



Differences in the analysis of the quality indexes and characteristic amino acids of the different grades of Wuyi Shuixian (*Camellia sinensis*) tea

Qi ZHANG^{1#} , Ying ZHANG^{2#}, Jiayi XIE¹, Jianghua YE¹, Xiaomin PANG³, Xiaoli JIA^{4*}

Abstract

The objective evaluation of the quality of the Wuyi rock tea remains challenging. In this study, 649 samples comprising four grades of Wuyi Shuixian tea from the “Spring Tea Competition, 2019” were collected to determine their quality indexes. The results showed that the higher the grade of the Wuyi Shuixian tea, the higher the individual scores from the sensory evaluation. The water extracts, catechin, aspartic acid, and tyrosine were significantly different among the different tea grades. Restricted principal coordinate analysis showed that different grades of Wuyi Shuixian tea could be effectively differentiated using the tea quality indexes and 15 free amino acids. The total quality score of the sensory evaluation was significantly negatively correlated with the hydrophobic amino acids, bitter amino acids, and ECG, and was significantly positively correlated with GCG. These results suggest that the high content of the multicomponents is the material basis of the high quality of the tea. The hydrophobic amino acids, bitter amino acids, ECG, and GCG can be used to evaluate the differences in grades of the Wuyi Shuixian tea. The results of this study are expected to provide a theoretical basis for objectively evaluating the differences in the qualities of the Wuyi Shuixian tea.

Keywords: Wuyi Shuixian tea; tea grades; sensory evaluation; characteristic amino acid; quality index.

Practical Application: Four grades of Wuyi Shuixian teas could be effectively distinguished by the tea quality indexes.

1 Introduction

Wuyi rock tea is one of the top 10 famous teas in China, named for its origin in the rocky mountain area of Wuyishan in the Fujian Province of China. Wuyi Shuixian tea (*Camellia sinensis*) is one of the main varieties of Wuyi rock tea and is an excellent national tea tree, which is widely planted in Wuyishan City (Qiu et al., 2018). Wuyi Shuixian tea has a strong aroma and mellow taste and is favored by the majority of consumers for its unique sweet and mellow taste. It has long enjoyed the reputation of “rock bone fragrance” and has become the leader of Oolong teas. To improve the tea industry and product quality in Wuyishan City, the Wuyishan Tea Bureau holds the “Spring Tea Competition” every year. This is a big event in the tea industry in Wuyishan City, and also the most recognized tea competition in the Wuyishan tea industry. The competition set up gold medal, first prize, and high quality prize teas. The winners can gain the rich bonus products and honors as well as the affirmation of tea quality.

The taste of tea infusion is mainly the sensory comprehensive stimulation caused by the substances that dissolve in tea infusion, including tea polyphenols, caffeine, catechin, amino acids, and soluble sugar (Chen et al., 2008). Generally, there are six main tastes of tea infusion: sour, sweet, bitter, salty, astringent, and umami; among these, bitter, astringent, and umami are the main tastes of tea infusion (Tarachiwin et al., 2007). The substances

that affect the bitter and astringent tastes of tea soup include flavanols, flavanones, other polyphenols, and caffeine, which can enrich the flavor of tea within a certain content range (Li et al., 2013). Catechins are the major chemical components of tea and are the main bitter and astringent taste contributors to tea infusion (Narukawa et al., 2011; Zhuang et al., 2020; Li et al., 2021; Pang et al., 2022). Narukawa et al. (2011) studied the bitter intensity of catechins in tea and found that the order of bitter intensity of catechins was epigallocatechin gallate (EGCG), epicatechin gallate (ECG), epigallocatechin (EGC), gallic acid (GA), gallic acid gallate (GCG). Zhang et al. (2016) found that EGC and EC were the main contributors to the sweet aftertaste, but the astringency in the tea infusion inhibited the sweet aftertaste of EGC and EC. Amino acids are an important component of tea infusion taste and are usually used as an evaluation index for tea quality (Gu et al., 2012; Akitomi et al., 2013; Yu & Yang, 2020). Miyauchi et al. (2014) reported that glutamine, arginine, and theanine can be used as indicators to evaluate the quality of Tencha. Some studies have shown that amino acid content is significantly positively correlated with the quality of many tea types, such as oolong and green teas (Wang et al., 2010; Wang & Ruan, 2009; Xu et al., 2012).

Currently, tea grade evaluation mainly relies on sensory evaluation, but few studies involved the relationship between the

Received 01 June, 2022

Accepted 26 July, 2022

¹ College of Tea and Food Science, Wuyi University, Wuyishan, Fujian, China

² College of Horticulture, Fujian Agriculture and Forestry University, Fuzhou, China

³ Information Technology and Laboratory Management Center, Wuyi University, Wuyishan, Fujian, China

⁴ College of Ecology and Resource Engineering, Wuyi University, Wuyishan, Fujian, China

*Corresponding author: jiaxl2010@126.com

#Contributed equally to this study

quality index and the grade of tea. In this study, we collected four grades of Wuyi Shuixian tea from the 'Spring Tea Competition, 2019' to determine the quality indexes, and the relationship between the quality indexes and grades of Wuyi Shuixian tea was further analyzed.

2 Materials and methods

2.1 Tea samples

The tea samples comprised 67 gold medal (SA), 142 first prize (SB), 152 high-quality award (SC), and 288 non-awarding (SD) teas.

2.2 Sensory evaluation

The quality scores of the teas were blindly assessed according to a standardized procedure (Chinese National Standard, 2018) by a tasting panel consisting of five professional tasters officially certified. The evaluation procedure is described below. Then, 5 g tea was infused in a 110 mL tea-tasting porcelain cup with freshly boiled water and covered for 1 min then the tea aroma was assessed by smelling the flavor through the cup cover. The tea was later poured into a tea bowl for 2 min to assess the color, taste, and infused leaf aroma. Next, the cup was refilled with freshly boiled water and covered for 2 min for tea aroma assessment, and then the tea was poured into a tea bowl for 3 min to assess the color, taste, and infused leaf aroma again. Subsequently, the cup was refilled with freshly boiled water, covered for 3 min for tea aroma assessment, and then the tea was poured into a tea bowl for 5 min to assess the color, taste, and infused leaf aroma. The total score of each sample was calculated by summing the individual scores for dry tea appearance, aroma, taste, infused leaves, and tea color adjusted by weighting factors 20%, 30%, 35%, 10%, and 5% respectively.

2.3 Determination of tea quality index

The composition of the water extract was measured according to the National Standards of the People's Republic of China (Chinese National Standard, 2013a). This determination was done in triplicate for each sample. The composition of catechins and tea polyphenols was measured according to the National Standards of the People's Republic of China (Chinese National Standard, 2008). The detection wavelength was 278 nm. The mobile phase (A) was 0.09% acetonitrile and 0.02% acetic acid in water, and the mobile phase (B) was 0.8% acetonitrile and 0.02% acetic acid in water with the following gradient elution program: A:B = 100:0 (0-10 min), A:B = 68:32 (10-25 min), and A:B = 100:0 (25-30 min). The operating column temperature was maintained at 35 °C. The injection volume was 10 µL, and the flow rate was 1 mL/min. Pure EGCG, ECG, EGC, GC, and GCG obtained from Sigma (USA) were used as standards for detection. The content of tea polyphenols was determined using the Folin-Ciocalteu phenol reagents. Gallic acid was used as a standard and detected at 765 nm. All measurements were performed in triplicate.

The total free amino acid content (Chinese National Standard, 2013c) was determined using the ninhydrin reagents. The caffeine

content was determined using ultraviolet spectrophotometry. Caffeine (Chinese National Standard, 2013b) was used as a standard and detected at 274 nm. The anthrone colorimetric method was used to assay soluble sugar (Wang, 2006) and the absorbance was recorded using a spectrophotometer at a wavelength of 620 nm. The composition of free amino acids was measured according to the National Standards of the People's Republic of China (Chinese National Standard, 2016a) using a Hitachi L-8900 automatic amino acid analyzer and external standards of mixed amino acids as calibrators. All measurements were performed in triplicate.

The composition of total flavonoids was measured according to the Industrial Standards for Entry-Exit Inspection and Quarantine of the People's Republic of China (Chinese National Standard, 2016b). The aluminum chloride-acetic acid-sodium acetate colorimetric method was used to assay flavonoids, and the absorbance was recorded using a spectrophotometer at a wavelength of 415 nm. All measurements were performed in triplicate.

2.4 Statistical analysis

All experimental data were presented as mean ± standard error (SE). They were analyzed using a one-way analysis of variance (ANOVA), and followed by the least significant difference (LSD). Restricted principal coordinate analysis (PCoA) and correlative analysis were all conducted with SPSS 20.0 software.

3 Results and discussion

3.1 Sensory evaluation

At present, sensory evaluation is the most important method used to judge differences in tea quality. The better the tea quality, the higher the sensory evaluation score. In China, the national standard for evaluating oolong tea is 100% in total, comprising 20% for the appearance of dry tea, 30% for tea aroma, 35% for taste, 10% for infused leaves, and 5% for tea color (Chinese National Standard, 2018). The sensory evaluation results of the different grades of Wuyi Shuixian tea showed that (Table 1) the sensory evaluation scores of the gold medal, first prize, high-quality award, and non-awarding grades of Wuyi Shuixian tea were all higher than 92.50, 86.00, 80.00, and 60.00, with average scores of 93.40, 88.38, 82.54, and 72.28, respectively.

3.2 Quality index of Wuyi Shuixian tea

Water extract is the main flavor and color substance of tea soup, which is highly related to the taste of tea infusion. The results showed that the water extract in the Wuyi Shuixian tea was in the range of 35.97-43.23% (Figure 1). It was the highest in the SA grade (43.23%) and was significantly different ($P < 0.05$) from those in the SC and SD grades. The water extracts for the different grades were SB (39.22%) > SD (38.50%) > SC (35.97%); however, there were no significant differences among them.

The soluble sugar content of Wuyi Shuixian tea was in the range of 46.76-48.08 (mg/g). The soluble sugar content for each grade was SC (48.08 mg/g) ≈ SB (48.00 mg/g) > SA (47.21 mg/g) > SD (46.76 mg/g); however, there were no significant differences

Table 1. Sensory evaluations results of Wuyi Shuixian tea with different grades.

Grade	Appearance (20%)	Aroma (30%)	Taste (35%)	Infused leaves (10%)	Liquor color (5%)	TQS
SA	94.62 ± 3.47	92.35 ± 2.41	93.49 ± 2.22	93.47 ± 3.24	94.01 ± 3.85	93.40 ± 3.81
SB	87.66 ± 2.42	88.08 ± 4.35	89.64 ± 3.20	86.35 ± 3.41	88.32 ± 4.62	88.38 ± 3.25
SC	81.98 ± 3.64	85.17 ± 5.20	84.28 ± 2.92	80.18 ± 2.04	82.54 ± 1.93	82.54 ± 5.48
SD	72.55 ± 2.03	70.62 ± 3.07	70.57 ± 3.52	63.15 ± 5.40	68.44 ± 2.25	72.28 ± 4.62

Means standard error (\pm SE) were score of the six panelists independently. SA: gold medal Wuyi Shuixian tea; SB: first prize Wuyi Shuixian tea; SC: high quality award Wuyi Shuixian tea; SD: non-awarding Wuyi Shuixian tea; TQS: total quality score.

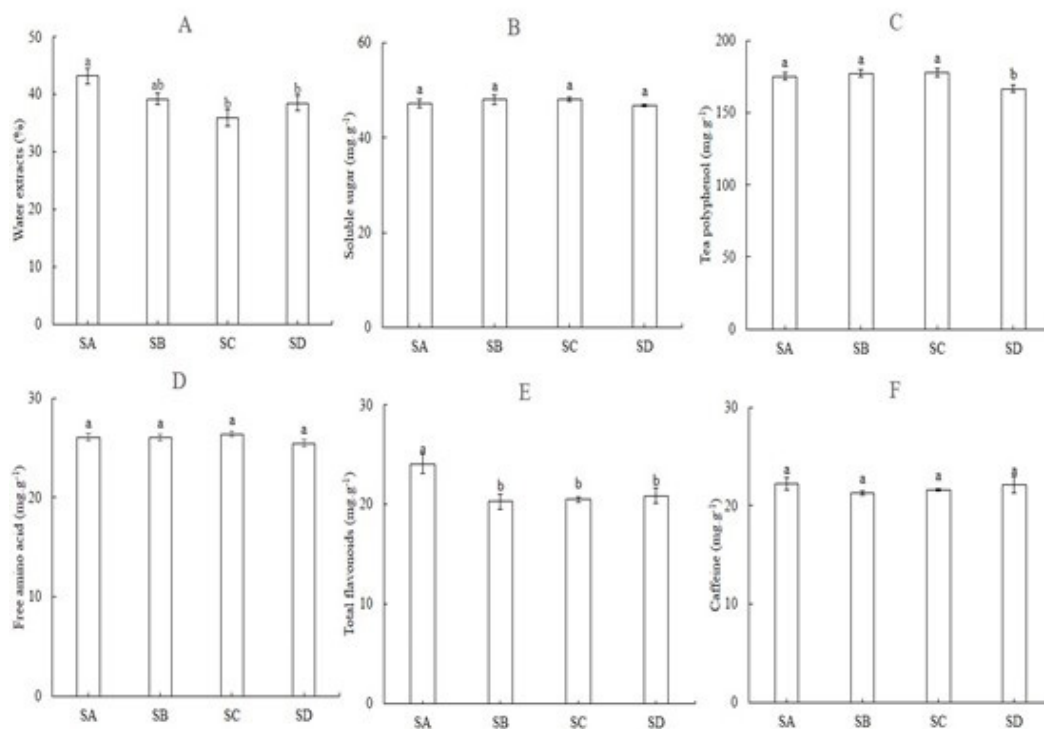


Figure 1. The quality indexes in different grades of Wuyi Shuixian tea. SA: gold medal Wuyi Shuixian tea; SB: first prize Wuyi Shuixian tea; SC: high quality award Wuyi Shuixian tea; SD: non-awarding Wuyi Shuixian tea. The lowercase letters represent a significant level at $P < 0.05$.

($P < 0.05$) among them. Although the soluble sugar content in the tea infusion was very low, it could reduce the bitterness of the tea infusion. Yin et al. (2014) showed that there was a synergistic effect of sweet-tasting between theanine and sucrose. The higher the soluble sugar content in tea infusion, the more the bitterness of tea infusion can be reduced.

Tea polyphenols are the most abundant inclusions in tea and are important flavor substances (Li et al., 2013). Tea polyphenols are related to the astringent and mellow flavors of tea infusion (Zhang, 2014). The content of tea polyphenols in Wuyi Shuixian tea was in the range of 166.67–177.99 mg/g. It was the lowest in the SD grade (166.67 mg/g), which was significantly different ($P < 0.05$) from the other grades. The content of tea polyphenols for the different grades was SA (175.37 mg/g) < SB (177.53 mg/g) < SC (177.99 mg/g); however, there were no significant differences ($P < 0.05$) among them.

Free amino acids are an important factor in the freshness of tea infusion (Xu et al., 2020). The high content of free amino

acids in tea leaves can increase the freshness of tea infusion (Chen & Yang, 2015). The total free amino acid content of Wuyi Shuixian tea was in the range of 25.45–26.40 mg/g. It was not significantly different ($P < 0.05$) among the four grades of tea samples. The ratio of phenol to amino acids of the SA, SB, SC, and SD grades was 6.72, 6.81, 6.74, and 6.55, respectively. The ratio of phenol to amino acids is a traditional quality evaluation index of tea (Liu et al., 2021). Generally, a high phenol to ammonia ratio contributes to tea infusion with a strong throat feeling, and low phenol to ammonia ratio contributes to tea infusion with a soft throat feeling. This study shows that the ratio of phenol to amino acids of the three winning teas were higher than those of the non-winning teas.

Flavonoids are a kind of tea polyphenols, which have a low content in tea infusion but have a great contribution to the taste of tea infusion due to their low taste threshold (Scharbert & Hofmann, 2005). The flavonoid content of Wuyi Shuixian tea was in the range of 20.29–24.09 mg/g. It was the highest in

the SA grade (24.09 mg/g), which was significantly different ($P < 0.05$) from those of the other grades. The flavonoid content of the different grades was SD (20.87 mg/g) > SC (20.54 mg/g) > SB (20.29 mg/g); however, there were no significant differences ($P < 0.05$) among them.

The caffeine content of Wuyi Shuixian tea was in the range of 21.26–22.28 mg/g. There were no significant differences ($P < 0.05$) among the four grades of tea samples. Studies have shown that caffeine is the main taste substance of tea soup, which is bitter in tea soup and is negatively correlated with the quality of tea (Xu et al., 2020; Scharbert & Hofmann, 2005). However, the appropriate amounts of caffeine can enhance the taste of tea infusion (Li et al., 2018).

3.3 Catechin components of the different grades of tea

Catechin is one of the main flavor compounds of tea polyphenols in tea infusion. The results of the catechin components of Wuyi Shuixian tea showed that (Figure 2), the EGCG content of Wuyi Shuixian tea was in the range of 30.42–32.18 mg/g. It was the lowest in SA (30.42 mg/g), which was significantly different ($P < 0.05$) from those of the other grades. The EGCG content of the different grades was SB (31.41 mg/g) < SD (31.85 mg/g) < SC (32.18 mg/g), and that of SC was significantly ($P < 0.05$) higher than that of SB. The ECG content was SD (9.48 mg/g) \approx SC (9.38 mg/g), which was significantly ($P < 0.05$) higher than that of SB (9.20 mg/g), and SB (9.20 mg/g) was significantly

($P < 0.05$) higher than that of SA (8.76 mg/g). The EGC content of SC (17.26 mg/g) was the highest, which was significantly different ($P < 0.05$) from those of the other grades. The EGC content of the different grades was SB (16.86 mg/g) > SD (16.66 mg/g) > SA (16.34 mg/g), and that of SB was significantly ($P < 0.05$) higher than that of SA. The GC content of SA (10.06 mg/g) was the lowest, which was significantly different ($P < 0.05$) from those of the other grades. The GC content was SC (10.69 mg/g) \approx SB (10.68 mg/g) > SD (10.42 mg/g); however, there were no significant differences ($P < 0.05$) among them. The GCG content of SD (1.09 mg/g) was the lowest, which was significantly different ($P < 0.05$) from those of SA and SB. The GCG content was SA (1.20 mg/g) \approx SB (1.19 mg/g) > SC (1.16 mg/g); however, there were no significant differences among them. The total catechin content of the different grades was SA (66.78 mg/g) < SB (69.34 mg/g) \approx SD (69.50 mg/g) < SC (70.68 mg/g).

EGCG was the highest of the five catechin components accounting for approximately 45%. The second was EGC and GC, accounting for approximately 24% and 15%, respectively. The third was ECG, accounting for approximately 13%. The lowest was GCG, accounting for approximately 1.0%. More specifically, the ester-type catechins (namely EGCG, ECG, and GCG) of the four grades of tea accounted for 60–61%, and the non-ester-type catechins (namely GC and EGC) accounted for 38–39%. The ratio of the ester-type to non-ester-type catechins was 1.53, 1.52, 1.53, and 1.57 in SA, SB, SC, and SD, respectively (Table 2). The bitterness of tea infusion mainly comes from catechin in

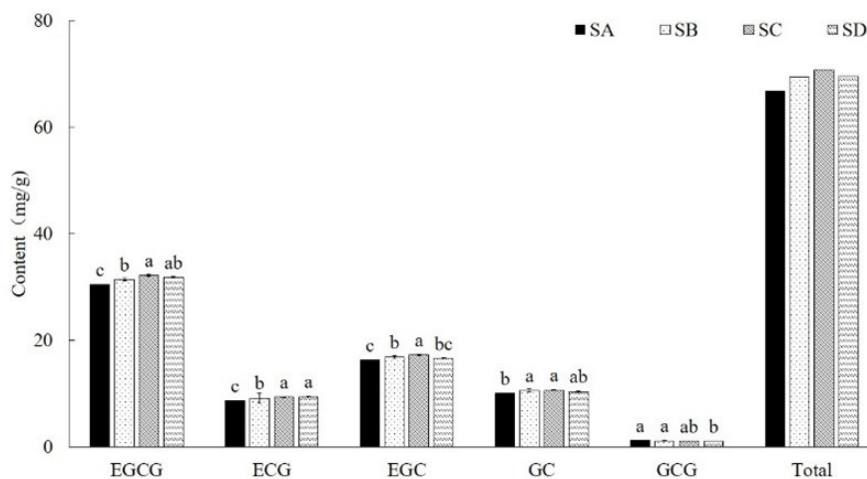


Figure 2. Catechins content of Wuyi Shuixian tea with different grades. SA: gold medal Wuyi Shuixian tea; SB: first prize Wuyi Shuixian tea; SC: high quality award Wuyi Shuixian tea; SD: non-awarding Wuyi Shuixian tea. The lowercase letters represent a significant level at $P < 0.05$.

Table 2. The proportion of different types of catechins in Wuyi Shuixian tea with different grades.

Composition	SA	SB	SC	SD
Ester catechins	60.47%	60.28%	60.45%	61.04%
Non-ester catechins	39.53%	39.72%	39.55%	38.96%
Ratio of ester to non-ester catechins	1.53	1.52	1.53	1.57

SA: gold medal Wuyi Shuixian tea; SB: first prize Wuyi Shuixian tea; SC: high quality award Wuyi Shuixian tea; SD: non-awarding Wuyi Shuixian tea. Ester catechins include GCG, ECG, and EGCG; non-ester catechins include GC and EGC.

polyphenols, but high catechin increases the bitterness of tea infusion (Narukawa et al., 2011). Previous studies have shown that tea infusion tastes mellower and contains less galloylated catechin (Hofmann et al., 2006; Ding et al., 1992). Our results show that the content of catechins in the high-grade tea samples was lower, the taste of tea infusion was softer, and the ratio of the ester-type to non-ester-type catechins was lower, which reduced the bitterness of the ester-type catechins.

3.4 Characteristic amino acid profiles in the different grades of tea

Free amino acids in tea leave important chemical components that affect the quality of tea (Ruan et al., 1998). Amino acids contribute to the overall quality in terms of freshness and mellowness of tea (Wang & Ruan, 2009). Although there were no significant differences in the total free amino acid contents among the four tea samples (Figure 1), the composition of amino acids in each of the four tea samples was further determined, and 15 types of amino acids were identified in the tea infusion (Table 3). The total contents of the 15 amino acids were SD (3.9468 mg/g) > SC (3.9238 mg/g) > SA (3.7708 mg/g) > SB (3.7017 mg/g).

Theanine is the amino acid with the highest component in tea infusion, and its flavor is umami and sweet (Scharbert et al., 2004). The correlation coefficient between theanine and tea quality was as high as 0.989 (Casimir et al., 1960). Theanine accounted for approximately 42-47% of the total contents of free amino acids, and theanine in the different grades was SC (1.7670 mg/g) > SA (1.7631 mg/g) > SB (1.7304 mg/g) > SD (1.6940 mg/g). The SD theanine content was significantly ($P < 0.05$) lower than those of SA, SB, and SC, and there were no significant differences among SA, SB, and SC. The aspartic acid and tyrosine contents were SD > SC > SA > SB, and there were no significant differences among the four grades of tea samples. The threonine content was SD (0.1131 mg/g) > SB (0.1079 mg/g) > SC (0.1022 mg/g) > SA (0.0893 mg/g), and that of SA was significantly ($P < 0.05$) lower than those of SB, SC, and SD; there were no significant differences among SD, SB, and SC. Leucine content was the highest in SC (0.0562 mg/g) and SD (0.0590 mg/g), and was significantly different ($P < 0.05$) from those of SA (0.0443 mg/g) and SB (0.0441 mg/g). The third component was glutamic acid, which accounted for approximately 8-9% of the total contents, but as with the other eight amino acids, there were no significant differences among the four tea samples (Table 3).

Table 3. The content of amino acids components in Wuyi Shuixian tea with different grades (mg/g).

Amino acid	SA	SB	SC	SD
Theanine	1.7631 ± 0.0201 ^a (46.76%)	1.7304 ± 0.0160 ^a (46.75%)	1.7670 ± 0.0205 ^a (45.21%)	1.6940 ± 0.0060 ^b (42.92%)
Aspartic acid	0.4443 ± 0.0026 ^c (11.78%)	0.4195 ± 0.0037 ^d (11.33%)	0.4659 ± 0.0090 ^b (11.87%)	0.4946 ± 0.0042 ^a (12.53%)
Threonine	0.0893 ± 0.0015 ^b (2.37%)	0.1079 ± 0.0032 ^a (2.92%)	0.1022 ± 0.0041 ^a (2.60%)	0.1131 ± 0.0033 ^a (2.87%)
Tyrosine	0.0924 ± 0.0019 ^c (2.45%)	0.0841 ± 0.0033 ^d (2.27%)	0.1044 ± 0.0029 ^b (2.66%)	0.1123 ± 0.0013 ^a (2.85%)
Leucine	0.0443 ± 0.0051 ^b (1.18%)	0.0441 ± 0.0096 ^b (1.19%)	0.0562 ± 0.0005 ^a (1.43%)	0.0590 ± 0.0008 ^a (1.50%)
Serine	0.1690 ± 0.0017 ^a (4.48%)	0.1696 ± 0.0055 ^a (4.58%)	0.1789 ± 0.0069 ^a (4.56%)	0.1776 ± 0.0053 ^a (4.50%)
Glutamic acid	0.3213 ± 0.0286 ^a (8.52%)	0.3248 ± 0.0054 ^a (8.77%)	0.3451 ± 0.0276 ^a (8.79%)	0.3516 ± 0.0067 ^a (8.91%)
Glycine	0.0216 ± 0.0129 ^a (0.57%)	0.0216 ± 0.0129 ^a (0.58%)	0.0207 ± 0.0109 ^a (0.53%)	0.0249 ± 0.0146 ^a (0.63%)
Lysine	0.0187 ± 0.0048 ^a (0.50%)	0.0184 ± 0.0045 ^a (0.50%)	0.0183 ± 0.0044 ^a (0.47%)	0.0201 ± 0.0098 ^a (0.51%)
Alanine	0.1533 ± 0.01 ^a (4.06%)	0.1461 ± 0.01 ^a (3.95%)	0.1937 ± 0.02 ^a (4.94%)	0.1598 ± 0.01 ^a (4.05%)
Histidine	0.1797 ± 0.03 ^a (4.77%)	0.1719 ± 0.03 ^a (4.64%)	0.1936 ± 0.01 ^a (4.93%)	0.2203 ± 0.01 ^a (5.58%)
Phenylalanine	0.1325 ± 0.0032 ^a (3.52%)	0.1342 ± 0.0068 ^a (3.63%)	0.1365 ± 0.0083 ^a (3.48%)	0.1435 ± 0.0047 ^a (3.64%)
Valine	0.1597 ± 0.0004 ^a (4.24%)	0.1583 ± 0.0021 ^a (4.28%)	0.1665 ± 0.0012 ^a (4.24%)	0.1766 ± 0.0018 ^a (4.47%)
Isoleucine	0.1052 ± 0.0040 ^a (2.79%)	0.1000 ± 0.0008 ^a (2.70%)	0.0989 ± 0.0045 ^a (2.52%)	0.1044 ± 0.0035 ^a (2.65%)
γ-Aminobutyric acid	0.0763 ± 0.0037 ^a (2.02%)	0.0707 ± 0.0011 ^a (1.91%)	0.0759 ± 0.0006 ^a (1.94%)	0.0948 ± 0.0022 ^a (2.40%)
Total	3.7708	3.7017	3.9238	3.9468

SA: gold medal Wuyi Shuixian tea; SB: first prize Wuyi Shuixian tea; SC: high quality award Wuyi Shuixian tea; SD: non-awarding Wuyi Shuixian tea. Means standard error (± SE) from three replications for each determination is shown. The lowercase letters represent a significant level at $P < 0.05$.

The amino acids in tea infusion do not all have the umami taste, but they also have bitter, astringent, fresh, sweet, and sour tastes (Yu & Yang, 2020). Research has shown that hydrophobic amino acids have a bitter taste, while some hydrophilic amino acids, due to their small molecular chains, readily bind to taste receptors to produce a sweet taste (Akitomi et al., 2013). An in-depth analysis of these special characteristics of amino acids of Wuyi Shuixian tea revealed that in these four tea samples (Table 4), the hydrophilic amino acids accounted for 87.74-88.32% and the hydrophobic amino acids accounted for 11.68-12.26%. The ratio of hydrophilic to hydrophobic amino acids was SC (7.56) > SA (7.53) > SB (7.48) > SD (7.16). The sweet amino acids accounted for 78.8-80.79% and the bitter amino acids accounted for 19.21-21.19% of the amino acid content. The ratio of sweet to bitter amino acids was SB (4.21) > SA (4.15) > SC (4.07) > SD (3.72). The umami amino acids accounted for 64.36-67.06%: SA (67.06%) > SB (66.85%) > SC (65.70%) > SD (64.36%). Although the trends of most of these characteristic amino acids were not completely consistent with the tea grades, the trend of the umami amino acid content was exactly consistent with the tea grades. L-Theanine contributes to the umami taste in tea infusion and is closely related to the quality of green tea (Juneja et al., 1999; Yamaguchi & Ninomiya, 2000). The proportion of theanine that accounted for the total content was 46.76%, 46.75%, 45.21%, and 42.92% in SA, SB, SC, and SD, respectively (Table 3). We suggest

that the proportion of umami amino acid content could be used to evaluate the quality of Wuyi Shuixian tea.

3.5 Restricted principal coordinate analysis

Sensory evaluation grade is more reflected in the color, aroma, taste, and shape of tea, and the determination of the quality indexes may explain more problems. The changes in the quality indexes may indicate the grade of Wuyi Shuixian tea. Thus, the restriction principal coordinate analysis (PCoA) of the quality indexes of Wuyi Shuixian tea was performed (Figure 3A). The coordinate axis 1 (PCoA1) could differentiate SA, SB, and SC from SD, which explained 79.04% of the difference. Differences among RA, RB, and RC were dominated by principal coordinate 2 (PCoA2), and the degree of explanation was 11.11%. The total degree of explanation of the quality indexes to the differences among the Wuyi Shuixian tea grades reached 73.46%. The PCoA of the 15 free amino acids in Wuyi Shuixian tea could differentiate Wuyi Shuixian tea into different grades (Figure 3B). The coordinate axis 1 (PCoA1) could differentiate SA and SB from SC and SD, which explained 74.82% of the difference. The coordinate axis 2 (PCoA2) could differentiate SA from SB and SC from SD. The total degree of explanation of the 15 free amino acids to the differences among the Wuyi Shuixian tea grades reached 58.02%.

Table 4. The characteristics of amino acids in Wuyi Shuixian tea with different grades.

Characteristic	SA	SB	SC	SD
Hydrophilic (%)	88.28	88.21	88.32	87.74
Hydrophobic (%)	11.72	11.79	11.68	12.26
Ratio of Hydrophilic: Hydrophobic	7.53	7.48	7.56	7.16
Sweet (%)	80.57	80.79	80.26	78.81
Bitter (%)	19.43	19.21	19.74	21.19
Ratio of Sweet: Bitter	4.15	4.21	4.07	3.72
Umami (%)	67.06	66.85	65.70	64.36

SA: gold medal Wuyi Shuixian tea; SB: first prize Wuyi Shuixian tea; SC: high quality award Wuyi Shuixian tea; SD: non-awarding Wuyi Shuixian tea. Hydrophilic amino acids include glycine, serine, threonine, tyrosine, aspartic acid, glutamic acid, histidine, γ -Aminobutyric acid, alanine, lysine and theanine. Hydrophobic amino acids include valine, isoleucine, leucine, and phenylalanine. Sweet amino acids include glutamic acid, glycine, alanine, asparagine, threonine, serine, γ -Aminobutyric acid, and theanine. Bitter amino acids include histidine, phenylalanine, valine, isoleucine, leucine, lysine and tyrosine. Umami amino acids include aspartic acid, glutamic acid and theanine.

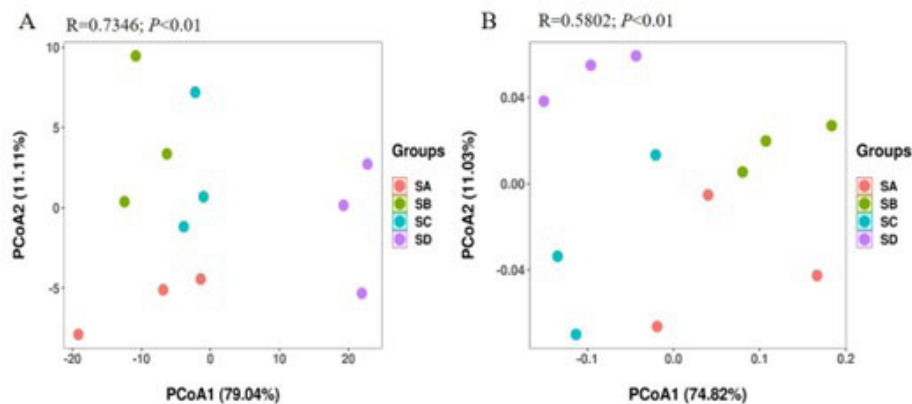


Figure 3. Restricted principal coordinate analysis (PCoA) of quality indexes of Wuyi Shuixian tea (A), of 15 free amino acid contents of Wuyi Shuixian tea (B). SA: gold medal Wuyi Shuixian tea; SB: first prize Wuyi Shuixian tea; SC: high quality award Wuyi Shuixian tea; SD: non-awarding Wuyi Shuixian tea.

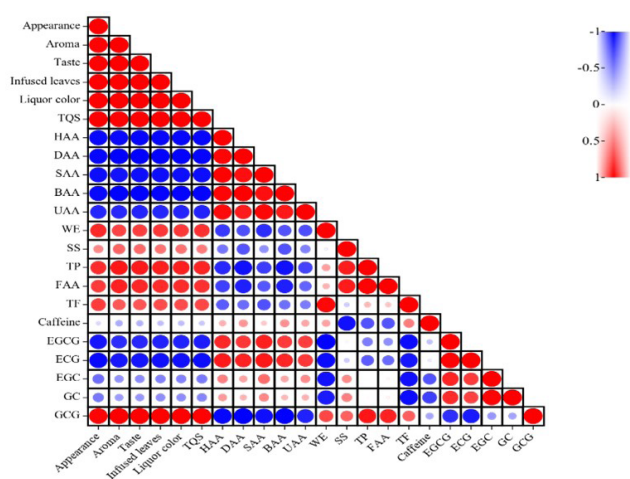


Figure 4. Correlation between quality indexes, characteristic amino acids and the score of sensory evaluation. TQS: total quality score; HAA: hydrophilic amino acids; DAA: hydrophobic amino acids; SAA: sweet amino acids; BAA: bitter amino acids; UAA: umami amino acids; WE: water extract; SS: soluble sugar; TP: tea polyphenol; FAA: free amino acid; TF: total flavonoid.

3.6 Correlation analysis

Correlation analysis was conducted to investigate the relationship between the quality indexes, characteristic amino acids, and the score of sensory evaluation. As shown in Figure 4, five quality features and the total quality score of Wuyi Shuixian tea were significantly negatively correlated with the hydrophobic and bitter amino acids and were significantly positively correlated with GCG ($P < 0.05$ or $P < 0.01$). ECGs were significantly negatively correlated with appearance and total quality score ($P < 0.05$). Therefore, the lower the contents of hydrophobic amino acids, bitter amino acids, and ECG, the higher the content of GCG, which may end up benefiting the good teas.

4 Conclusion

In this study, we conducted a detailed analysis of the quality indexes and characteristic amino acids of the different grades of Wuyi Shuixian tea and their correlation with the sensory evaluation. We found that the scores of sensory evaluation were significantly negatively correlated with the hydrophobic amino acids, bitter amino acids, and ECG, and were significantly positively correlated with GCG ($P < 0.01$ or $P < 0.05$). The different grades of Wuyi Shuixian tea could be effectively differentiated using the characteristic amino acids of tea infusion. Accordingly, we suggest that the hydrophobic amino acids, bitter amino acids, and catechin can be used to evaluate the differences in the grades of Wuyi Shuixian tea, and the ratio of umami amino acids is an important hallmark of the top of Wuyi Shuixian tea.

Abbreviations

TQS: total quality score. HAA: hydrophilic amino acids. DAA: hydrophobic amino acids. SAA: sweet amino acids. BAA: bitter amino acids. UAA: umami amino acids. WE: water extract. SS: soluble sugar. TP: tea polyphenol. FAA: free amino acid. TF: total flavonoid. GC: gallic acid. EGC: epigallocatechin.

GCG: gallic acid gallate. ECG: epigallocatechin gallate. EGCG: epigallocatechin gallate.

Acknowledgements

This work was supported by Natural Science Foundation of Fujian Province (grant numbers 2017J01649, 2020J05214), Scientific Research Project of Introducing Talents, Wuyi University (grant numbers YJ201909), Fujian Provincial First-class Undergraduate Major (Tea Science) Construction (grant numbers SJZY2019004), Teachers and students create team projects of Wuyi University (grant numbers 2021-SSTD-05, 2021-SSTD-01) and Fujian Province Young and Middle-aged Teacher Education Research Project (Science and Technology) (grant numbers JAT190786, JAT200664).

References

- Akitomi, H., Tahara, Y., Yasuura, M., Kobayashi, Y., Ikezaki, H., & Toko, K. (2013). Quantification of tastes of amino acids using taste sensors. *Sensors and Actuators B: Chemical*, 179, 276-281. <http://dx.doi.org/10.1016/j.snb.2012.09.014>.
- Casimir, J., Jadot, J., & Renard, M. (1960). Separation and characterization of N-ethyl-gamma-glutamine from *Xerocomus badius*. *Biochimica et Biophysica Acta*, 39(3), 462-468. [http://dx.doi.org/10.1016/0006-3002\(60\)90199-2](http://dx.doi.org/10.1016/0006-3002(60)90199-2). PMID:13808157.
- Chen, H. K., & Yang, J. F. (2015). Analysis of the chemical component on quality of Rougui tea in different rock areas. *Shipin Anquan Zhiliang Jiance Xuebao*, 6(4), 1287-1294.
- Chen, H., Zhang, M., Qu, Z., & Xie, B. (2008). Antioxidant activities of different fractions of polysaccharide conjugates from green tea (*Camellia Sinensis*). *Food Chemistry*, 106(2), 559-563. <http://dx.doi.org/10.1016/j.foodchem.2007.06.040>.
- Chinese National Standard. (2008). *GB/T 8313-2008: determination of total polyphenols and catechins content in tea*. Beijing: China Standard Publishing House.
- Chinese National Standard. (2013a). *GB/T 8305-2013: determination of tea extract*. Beijing: China Standard Publishing House.
- Chinese National Standard. (2013b). *GB/T 8312-2013: determination of tea caffeine*. Beijing: China Standard Publishing House.
- Chinese National Standard. (2013c). *GB/T 8314-2013: determination of total free amino acids in tea*. Beijing: China Standard Publishing House.
- Chinese National Standard. (2016a). *GB/T 5009.124-2016: determination of amino acids in foods*. Beijing: China Standard Publishing House.
- Chinese National Standard. (2016b). *SN/T 4592-2016: determination of total flavonoids in food for export*. Beijing: China Standard Publishing House.
- Chinese National Standard. (2018). *GB/T 23776-2018: methodology of sensory evaluation of tea*. Beijing: China Standard Publishing House.
- Ding, Z., Kuhr, S., & Engelhardt, U. H. (1992). Influence of catechins and theaflavins on the astringent taste of black tea brews. *Zeitschrift für Lebensmittel-Untersuchung und -Forschung*, 195(2), 108-111. <http://dx.doi.org/10.1007/BF01201768>.
- Gu, Q., Lu, J. S., & Ye, B. C. (2012). *Tea chemistry*. Anhui: Press of University of Science and Technology of China.
- Hofmann, T., Scharbert, S., & Stark, T. (2006). Molecular and gustatory characterization of the impact taste compounds in black tea infusions. *Developments in Food Science*, 43, 3-8. [http://dx.doi.org/10.1016/S0167-4501\(06\)80002-6](http://dx.doi.org/10.1016/S0167-4501(06)80002-6).

- Juneja, L. R., Chu, D. C., Okubo, T., Nagato, Y., & Yokogoshi, H. (1999). L-theanine—a unique amino acid of green tea and its relaxation effect in humans. *Trends in Food Science & Technology*, 10(6-7), 199-204. [http://dx.doi.org/10.1016/S0924-2244\(99\)00044-8](http://dx.doi.org/10.1016/S0924-2244(99)00044-8).
- Li, S. H., Lian, X. M., Wang, F. Q., Chen, R. B., & Hong, D. U. (2018). Analysis of flavor components of Wuyi Rock tea. *Journal of Fuyang Normal University*, 35(1), 33-42.
- Li, S., Lo, C., Pan, M., Lai, C., & Ho, C. (2013). Black tea: chemical analysis and stability. *Food & Function*, 4(1), 10-18. <http://dx.doi.org/10.1039/C2FO30093A>. PMID:23037977.
- Li, T., Xu, S., Wang, Y., Wei, Y., Shi, L., Xiao, Z., Liu, Z., Deng, W.-W., & Ning, J. (2021). Quality chemical analysis of crush-tear-curl (CTC) black tea from different geographical regions based on UHPLC-Orbitrap-MS. *Journal of Food Science*, 86(9), 3909-3925. <http://dx.doi.org/10.1111/1750-3841.15871>. PMID:34390261.
- Liu, S. B., Xiang, X., Li, L. L., Qu, Q. Y., Xu, Y. Y., Wang, J. G., Li, S., Xiao, W. J., & Gong, Z. H. (2021). Effect of raw material combination of fresh leaves of different tea varieties on the quality of processed blacktea. *Journal of Tea Communication*, 48(2), 259-265.
- Miyauchi, S., Yuki, T., Fuji, H., Kojima, K., Yonetani, T., Tomio, A., Bamba, T., & Fukusaki, E. (2014). High-quality green tea leaf production by artificial cultivation under growth chamber conditions considering amino acids profile. *Journal of Bioscience and Bioengineering*, 118(6), 710-715. <http://dx.doi.org/10.1016/j.jbiosc.2014.05.008>. PMID:24915994.
- Narukawa, M., Noga, C., Ueno, Y., Sato, T., Misaka, T., & Watanabe, T. (2011). Evaluation of the bitterness of green tea catechins by a cell-based assay with the human bitter taste receptor hTAS2R39. *Biochemical and Biophysical Research Communications*, 405(4), 620-625. <http://dx.doi.org/10.1016/j.bbrc.2011.01.079>. PMID:21272567.
- Pang, X. M., Chen, F. Y., Liu, G. Y., Zhang, Q., Ye, J. H., Lei, W. X., Jia, X. L., & He, H. B. (2022). Comparative analysis on the quality of Wuyi Rougui (*Camellia sinensis*) tea with different grades. *Food Science and Technology*, 42, e115321. <http://dx.doi.org/10.1590/fst.115321>.
- Qiu, X. H., Chen, S. R., Zhang, Q., Gao, C., Zhao, F., & Ye, N. X. (2018). Quality and chemical characteristic of Wuyi Shuixian tea. *Acta Tea Sinica*, 59(1), 47-52.
- Ruan, J. Y., Wu, X., Ye, Y., & Hardter, R. (1998). Effect of potassium, magnesium and sulphur applied in different forms of fertilisers on free amino acid content in leaves of tea (*Camellia sinensis* L.). *Journal of the Science of Food and Agriculture*, 76(3), 389-396. [http://dx.doi.org/10.1002/\(SICI\)1097-0010\(199803\)76:3<389::AID-JSFA963>3.0.CO;2-X](http://dx.doi.org/10.1002/(SICI)1097-0010(199803)76:3<389::AID-JSFA963>3.0.CO;2-X).
- Scharbert, S., & Hofmann, T. (2005). Molecular definition of black tea taste by means of quantitative studies, taste reconstitution, and omission experiment. *Journal of Agricultural and Food Chemistry*, 53(13), 5377-5384. <http://dx.doi.org/10.1021/jf050294d>. PMID:15969522.
- Scharbert, S., Holzmann, N., & Hofmann, T. (2004). Identification of the astringent taste compounds in black tea infusions by combining instrumental analysis and human bioresponse. *Journal of Agricultural and Food Chemistry*, 52(11), 3498-3508. <http://dx.doi.org/10.1021/jf049802u>. PMID:15161222.
- Tarachiwin, L., Ute, K., Kobayashi, A., & Fukusaki, E. (2007). ¹H-NMR based metabolic profiling in the evaluation of Japanese green tea quality. *Journal of Agricultural and Food Chemistry*, 55(23), 9330-9336. <http://dx.doi.org/10.1021/jf071956x>. PMID:17944534.
- Wang, K., & Ruan, J. Y. (2009). Analysis of chemical components in green tea in relation with perceived quality, a case study with Longjing teas. *International Journal of Food Science & Technology*, 44(12), 2476-2484. <http://dx.doi.org/10.1111/j.1365-2621.2009.02040.x>.
- Wang, K., Liu, F., Liu, Z., Huang, J., Xu, Z., Li, Y., Chen, J., Gong, Y., & Yang, X. (2010). Analysis of chemical components in oolong tea in relation to perceived quality. *International Journal of Food Science & Technology*, 45(5), 913-920. <http://dx.doi.org/10.1111/j.1365-2621.2010.02224.x>.
- Wang, X. K. (2006). *Principles and techniques of plant physiological and biochemical experiments*. Beijing: Higher Education Press.
- Xu, W., Song, Q., Li, D., & Wan, X. (2012). Discrimination of the production season of Chinese green tea by chemical analysis in combination with supervised pattern recognition. *Journal of Agricultural and Food Chemistry*, 60(28), 7064-7070. <http://dx.doi.org/10.1021/jf301340z>. PMID:22720840.
- Xu, X. Y., Chen, S., Yu, X. M., Zhao, X. M., Lin, H. Z., Liu, G. Y., Su, F., Gao, F., Sun, Y., & Hao, Z. L. (2020). Quality differences of different grades of Wuyi Rougui tea with different baking degrees. *Shipin Kexue*, 41(13), 22-28.
- Yamaguchi, S., & Ninomiya, K. (2000). Umami and food palatability. *The Journal of Nutrition*, 130(4), 921S-926S. <http://dx.doi.org/10.1093/jn/130.4.921S>. PMID:10736353.
- Yin, J. F., Zhang, Y. N., Du, Q. Z., Chen, J. X., Yuan, H. B., & Xu, Y. Q. (2014). Effect of Ca²⁺ concentration on the tastes from the main chemicals in green tea infusions. *Food Research International*, 62, 941-946. <http://dx.doi.org/10.1016/j.foodres.2014.05.016>.
- Yu, Z., & Yang, Z. (2020). Understanding different regulatory mechanisms of proteinaceous and non-proteinaceous amino acid formation in tea (*Camellia sinensis*) provides new insights into the safe and effective alteration of tea flavor and function. *Critical Reviews in Food Science and Nutrition*, 60(5), 844-858. <http://dx.doi.org/10.1080/10408398.2018.1552245>. PMID:30614265.
- Zhang, X. B. (2014). Differences of polyphenols content in Anxi Tieguanyin tea among different seasons and relationship between polyphenols and tea quality. *Agricultural Science and Technology*, 15(7), 1191-1195.
- Zhang, Y. N., Yin, J. F., Chen, J. X., Wang, F., Du, Q. Z., Jiang, Y. W., & Xu, Y. Q. (2016). Improving the sweet aftertaste of green tea infusion with tannase. *Food Chemistry*, 192(5), 470-476. <http://dx.doi.org/10.1016/j.foodchem.2015.07.046>. PMID:26304374.
- Zhuang, J., Dai, X., Zhu, M., Zhang, S., Dai, Q., Jiang, X., Liu, Y., Gao, L., & Xia, T. (2020). Evaluation of astringent taste of green tea through mass spectrometry-based targeted metabolic profiling of polyphenols. *Food Chemistry*, 305, 125507. <http://dx.doi.org/10.1016/j.foodchem.2019.125507>. PMID:31622805.