Supplementary materials

The effect of steaming and fermentation on nutritive values, antioxidant activities, and inhibitory properties of tea leaves

Chaowanee Chupeerach[1,2], Amornrat Aursalung[1], Thareerat Watcharachaisoponsiri[1], Kanyawee Whanmek[1], Parunya Thiyajai[1], Kachakot Yosphan[1], Varittha Sritalahareuthai[1], Yuraporn Sahasakul[1,2], Chalat Santivarangkna[1,2] and Uthaiwan Suttisansanee[1,2,*]

1. Institute of Nutrition, Mahidol University, Salaya, Phuttamonthon, Nakhon Pathom 73170, Thailand; chaowanee.chu@mahidol.ac.th (C.C.); amornrat.aur@mahidol.ac.th (A.A.); thareerat.wat@hotmail.com (T.W.); kanyaweebiosci@gmail.com (K.W.); parunya.thy@mahidol.ac.th (P.T.); kachakot.yp@gmail.com (K.Y.); varittha.sri@hotmail.com (V.S.); yuraporn.sah@mahidol.ac.th (Y.S.); chalat.san@mahidol.ac.th (C.S.); uthaiwan.sut@mahidol.ac.th (U.S.)

2. Food and Nutrition Academic and Research Cluster, Institute of Nutrition, Mahidol University, Salaya, Phuttamonthon, Nakhon Pathom 73170, Thailand

* Correspondence: uthaiwan.sut@mahidol.ac.th; Tel.: +662-800-2380 ext. 422

Supplementary Table S1:

Nutrient compositions of fresh, steamed, and fermented tea leaves per 100 g fresh weight.

Nutrients	Nutritive values (per 100 g fresh weight)		
	Fresh leaves	Steamed leaves	Fermented leaves
Energy (kcal)	24.36 ± 0.64 ^{c}	78.66 ± 4.74 ^{a}	61.14 ± 4.52 ^{b}
Moisture content (g)	93.46 ± 0.15 ^{a}	79.65 ± 1.19 ^{c}	84.32 ± 1.13 ^{b}
Protein (g)	3.30 ± 0.00 ^{c}	5.02 ± 0.02 ^{a}	3.53 ± 0.03 ^{b}
Fat (g)	0.12 ± 0.01 ^{c}	0.36 ± 0.01 ^{b}	0.50 ± 0.00 ^{a}
Carbohydrate (g)	2.54 ± 0.14 ^{c}	13.85 ± 1.17 ^{a}	10.63 ± 1.10 ^{b}
Total dietary fiber (g)	5.76 ± 0.15 ^{c}	10.56 ± 0.04 ^{a}	7.84 ± 0.01 ^{b}
Soluble dietary fiber (g)	1.00 ± 0.04 ^{c}	2.74 ± 0.02 ^{a}	2.16 ± 0.11 ^{b}
Insoluble dietary fiber (g)	4.76 ± 0.11 ^{c}	7.82 ± 0.01 ^{a}	5.68 ± 0.12 ^{b}
Ash (g)	0.60 ± 0.01 ^{c}	1.13 ± 0.00 ^{a}	1.02 ± 0.00 ^{b}
Minerals			
Calcium (mg)	38.42 ± 0.09 ^{c}	103.80 ± 0.52 ^{a}	70.90 ± 0.19 ^{b}
Sodium (mg)	14.84 ± 0.66 ^{c}	21.75 ± 0.73 ^{b}	110.30 ± 0.59 ^{a}
Potassium (mg)	178.24 ± 1.28 ^{c}	290.85 ± 0.71 ^{a}	272.47 ± 7.33 ^{b}
Magnesium (mg)	15.97 ± 0.50 ^{c}	29.76 ± 0.19 ^{a}	20.00 ± 0.58 ^{b}
Iron (mg)	0.47 ± 0.02 ^{c}	1.21 ± 0.00 ^{a}	1.05 ± 0.01 ^{b}
Zinc (mg)	0.57 ± 0.02 ^{a}	0.36 ± 0.02 ^{b}	0.32 ± 0.01 ^{c}
Vitamins			
Vitamin B1 (mg)	0.17 ± 0.01 ^{c}	0.85 ± 0.04 ^{a}	0.40 ± 0.00 ^{b}
Vitamin B2 (mg)	0.10 ± 0.01 ^{b}	0.14 ± 0.01 ^{a}	0.05 ± 0.00 ^{c}
Niacin (B3) (mg)	0.71 ± 0.01 ^{b}	0.75 ± 0.02 ^{a}	0.42 ± 0.00 ^{c}
Vitamin C (mg)	3.84 ± 0.15 ^{b}	9.35 ± 0.27 ^{a}	ND

All data were expressed as mean ± SD of triplicate experiments; ND: not detected. $^{a-c}$ showed significant difference ($p < 0.05$) of the same nutrients in different samples using one way ANOVA followed by Duncan's *post hoc* test.

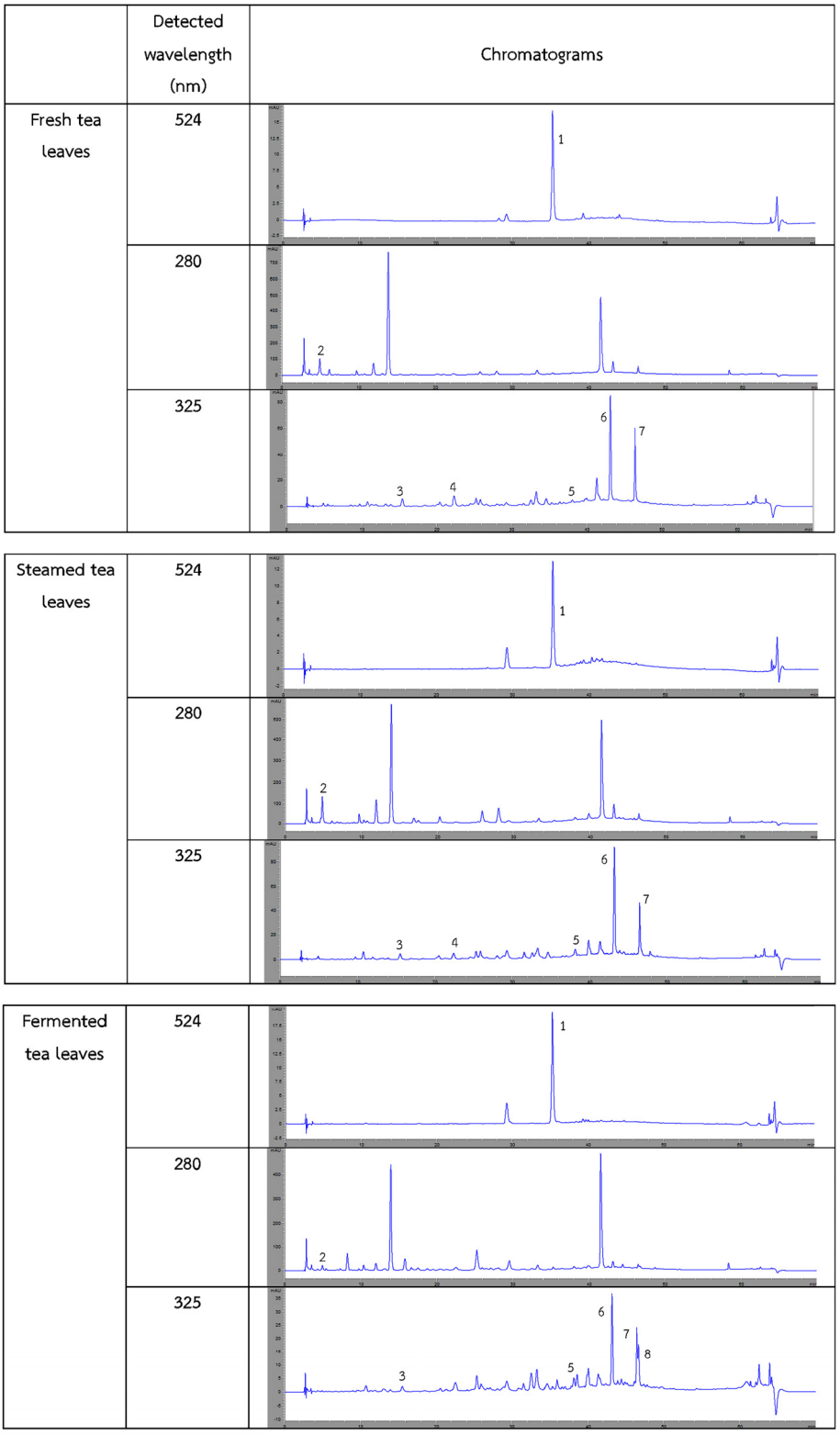
Supplementary Table S2:

The validation parameters used for HPLC analysis.

Standards	Retention time (min)	Linear range (μg/mL)	Linear regression equation	Correlation coefficient (R^2)	LOQ (μg/mL)	LOQ (μg/mL)	RSD (%)
Phenolic acids							
Gallic acid	5.01-5.27	0.78-200.00	y = 38.814x + 43.836	0.9998	2.66	8.07	1.37
4-Hydroxybenzoic acid	13.58-14.53	0.39-100.00	y = 54.999x - 11.469	0.9994	0.32	0.98	1.86
Chlorogenic acid	13.94-14.56	0.39-100.00	y = 45.107x - 8.1659	1.0000	0.03	0.10	1.59
Caffeic acid	17.06-17.80	0.39-100.00	y = 76.614x + 15.797	0.9998	0.59	1.78	1.70
Syringic acid	17.95-18.83	0.65-166.67	y = 37.286x - 24.557	0.9990	1.12	3.41	1.63
p-Coumaric acid	24.72-26.21	0.56-142.86	y = 44.809x - 22.611	0.9991	0.57	1.73	1.97
Ferulic acid	27.88-28.92	0.56-142.86	y = 42.477x - 16.212	0.9990	0.49	1.48	1.58
Sinapic acid	28.61-29.45	0.39-100.00	y = 63.807x + 10.841	1.0000	0.22	0.68	1.01
Flavonoids							
Hesperidin	38.06-38.64	0.65-333.33	y = 24.424x + 76.057	0.9990	2.56	7.77	0.64
Myricetin	39.45-40.02	0.65-166.67	y = 43.048x + 16.879	0.9999	0.37	1.12	0.32
Quercetin	44.58-44.75	0.65-166.67	y = 46.975x + 38.753	0.9999	0.19	0.58	0.10
Luteolin	44.87-45.54	0.65-166.67	y = 44.717x + 28.899	0.9999	1.19	3.62	0.52
Naringenin	45-41-45.87	0.65-333.33	y = 30.205x + 46.292	0.9999	0.46	1.38	0.45
Kaempferol	47.74-47.96	0.65-166.67	y = 47.952x + 43.855	0.9999	0.14	0.42	0.12
Apigenin	47.61-48.10	0.39-200.00	y = 40.778x + 34.994	0.9999	2.22	6.73	0.44
Isorhamnetin	48.15-48.69	1.30-333.33	y = 16.302x + 46.483	0.9992	0.45	1.37	0.39
Anthocyanidins							
Delphinidin	9.44-9.48	2.58-330.00	y = 1.6280x + 5.0454	0.9992	0.77	2.34	0.45
Cyanidin	16.74-16.85	2.58-330.00	y = 1.5641x + 4.4497	0.9995	0.05	0.15	0.58
Petunidin	18.26-18.33	3.91-125.00	y = 1.2904x + 4.4487	0.9951	0.78	2.36	0.28
Peonidin	36.62-36.96	2.58-330.00	y = 1.8154x + 4.1796	0.9995	3.08	9.34	0.73
Malvidin	42.51-42.61	3.91-125.00	y = 0.6093x + 4.1012	0.9945	1.82	5.53	0.24

Supplementary Figure S1:

High-performance liquid chromatograms of fresh, steamed, and fermented tea leaves detected at 280, 325, and 524 nm.



1 = cyanidin, 2 = gallic acid, 3 = caffeic acid, 4 = *p*-courmaric acid, 5 = myricetin, 6= querceitn, 7 = kaempferol, 8 = apigenin