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Chemical Composition, Plant Secondary Metabolites, and Minerals of Green and Black Teas and the Effect of Different Tea-to-Water Ratios during Their Extraction on the Composition of Their Spent Leaves as Potential Additives for Ruminants

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ABSTRACT: This study characterized the chemical composition of green and black teas as well as their spent tea leaves (STL) following boiling in water with different tea-to-water ratios. The green and black tea leaves had statistically similar (g/kg dry matter (DM), unless stated otherwise) DM (937 vs 942 g/kg sample), crude protein (240 vs 242), and ash (61.8 vs 61.4), but green tea had significantly higher (g/kg DM) total phenols (231 vs 151), total tannins (204 vs 133), condensed tannins (176 vs 101), and total saponins (276 vs 86.1) and lower neutral detergent fiber (254 vs 323) and acid detergent fiber (211 vs 309) than the black tea leaves. There was no significant difference between the green and black tea leaves for most mineral components except Mn, which was significantly higher in green tea leaves, and Na and Cu, which were significantly higher in black tea leaves. A higher tea-to-water ratio during extraction significantly reduced the loss of soluble compounds into water and hence yielded more nutrient-rich STL. On the basis of these analyses it appears that the green and black tea leaves alongside their STL have the potential for use as sources of protein, fiber, secondary metabolites, and minerals in ruminant diets. The presence of high levels of plant secondary metabolites in either tea leaves or their STL suggests that they may have potential for use as natural additives in ruminant diets.

KEYWORDS: *green tea, black tea, spent tea leaves, chemical composition, phenols, tannins, saponins, ruminants*

INTRODUCTION

Tea is one of the most popular drinks in the world and is perceived as being healthy. Tea drinks have been reported to have chemopreventive effects that inactivate potentially harmful free radical oxygen in the body system.[1−5] Moreover, tea drinks are known to prevent obesity,[6] breast cancer,[7] and coronary heart[8] and cardiovascular[9] diseases. Commercially, for example, in the preparation of bottled tea drinks, spent tea leaves (STL), which remain after preparation of the tea drink, are collected as a waste product. The utilization of this waste as a source of nutrients and bioactive compounds for ruminant feeding has been suggested for years,[10−20] and this is encouraging for a zero-waste agricultural system, safer environment, and feed cost efficiency. Tea leaves are rich in secondary metabolites such as phenolic antioxidants,[21−24] proteins, amino acids, lipids, sugars, vitamin, fiber,[25] and minerals.[26,27] However, not all of these chemicals are entirely dissolved during water extraction, and some proteins, fiber, lipids, minerals, and phenolic compounds can be retained in STL.[13,15,20] In fact, the solubility of these compounds during water extraction is likely to be influenced by tea-to-water ratios. Therefore, the tea beverage industries may prefer to apply higher tea-to-water ratios during extraction to obtain more concentrated tea drinks and consequently nutrient-rich STL. In ruminants, plant secondary metabolites such as phenols and tannins may increase the availability of rumen bypass protein and non-ammonia nitrogen supply, which can be absorbed in the small intestine due to their binding ability to plant proteins.[28,29] Tannins have the potential to reduce rumen methane production.[28,29] Similarly, tea saponins can reduce methane and ammonia production[30−32] by reducing protozoa and supposedly lowering the methanogenic activity of relevant microbes.[30,31] Tannin suplementation can improve animal health by reducing gastrointestinal nematotes[33,34] and improve the quality of ruminant products such as milk and meat by increasing the rumenic acid and polyunsaturated fatty acids and decreasing saturated fatty acids through altered biohydrogenation by changing the microbial population in the rumen.[35−37] Moreover, tea leaves have a considerable amount of minerals such as Ca, Cu, Fe, Mg, Mn, and Zn,[26,27] which must be provided in the diets of ruminants to meet their requirements for optimum growth.[38,39] The objectives of this study were (1) to characterize chemical components, secondary metabolites, and minerals in green and black tea leaves along with their STL and (2) to test the hypothesis that a higher tea-to-water ratio would affect the extraction of soluble compounds into water to yield more nutrient-rich STL.

MATERIALS AND METHODS

Sample Preparation. Green and black tea leaves were obtained from a tea processing company (PT. Kabepe Chakra), located in Bandung, West Java, Indonesia. The green tea was graded as *Sow Mee* (code SM #315), and the black tea was graded as *Broken Orange Pekoe*

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Fanning (code BOPF #355). Each tea batch was tested for standard quality before it was marketed. The above-mentioned tea grades were selected for sampling in this study because these were the most consistent grades being used by the local tea beverage industries. The fresh tea leaves were initially plucked from *Camellia sinensis* var. Assamica tea plants from the same farm. The farm has its land elevation of 1350−1500 m above the sea level and andosol soils. Plucked leaves were then subjected to either green and black tea processing by the company as illustrated in Figure 1.

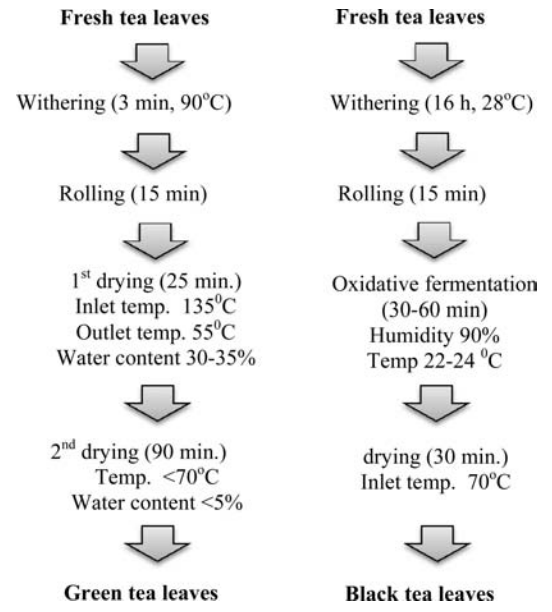

Figure 1. Typical sequences of green and black tea manufacturing in Indonesia.

After withering, the green tea is made by subjecting the fresh tea leaves to only the rolling and drying process. Black tea, however, is made by withering, rolling, and the oxidative fermentation process before drying. Representative samples of green and black tea leaves were collected from three different batches as replicates ($n = 3$). Each of these tea samples were then used by following a 3 × 2 factorial arrangement to obtain STL by extracting three different amounts (T1 = 2.8 g, T2 = 5.6 g, and T3 = 11.2 g) of the two tea types (green and black) in a fixed volume of 300 mL of boiling water for 5 min. Here, the T2 ratio was chosen to represent the ratio that is commonly used by the company to prepare tea drinks, whereas T1 and T3 ratios were selected to test how the lower and higher ratios can affect the tea extraction process to obtain variable qualities of STL. These changed ratios may be adopted by the industry to obtain tea drinks with modified organoleptic properties for humans and consequently STL with better nutrients for ruminants. After the tea extractions, each of the 18 STL (3 × 2 factorial, in triplicate) alongside the original green and black tea leaves was analyzed, in duplicate, for their chemical compositions as described below.

Chemical Analysis. Before chemical analysis, both tea leaves and their dried STL were ground through a 1 mm sieve using a sample mill (Cyclotec 1093, Tecator, Sweden). AOAC[40] methods were used to determine dry matter (DM), ash, organic matter (OM), and ether extract (EE), whereas total nitrogen (N) (N × 6.25 = crude protein, CP) and sulfur (S) were simultaneously analyzed by Elementar Vario Macro Cube (Elementar, Hanau Germany). The neutral detergent fiber (NDF) content was determined according to the method of Van Soest et al.[41] but without using amylase, sodium sulfite, and dekalin, whereas acid detergent fiber (ADF) was determined as reported by Van Soest.[42] The NDF and ADF contents were calculated by excluding ash. Total phenols (TP) and total tannins (TT) were analyzed using the Folin−Ciocalteu method as described by Makkar[43] with tannic acid (Fisher Scientific, Loughborough, UK) as the reference standard, whereas condensed tannins (CT) were analyzed according to the method of Makkar[43] with epigallocatechin gallate (Sigma-Aldrich, Gillingham, UK) as the reference standard. The procedure of Makkar et al.[44] was used for total saponin (TS) analysis using diosgenin (Molekula Ltd., Gillingham, UK) as a standard. Minerals were analyzed on a Varian Vista-MPX CCD by simultaneous inductively coupled plasma−optical emission spectroscopy (ICP-OES) (Varian Inc., Australia) by following the extraction procedure as described by Chaudhry and Jabeen.[45] Briefly, about 0.5 g of sample was weighed in a beaker and 9 mL of nitric acid added and kept overnight before 1 mL of perchloric acid was added. The mixture was then heated gradually to 150 °C on a hot plate until red NO_2 fumes turned colorless and the volume was reduced to around 1 mL. After cooling, the digested content was dissolved with distilled water, filtered (Whatman paper no. 541), and transferred into a 25 mL volumetric flask. Further dilutions were made with demineralized water as required to suit the standard curve calibrations. Commercial Ca, Zn, Ni, and Cu solutions (May and Baker Ltd., Dagenham, UK), $Mg(NO_3)_2$, $Mn(NO_3)_2$, $Fe(NO_3)_2$, and $Pb(NO_3)_2$ solutions, Cd (cadmium coarse powder), Cr (chromium(III) chloride 95%), Na (sodium chloride 99.5%) (BDH Chemicals, UK), P (sodium phosphate ≥99%) (Sigma-Aldrich), and K (potassium chloride, 99.8%) (Fisher Scientific) were used to prepare standard solutions at either 0−1.0 mg/kg, to represent lower, or 0−50 mg/kg, for higher, ranges of sample mineral concentrations.

Statistical Analysis. Minitab 16 software was utilized in all statistical analyses. One-way analysis of variance (ANOVA) was used to compare green with black teas and also original tea leaves with their STL across the tea-to-water ratios for each of their chemical components. Meanwhile, two-way ANOVA using the General Linear Model procedure was used to examine the statistical effects of tea types and tea-to-water ratios alongside their interaction on the chemical components of the STL from each extraction at $P < 0.05$ or below. Tukey's test was applied to compare means for statistical difference at $P \leq 0.05$. All data were analyzed for normality, and the data passed the Anderson−Darling normality test at $P > 0.05$.

RESULTS

Chemical Compositions of Green and Black Tea Leaves. Table 1 shows the proximate and fiber contents for the green and black tea leaves. The green and black tea leaves

Table 1. Mean (± SD, g/kg DM; $n = 6$) Chemical Compositions of Green and Black Tea Leaves Together with Their Standard Error of Means (SEM) and Significances[a]

composition	green tea	black tea	SEM with significance
DM	937 ± 3.56	942 ± 5.61	2.31^{NS}
OM	938 ± 1.67	939 ± 1.73	1.54^{NS}
CP	240 ± 1.02	242. ± 1.38	0.92^{NS}
EE	20.8 ± 3.29	12.6 ± 4.06	2.80**
ash	61.8 ± 1.67	61.4 ± 1.73	1.54^{NS}
S	2.74 ± 0.15	2.53 ± 0.22	0.11^{NS}
NDF	254 ± 12.0	323 ± 15.6	6.21***
ADF	211 ± 7.89	309 ± 9.02	3.82***
TP	231 ± 17.0	151 ± 9.61	7.98**
TT	204 ± 12.1	133 ± 6.79	5.69**
CT	176 ± 4.73	101 ± 22.8	9.49**
TS	276 ± 15.6	86.1 ± 3.69	6.56***

[a] Mean values were significantly different at $P < 0.05$ (*), $P < 0.01$ (**), or $P < 0.001$ (***); NS, nonsignificant; SD, standard deviation; *n*, number of replications; SEM, standard error of mean; DM, dry matter (g/kg sample); OM, organic matter; CP, crude protein; EE, ether extract; S, sulfur; NDF, neutral detergent fiber; ADF, acid detergent fiber; TP, total phenols; TT, total tannins; CT, condensed tannins; TS, total saponins.

had similar DM, CP, OM, ash, and S contents. Green tea leaves had a significantly higher EE content and significantly lower NDF and ADF contents than the black tea leaves. The green tea leaves also had significantly higher TP, TT, CT, and TS than the black tea leaves. There was no difference between green and black teas for most mineral components except Mn, which was significantly higher in green tea leaves than in black tea leaves, and Na and Cu, which were significantly lower in green tea leaves compared with black tea leaves (Table 2).

Table 2. Mean (± SD, mg/kg DM; *n* = 6) Mineral Components of Green and Black Tea Leaves Together with Their Standard Error of Means (SEM) and Significances[a]

composition	green tea	black tea	SEM with significance
Ca	6699 ± 179.6	6441 ± 648.6	274.8$^{\mathrm{NS}}$
K	8095 ± 744.3	7808 ± 233.7	318.5$^{\mathrm{NS}}$
P	2521 ± 55.00	2413 ± 241.8	101.2$^{\mathrm{NS}}$
Mg	1993 ± 49.60	1726 ± 169.6	72.17$^{\mathrm{NS}}$
Mn	663 ± 17.63	527 ± 50.95	22.00*
Fe	119 ± 5.31	116 ± 11.89	5.32$^{\mathrm{NS}}$
Na	78.2 ± 4.87	150 ± 11.37	5.05**
Cu	16.9 ± 0.54	23.8 ± 3.96	1.63*
Zn	21.2 ± 0.57	21.7 ± 2.45	1.03$^{\mathrm{NS}}$
Ni	1.58 ± 0.07	1.69 ± 0.22	0.09$^{\mathrm{NS}}$
Cr	1.32 ± 0.26	1.22 ± 0.12	0.12$^{\mathrm{NS}}$
Pb	0.51 ± 0.12	0.59 ± 0.18	0.09$^{\mathrm{NS}}$
Cd	0.04 ± 0.03	0.04 ± 0.02	0.01$^{\mathrm{NS}}$

[a] Mean values were significantly different at $P < 0.05$ (*), $P < 0.01$ (**), or $P < 0.001$ (***); NS, nonsignificant; SD, standard deviation; *n*, number of replications; SEM, standard error of mean.

Effect of Different Tea-to-Water Ratios on Chemical Components of Spent Tea Leaves. Tables 3 and 4 present the means of chemical components and minerals for only the main effects of STL types and tea-to-water ratios that were mostly significant but not their interactions. The green STL had significantly higher DM, CP, EE, ash, S, TP, TT, CT, and TS than black STL. However, green STL had lower WHC, OM, NDF, and ADF than black STL. For minerals, green STL had significantly higher concentrations of Ca, P, Mg, and Mn but lower concentrations of Fe, Na, Cu, Zn, Ni, Cr, and Pb than black STL. There were no significant differences between green and black STL for K and Cd.

Increasing tea-to-water ratios from T1 to T3 caused significant increases in DM, CP, ash, TP, TT, CT, and TS contents, but significantly decreased the WHC, OM, S, NDF, and ADF contents in STL. However, increasing tea-to-water ratios from T1 to T3 had no effect on most of the mineral components of STL except for an increase in K and a decrease in Ca and Mg in STL. Changing the tea-to-water ratio from T1 to T2 had no significant effect on most chemical components in STL except OM, CP, CT, and K (Tables 3 and 4).

Chemical Composition Changes between Original Tea Leaves and Their STL. Across the tea-to-water ratios, the extraction significantly altered most soluble chemical components from the original tea leaves to their STL except EE, S, Na, Cu, Cr, Pb, and Cd for the green tea and CP, S, CT, Mg, Mn, Na, Cu, Zn, Cr, Pb, and Cd for the black tea leaves. In green tea, the extraction significantly reduced DM (84.9%), ash (26.5%), TP (43.7%), TT (38.2%), CT (40.3%), and TS (74.5%) but significantly increased CP (5.0%), EE (10.6%), NDF (49.8%), and ADF (36.4%). Likewise, the green tea also showed significant changes in mineral components such as reductions in K (67.3%), P (12.3%), Mg (7.38%), Zn (9.43%), and Ni (69.0%) but significant increases in Ca (32.2%), Mn (11.9%), and Fe (18.5%) when compared with their green STL. In black tea, similar extraction significantly decreased DM (86.1%), ash (32.6%), TP (34.4%), TT (32.2%), and TS (54.4%) but significantly increased EE (14.3%), NDF (42.7%), and ADF (32.7%). For minerals, there were significant decreases in K (66.2%), P (20.9%), and Ni (30.8%) but significant increases in Ca (29.5%) and Fe (37.9%) in the black STL compared with the original black tea.

■ DISCUSSION

The lower EE and plant secondary metabolites in the black tea than in the green tea leaves were likely due to the degradation

Table 3. Mean Chemical Components (g/kg DM) of STL for the Main Effect of STL Tea Types (ST) and Tea-to-Water Ratios (Ratio, T1 = 2.8 g, T2 = 5.6 g, and T3 = 11.2 g/300 mL) Together with Their SEM and Significances[a]

	ST (*n* = 18)		tea-to-water ratios (*n* = 12)			SEM with significance		
composition	green	black	T1	T2	T3	ST	ratio	ST × ratio
DM	141	131	130 b	137 ab	140.8 a	1.72**	2.10*	2.97$^{\mathrm{NS}}$
WHC	61.1	66.4	67.0 a	63.2 ab	61.2 b	0.09**	0.11**	0.16$^{\mathrm{NS}}$
OM	955	959	959 a	956 b	955 b	0.57***	0.90**	0.99$^{\mathrm{NS}}$
CP	252	240	240 b	248 a	249 a	0.89***	1.09***	1.54$^{\mathrm{NS}}$
EE	23.0	14.4	18.3	18.1	19.7	0.56***	0.69$^{\mathrm{NS}}$	0.98$^{\mathrm{NS}}$
ash	45.4	41.4	41.0 b	43.8 b	45.4 a	0.57***	0.70**	0.99$^{\mathrm{NS}}$
S	2.90	2.58	2.90 a	2.73 ab	2.58 b	0.06**	0.07*	0.10$^{\mathrm{NS}}$
NDF	394	461	440 a	430 a	413 b	2.92***	3.57**	5.05$^{\mathrm{NS}}$
ADF	285	410	357 a	352 a	330 b	4.19***	5.13**	7.26$^{\mathrm{NS}}$
TP	130	99.0	108 b	113 b	122 a	1.76***	2.16**	3.05$^{\mathrm{NS}}$
TT	126	90.2	102 b	107 b	115 a	1.74***	2.13**	3.01$^{\mathrm{NS}}$
CT	105	77.3	64.0 b	93.8 a	116 a	4.96**	6.07***	8.59$^{\mathrm{NS}}$
TS	70.1	39.3	46.0 b	53.0 b	65.1 a	2.13***	2.61**	3.68$^{\mathrm{NS}}$

[a] Mean values with different letters in the same row were significantly different at $P < 0.05$ (*), $P < 0.01$ (**), or $P < 0.001$ (***); NS, nonsignificant; *n*, number of replication; SEM, standard error of mean; DM, dry matter (g/kg sample); WHC, water-holding capacity (g H_2O/kg DM); OM, organic matter; CP, crude protein; EE, ether extract; S, sulfur; NDF, neutral detergent fiber; ADF, acid detergent fiber; TP, total phenols; TT, total tannins; CT, condensed tannins; TS, total saponins.

Table 4. Mean Mineral Components (mg/kg DM) of STL for the Main Effect of STL Tea Types (ST) and Tea-to-Water Ratios (Ratio, T1 = 2.8 g, T2 = 5.6 g, and T3 = 11.2 g/300 mL) Together with Their SEM and Significance[a]

	ST (n = 18)		tea-to-water ratios (n = 12)			SEM with significance		
composition	green	black	T1	T2	T3	ST	ratios	ST × ratio
Ca	8860	8339	8799 a	8581 ab	8418 b	59.5***	72.8*	103^{NS}
K	2644	2642	1913 c	2532 b	3485 a	27.0^{NS}	33.1***	46.8**
P	2211	1908	2028	2058	2092	13.6***	16.7^{NS}	23.6^{NS}
Mg	1846	1638	1785 a	1744 ab	1696 b	11.9***	14.6**	20.7^{NS}
Mn	742	535	639	642	636	5.29***	6.48^{NS}	9.16^{NS}
Fe	141	160	152	156	142	3.71**	4.54^{NS}	6.43^{NS}
Na	98.6	190	118	137	177	14.8***	18.1^{NS}	25.7^{NS}
Cu	16.4	23.9	20.2	20.3	20.1	0.41***	0.50^{NS}	0.71^{NS}
Zn	19.2	22.2	20.8	20.9	20.3	0.16***	0.20^{NS}	0.28^{NS}
Ni	0.49	1.17	0.78	0.83	0.89	0.04***	0.05^{NS}	0.07^{NS}
Cr	1.12	1.42	1.36	1.26	1.17	0.04**	0.06^{NS}	0.08^{NS}
Pb	0.47	0.66	0.53	0.56	0.61	0.05*	0.06^{NS}	0.09^{NS}
Cd	0.04	0.04	0.04	0.05	0.04	0.00^{NS}	0.00^{NS}	0.00^{NS}

[a] Mean values with different letters in the same row were significantly different at $P < 0.05$ (*), $P < 0.01$ (**), or $P < 0.001$ (***); n, number of replications; NS, nonsignificant; SEM, standard error of mean.

of these components during the oxidative fermentation of black tea manufacturing.[46,47] Despite the reduction of some components in the tea leaves, this process of tea preparation was intended to improve extrinsic qualities such as the color, flavor, brightness, and taste of the tea drinks.[48,49] According to Yamamoto et al.,[25] the CP contents of the black and green tea leaves ranged from 182 to 307 g/kg DM, respectively, which were in line with the CP contents reported in this study. However, the TP composition of green tea (231 g/kg DM) of this study was higher than those from studies by Anesini et al.[50] (143−210 g/kg DM) and Khokhar and Magnusdottir[23] (87.0−106 g/kg DM), whereas the TP in black tea measured in this study (151 g/kg DM) was also higher than in the study by Khokhar and Magnusdottir[23] (80.5−135 g/kg DM) but within the range of the study by Anesini et al.[50] (84.2−176 g/kg DM). In comparison with the black tea that was reported by Salahinejad and Aflaki,[26] the black tea in this study had higher Ca but lower Cu, Fe, Mn, Mg, Zn, Ni, Cr, Pb, and Cd. However, Shen and Chen[27] reported lower Fe, Mg, and Zn in black tea and lower Cu, Fe, Mg, and Zn in green tea compared with the black and green teas of this study. These chemical differences could be expected because worldwide there are various qualities, brands, and grades of both green and black teas that are bound to affect the chemical composition of different tea types. These differences in chemical compositions also reflected the differences in varieties, soil types, and manufacturing processes that different tea leaves have been exposed to during their different phases of growth and processing. For example, the samples of this study were obtained from *C. sinensis* var. Asamica cultivated on the Java island of Indonesia, whereas the samples of Anesini et al.[50] were from *C. sinensis* (L.) O. Kuntze cultivated in northern Argentina, and Salahinejad and Aflaki[26] used some local commercial teas cultivated in northern Iran as well as imported samples from India and Ceylon.

As the STL are usually obtained as wet materials, their DM contents are expected to be <500 g/kg sample. It was reported that STL (green type), obtained from tea beverage companies were low in DM content, ranging from 190 to 250 g/kg sample,[13,15,20,51] which on average was higher than the STL DM of this study. The higher DM contents in STL from the previous authors were expected as those STL were collected from the factories, and by the time the samples were transported to a laboratory for DM determination, substantial amounts of moisture was perhaps evaporated or leached out of the samples. Conversely, the DM in STL of this study was immediately determined before any water evaporation took place in our laboratory. The previous authors had also reported slightly greater CP (276−311 g/kg DM) and lipid contents (57 g/kg DM) in green STL than this study,[20] which may be related to the higher OM content (970 g/kg DM) of material in their study. Conversely, the STL from this study had lower NDF but higher ADF contents than the green STL reported by Xu et al.[20] (410 and 261 g/kg DM, respectively) and Kondo et al.[13] (439 and 263 g/kg DM, respectively). Furthermore, previously reported (g/kg DM) TP (99−97), TT (89−85), and CT (23.7−96.5) values, respectively,[13,15,20] were lower than those in green STL of this study (130, 126, and 105, respectively). As found in this study, these differences could be attributed to the variation in tea-to-water ratios and other unknown processing methods that were used for the processing of tea leaves and extraction of tea drinks in different studies.[13,15,20,51]

Along with CP and ash, secondary metabolites such as TP, TT, CT, and TS were significantly increased as the tea-to-water ratios were increased, which could be linked with the significant decreases in the WHC resulting in more nutrient-rich STL. The CP, EE, and ash appeared to be less soluble than secondary metabolites in water because the concentrations of these two chemicals were less changed compared with the plant secondary metabolites in both green and black STL.

On the basis of this study, it appeared that the green and black tea leaves along with their STL had relatively high protein, fiber, plant secondary metabolites, and mineral components that can be useful as additives for ruminant diets. Information on the use of original tea leaves as a ruminant feed additive is still limited, perhaps due to the competition for their use for humans. However, the utilization of STL as potential sources of protein and fiber to feed ruminants has been suggested for years.[10−13,15−17,19,20,52] Some authors associated the presence of plant secondary metabolites such as tannins in STL as antinutrients that could reduce the solubility and rumen degradability of most plant proteins due to their ability to form undegradable protein complexes and hence reduced rumen ammonia production.[11,12] However, these protected proteins can be beneficial as bypass proteins along with the non-ammonia nitrogen supply to be absorbed in the small intestine of ruminant animals.[28,29,53−55] Although ammonia is an important source of N for rumen microbes, its overproduction or fast production may exceed the ability of microbes to utilize it. This can lead to an excessive NH_3 supply that after absorption through the rumen wall can enter the bloodstream and liver and eventually be excreted in urine as a waste.[56,57]

Babayemi et al.[52] estimated that rumen methane production from original tea leaves was lower than from their STL counterparts, and this was related to the higher secondary metabolites in tea leaves than in their STL. Hu et al.,[31] Mao et al.,[32] and Zhou et al.[58] reported that tea saponin extracts could reduce rumen methane production. Ishihara et al.[59] also reported that green tea extract could improve intestinal microflora balance and inhibit digestive and respiratory diseases in ruminants. Other studies reported that tannin extract supplementations into ruminant diets from either *Leucaena leucephala*,[60] *Acacia mearnsii*,[61] or *Lespedeza cuneata*[62,63] had the potential to reduce methane production. Similarly, it was reported that saponin extract addition into the diets from either *Achyranthus aspara*, *Tribulus terrestris*, *Albizia lebbeck*,[64] or *Gynostemma pentaphyllum*[65] could decrease methane releases from ruminants. Moreover, it was also found that tannin extract from *Pistachia lentiscus*, *Phillyrea latifolia*,[33] and *Havardia albicans*[34] could inhibit gastrointestinal nematodes in ruminants. In addition, Botura et al.[66] reported that saponin extract from *Agave sisalana* waste reduced total parasite egg counts in lamb feces without causing any toxicity as assessed by histological analysis of the liver and kidneys. Due to the potential advantageous effect of plant secondary metabolites such as tannins and saponins along with CP and other soluble nutrients in STL for ruminants, tea beverage industries may consider increasing tea-to-water ratios during their tea drink preparation to obtain a concentrated tea drink and consequently nutrient-rich STL but less ADF and NDF contents as found in this study. Reducing water during tea drink preparation can also be beneficial for tea beverage companies because there will be less requirement of space to store tea drink, less energy for heating smaller volumes during extraction, and less water containing STL with longer shelf life.

It has been reported that feeding STL or other tannin-rich plants has been associated with reduced feed intake due to their low palatability,[16,29,67] which may affect animal performance. This obstacle can be solved by mixing the STL with other palatable diets in the form of total mixed rations. Ensiling treatment can be a preferable option to improve the quality and to preserve the STL with its high water content. In ensiled total mixed rations for ruminants, green STL could substitute about 5% of soybean meal and alfalfa hay,[17] 10% of soybean meal and soybean hull,[18] 15% of brewer's grain,[20] and 20% of whole-crop oats[13] without affecting feed intake and animal productivity.

It can be concluded that tea leaves and their STL are good sources of protein, fiber, plant secondary metabolites, and minerals for their inclusion in ruminant diets. Because the concentration of CP and plant secondary metabolites can be enhanced in STL by increasing the tea-to-water ratio during the preparation of tea drinks, this approach may be adopted by the tea industry to obtain more nutrient-rich STL for their later use as feed additives for ruminant animals. Also, by using such increased tea-to-water ratios the tea beverage companies can produce less volume of more concentrated drinks, which will require less storage and heating and hence less overall cost of tea production. The presence of high levels of plant secondary metabolites in original tea leaves and their nutrient-rich STL suggests that they may have potential for use as a natural additive in ruminant diets. However, animal trials are needed to test the suitability of tea products for their use in formulating nutritious diets to improve ruminant health and vitality.

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Notes

The authors declare no competing financial interest.

■ ABBREVIATIONS USED

STL, spent tea leaves; DM, dry matter; OM, organic matter; EE, ether extract; CP, crude protein; NDF, neutral detergent fiber; ADF, acid detergent fiber; TP, total phenols; TT, total tannins; CT, condensed tannins; TS, total saponins; WHC, water-holding capacity

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