture at commercial scale.

Table 3. Changes in Chemical Composition Following Different Rolling Procedures

rolling conditions	total phenol %	total catechin %
microwave oven inactivated, rolled for 15 min at low pressure and for 15 min at high pressure	12.21	12.04
microwave oven inactivated, rolled for 10 min at low pressure and for 20 min at high pressure	12.31	12.08
microwave oven inactivated, rolled for 5 min at low pressure, for 20 min at high pressure, and for 5 min at low pressure	12.21	11.16
microwave oven inactivated, rolled for 15 min at low pressure and for 15 min at high pressure	12.42	12.07
microwave oven inactivated, rolled for 10 min at low pressure, for 15 min at high pressure, and for 5 min at low pressure	12.42	12.13
microwave oven inactivated, rolled for 30 min at low pressure	12.43	11.01
microwave oven inactivated, rolled for 20 min at high pressure	12.34	11.05
SEM	0.356	0.228
CD 5%	1.502	0.985
CV	0.589	0.658

Table 4. Changes in Chemical Composition Following Different Methods of Drying

manufacture conditions	total phenol %	total catechin %
inactivated, rolled, half-dried by conventional dryer, second roll, finally and completely dried by microwave oven	12.04	11.01
inactivated, rolled, half-dried by conventional dryer, second roll, finally and completely dried by oven	12.04	11.71
inactivated, rolled, half-dried by conventional dryer, rolled, finally and completely sun-dried	11.64	10.87
inactivated, rolled, half-dried by conventional dryer, second roll, no complete drying	12.00	11.17
inactivated, first roll, half-dried by conventional dryer	12.07	11.31
inactivated shoot (microwaved)	12.21	12.03
untreated	12.58	12.04
SEM	0.428	0.352
CD 5%	1.528	1.211
CV	1.852	1.228

Table 5. Total Polyphenol and Total Catechin Extracted at Different Temperatures in a Cup

sample processing conditions	tasters' opinion ^a	polyphenol (mg/cup, 80 °C)	polyphenol (mg/cup, 100 °C)	catechin (mg/cup, 80 °C)	catechin (mg/cup, 100 °C
oven-inactivated and sun-dried	+	62.0	63.3	53.1	56.1
oven-inactivated and oven-dried	+++	10.8	108.0	85.8	94.2
oven-inactivated and microwave-dried	+++	106.5	116.4	80.7	97.5
steam-inactivated and sun-dried	+	55.8	94.2	51.9	86.1
steam-inactivated and oven-dried	++	94.2	95.7	67.2	61.8
steam-inactivated and microwave-dried	++	96.6	107.7	76.8	82.5
oasting and sun-dried	+	54.3	65.7	48.6	62.1
oasting and oven-dried	++	86.4	93.3	73.8	84.3
oasted and microwave-dried	++	108.0	117.0	96.9	92.4
nicrowave-inactivated and sun-dried	+	63.9	73.2	56.4	63.6
nicrowave-inactivated and oven-dried	++	97.8	99.3	90.0	95.1
nicrowave-inactivated and microwave-dried	++++	116.4	112.2	103.8	101.4
SEM		0.214	0.29	0.058	0.194
CD 5%		0.789	1.28	0.224	0.759
CV		0.884	1.42	0.513	0.118

^a + indicates briskness in the cup; ++++ indicates highly brisk tea.

In conclusion, total phenols and total catechin contents as well as leaf style and infusion of green teas manufactured following different inactivation and drying methods showed that the teas made following oven and microwave inactivation and microwave drying are better in quality compared to the other sets of treatments.

ACKNOWLEDGMENT

We express gratitude to the Director, Institute of Himalayan Bioresource Technology (CSIR), for valuable suggestions and encouragement. We thank R. S. Shekhawat, V. S. Dhadwal, and Khushal Chand for help in tea sampling and Dinesh Singh and Harmesh Bali for help in tea manufacture.

LITERATURE CITED

- Koketsu, M. Antioxidant activity of tea polyphenols. In *Chemistry and Application of Green Tea*, 1st ed.; Yamamoto, T., Juneja, L. R., Chu D. C., Kim, M., Eds.; CRC Press: London, U.K., 1997; pp 37–50.
- (2) Sakanaka, S.; Kim, M.; Taniguchi, M.; Yamamoto, T. Antibacterial substances in green tea extract against *Streptococcus mutans*, a cariogenic bacterium. *Agric. Biol. Chem.* **1989**, *53*, 2307.
- (3) Mukhtar, H.; Katiyar, S. K.; Agarwal, R. Green tea and skin anticarcinogenic effects. J. Invest. Dermatol. 1994, 3, 102.
- (4) Anderson, R. A.; Polansky, M. M. Tea enhances insulin activity. J. Agric. Food Chem. 2002, 50, 7182–7186.
- (5) Roberts, E. A.; Smith, R. F. The phenolic substances of manufactured tea. IX. The spectrometric evaluation of liquor. J. Sci. Food Agric. 1963, 14, 689–700.

- (6) Nakagawa, M. Chemical composition and taste of green tea. Jpn. Agric. Res. Q. 1975, 9, 156–160.
- (7) Nakagawa, M.; Anan, T.; Ishima, N. The relationship of green tea taste with its chemical composition. *Bull. Natl. Res. Inst. Tea* **1976**, *17*, 69–123.
- (8) Saijo, R. Isolation and chemical structure of new catechins from fresh tea leaves. *Agric. Biol. Chem.* **1982**, *46*, 1969–7190.
- (9) Gulati, A.; Ravindranath, S. D. Seasonal variations in quality of Kangra tea (*Camellia sinensis* (L) O Kuntze) in Himachal Pradesh. J. Sci. Food Agric. **1996**, 71, 231–236.
- (10) Samarasingham, S. A method for processing green tea. Sri Lankan J. Tea Sci. 1990, 59 (1), 16–23.
- (11) Singh, H. P.; Ravindranath, S. D. Occurrence and distribution of PPO activity in floral organs of some standard and local cultivars of tea. J. Sci. Food Agric. 1994, 64, 117–120.

- (12) Singh, H. P.; Ravindranath, S. D.; Singh, C. Analysis of tea shoot catechins: Spectrophotometric quantitation and selective visualization on two-dimensional paper chromatograms using diazotized sulfanilamide. J. Agric. Food Chem. 1999, 47, 1041– 1045.
- (13) Bray, H. G.; Thrope, W. V. Estimation of phenols. In *Methods of Biochemical Analysis*; Glick, D., Ed.; Interscience Publishing: New York, 1954; Vol. 1, pp 27–52.

Received for review December 18, 2002. Revised manuscript received March 7, 2003. Accepted April 24, 2003. We are grateful to the National Tea Research Foundation, Kolkata, for providing financial support. R.R. acknowledges financial assistance from NTRF.

JF026227Q