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Study on the sensory and safety quality characteristics of "High mountain tea" and its formation causes: A comprehensive basic research

Cong-ming Wang¹, Xiao Du^{1*}, Cong-ning Nie², Xiang Zhang³, Xiao-tan Qin¹, Qian Li¹

Abstract

High mountain tea (HT) is an important resource for high quality tea because of its superior ecological environment and rich contents. In this study, we used sensory evaluation, gas chromatography–mass spectrometry (GC-MS), liquid chromatography–mass spectrometry (LC-MS), flavor activity value and risk factors to characterize the flavor (aroma and taste) and safety (heavy metals and pesticide residues) characteristics of HT, and then combined with the production process and environmental conditions to explore the reasons for its quality formation. The results showed that, the aroma sensory characteristics of HT were tender and green, accompanied by sweet and slight chestnut. 8 aroma compounds were identified as the important contributing substances to aroma characteristics and the high ratio of "green substances" and "chestnut substances" was the main reason for its unique aroma characteristics. The taste sensory characteristics of HT were high in freshness and sweetness, low in bitterness and astringency. High content of soluble sugar, non ester catechins, sweet free amino acids and low content of caffeine and tea polyphenols are the main reasons for its taste characteristics. Low temperature stress is the most fundamental reason for the formation of flavor characteristics of HT. In addition, the pollution risks of HT of 5 heavy metals and 50 pesticide residues are all less than 1. The complex ecosystem and low level of chemical control are the main reasons for the high safety quality of HT. All in all, these findings enable us to have a more comprehensive understanding of the quality characteristics and formation reasons of HT.

Abbreviations of experimental materials

HT	High mountain tea
HS	High mountain Shihua
HG	High mountain Ganlu
LT	Low mountain tea
LS	Low mountain Shihua
LG	Low mountain Ganlu

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Introduction

In China, there has always been a saying that "High mountain clouds make good tea". High mountain tea (HT) generally refers to the tea products produced in high altitude mountainous areas, where the ecological environment is generally superior, with low temperature, less sunshine and more clouds. Tea in these mountainous areas generally grows slowly, and the contents of tea (taste and aroma substances) are rich¹⁻⁴. At the same time, due to the low yield of *Camellia*, and the shortage of labor force in mountainous areas, most of the HT areas adopt "natural agricultural method" cultivation, with less chemical fertilizer and pesticide application, and high quality and safety of tea⁵⁻⁷. For a long time in the past, HT has always been synonymous with "high-end tea" and "luxury tea", and ordinary people seldom have access to it. With the improvement of people's living standards, the pursuit of quality and health is becoming higher and higher. HT has gradually become the object of concern.

In the past, people's perception of "High mountain clouds make good tea" was just a basic knowledge of tea consumption and production, or a common cognition of tea drinkers. In fact, the flavor characteristics of tea depend on the coordination of its components and their proportions⁸, and in modern food flavor research, the flavor characteristics of tea are characterized systematically and quantitatively by constructing the "flavor profile" of tea sensory factors and the "contribution value" of flavor active components, and the sensory flavor and aroma components are analyzed to evaluate the quality of tea⁹⁻¹¹.

Mengding Mountain, located in Sichuan Province of China, is one of the main tea producing areas in China. The nectar produced in this area was once rated as one of the top ten famous teas in China. According to the investigation, the elevation of the main tea gardens in this area is 600-900m, and there are a large number of "semi abandoned" HT areas in the deep mountain areas with the altitude of 900-1200m. These tea areas have less people and more land. Most of them adopt "natural agricultural method" cultivation, less chemical fertilizer and pesticide application, and slow development of tea buds and leaves caused by low temperature, which makes the production cost of tea in this region 55%-75% higher than that in the same region at low altitude, and the tea price is about 100%-200% higher. However, due to the lack of convincing research on the quality characteristics of tea in this region, people's recognition of the tea produced in this region is low. At the same time, due to the high price of tea, the sales volume of tea in this region is low. This leads to a vicious circle, which eventually leads to a large number of tea gardens in the region are semi abandoned and unattended.

In order to provide potential strategies for the development of HT and the sustainable development of tea, we selected HT (900-1200 m) as the representative test site, compared with LT (600-900 m) in this area, and analyzed the quality characteristics of high mountain ecological tea from two aspects of flavor quality and safety quality by using modern food flavor method.

Results

Sensory characteristics of HT

Using quantitative descriptive analysis, five flavor factors of each aroma and flavor of tea samples from different regions and different manufacturing processes were sensory scored according to the method described in 2.3, and the flavor profiles of tea samples were plotted according to the scoring mean (Fig. 1).

In terms of aroma, the aroma profile of HS (High mountain Shihua) showed a tender green predominance accompanied by a faint sweet whose average scores of green, chestnut, tender, fresh, sweet were 3.14, 0.19, 3.97, 0.57, 1.37, respectively. HG (High mountain Ganlu) aroma was described as tender, green, sweet dominated and accompanied by faint chestnut, with scores of 3.54, 1.44, 3.92, 1.18, 2.84 for green, chestnut, tender, fresh, sweet, respectively. The main reason for this difference is mainly due to the difference in the two processing modes, and the fabrication process of Ganlu is more complicated with multiple rubbing twists as well as drying processes, which have a greater impact on the tea's internal components, making its tea aroma profile appear more "plump".

Compared with that of shallow cumulus tea, the aroma profile shape of HS (High mountain Shihua) was basically consistent with that of LS (Low mountain Shihua), and the only difference was that the aroma profile of HS was slightly greater than that of LS, which illustrated that HS had a higher aroma intensity than that of LS. Because Shihua is processed in a simple way to better retain fresh leaf original aroma, we guess that the differences caused are most likely due to the greater abundance of aroma substances contained by alpine fresh leaves themselves. HG (High mountain Ganlu) had quite different aroma profiles from LG (Low mountain Ganlu)(Fig. 1d).HG exhibited high tender (3.92), green (3.54), sweet (2.84) as well as low chestnut (1.44), while LG exhibited high chestnut (3.43).Green versus tender are mainly derived from some alcohols, aldehydes and esters with low boiling points, cis-3-hexenol, nonanal and methyl salicylate¹².The chestnut is mainly derived from pyrrole and furan substances produced by Maillard reaction during tea processing¹³, and the chestnut type substances produced by processing should be basically comparable due to the same processing way adopted in the test, so that in-depth exploration of the causes responsible for alpine vs shallow hills requires identification by separation of aroma substances from tea leaves.

In terms of taste, the HS taste profile showed a predominance of fresh versus bitter, accompanied by a faint sweet, with the scores of fresh, sweet, bitter, acid, puckery being 3.68, 2.61, 3.18, 0, 2.74, respectively. The HG flavor profile showed a predominance of fresh versus sweet accompanied by an appropriate bitter degree as well as faint acid, and its fresh, sweet, bitter, acid, puckery scores were 3.07, 3.54, 1.67, 0, 1.62, respectively.

Compared with LT, the sweet score of HS was 2.61, which was significantly higher than that of LS (1.67), while there was no significant difference in other fresh, bitterness, puckery and acid. The soluble sugar in tea is the main contributor to the sweet of tea¹⁴,

while the large temperature difference between day and night in high mountain is conducive to the accumulation of soluble sugar in tea, which may be the main reason for the sweet of HS higher than LS. Compared with LG, except the acid, there were significant differences in fresh, bitterness, puckery and sweet. HG and LG showed two completely different taste characteristics. The scores of fresh and sweet of HG were 3.07 and 3.54 respectively, which were 22.31% and 50.00% higher than those of LG, while the scores of puckery and bitterness of HG were significantly lower than those of shallow hill. Studies have shown that puckery and bitterness are important factors to reduce the taste of tea and consumers' preference¹⁵. In this respect, HG's taste characteristics of high sweet, high fresh and low bitterness are more easily accepted by consumers.

Content and activity of aroma compounds in tea

Content of aroma compounds in tea: The content and proportion of tea aroma components directly determine the aroma sensory characteristics of tea. There are hundreds of aroma compounds in teas, including esters, alcohols, acids, ketones, terpenes, and so on¹⁶. In this study, 38 aroma components were detected in two types of HT samples, including 13 alcohols, 3 ketones, 7 aldehydes, 4 esters, 8 hydrocarbons and 3 heterocyclic compounds (Table 1). A total of 28 aroma compounds were detected in HS (Table 1), and the aroma compounds with higher contents were (z) - 3-Hexen-1-ol, benzyl alcohol, 2-ethylhexanol, phenyl alcohol, nonanal, valeraldehyde, methyl salicylate, which were 4.01 mg / kg, 3.46 mg / kg, 2.62 mg / kg, 1.94 mg / kg, 1.65 mg / kg, 1.59 mg / kg, 1.58 mg / kg, respectively. A total of 37 aroma compounds were detected in HG (Table 1), and the aroma compounds with higher contents were hexanal, heptaldehyde, Longifolene, nonanal, 1-penten-3-ol, valeraldehyde and linalool s, which were 4.53 mg / kg, 4.53 mg / kg, 2.99 mg / kg, 2.85 mg / kg, 2.77 mg / kg, 2.45 mg / kg and 2.29 mg / kg, respectively. Among these higher aroma compounds, four were common to both types of HT, namely, (z) - 3-Hexen-1-ol, benzyl alcohol, valeraldehyde, nonanal, and these could be the main aroma components that formed the aromatic quality of HT.

Compared with LT, the content and species of volatile components in HT were higher than those in LT, no matter Shihua or Ganlu (Table 1). In HT and LT, the aroma compounds with large difference (difference > 50 %) and high content (content > 1 mg / kg) were: (z) - 3-Hexen-1-ol, 2-ethylhexanol, linalool, linalool oxide II, nerolidol, geranylacetone, heptaldehyde, methyl salicylate, (z) - 3-Hexenyl hexanoate, alpha pinene, alpha caryophyllene and 3-carene. These aroma components may dominate the difference of aroma sense between high mountain and LT. The content of alcohols in HT, such as (z) - 3-Hexen-1-ol, nerolidol and linalool, was higher than that in LT, while the content of terpenoids, such as alpha pinene, alpha caryophyllene and 3-carene, was lower than that in LT. In aroma description, most of these alcohols present green, flower and fruit aroma, while most of the terpenoids present woody aroma. The presence of these woody aroma compounds may mask the expression of green and make tea present other aroma.

2-pentylfuran with caramel flavor is often considered as one of the important aroma components influencing the formation of chestnut^{17; 18}. In this study, the content of 2-pentylfuran in HG and LG was 0.52 mg / kg and 0.59 mg / kg respectively, and there was no significant difference between them (Table 1). We speculate that the reason why HG has the characteristics of high green and low chestnut aroma is that it contains a large number of green alcohols, thus masking the expression of chestnut and woody substances. In LG, the expression of green alcohols is weakened by more woody compounds, while the expression of chestnut compounds is promoted by woody compounds, which makes LG finally show a strong chestnut.

Activity values of aroma compounds in tea: In the study of tea flavor, odor activity value (OAV) is often used to characterize the contribution of aroma compounds to the overall aroma¹⁹. It is generally recognized that the aroma components with OAV > 1 have an important contribution to the overall aroma of tea samples, and the higher the OAV value, the higher the contribution to the overall aroma. In this study, there were 28 aroma compounds with OAV > 1 in HT, including 21 aroma compounds in HS and 27 aroma compounds in HG (Table 1). There were 8 aroma compounds with OAV > 100 both in HS and HG, which were linalool (912.00 / 1529.60), alpha ionone (639.36 / 1373.76), nonanal (471.99 / 813.64), hexanal (317.95 / 905.47), linalool oxide II (166.40 / 320.00), 2, 2-Dimethyl-6-methylenecyclohexane-1-carbaldehyde (147.92 / 171.42), Linalool oxide I (125.60 / 216.00), Geraniol (125.18 / 139.78). These aroma components may be the key components to form the unique aroma characteristics of high mountain. On the other hand, there are four compounds with OAV < 1 both in HS and HG, which are 2-acetyl pyrrole (0.01 / 0.01), 1-pentanol (0.04 / 0.02), benzyl alcohol (0.63 / 0.38) and benzyl benzoate (0.79 / 0.81). These aroma components may not be the key components of HT aroma characteristics.

In sensory evaluation, HS aroma was mainly described as tender and green, accompanied by faint sweet, while HG aroma was mainly described as tender, green and sweet, accompanied by faint chestnut (Fig. 1B). According to the OAV of aroma substances and their aroma description, linalool aroma was described as "coriander, floral, lavender, lemon, rose", alpha ionone as "violet, fruit, wood", nonanal as "fat, floral, green, lemon", hexanal as "apple, fat, fresh, green", linalool oxide I and linalool oxide II as "floral", 2-dimethyl-6-methylenecyclohexane-1-carbaldehyde as "fresh fruit, Camphor", geraniol as "geranium, lemon peel, passage fruit, peach, rose". These aroma substances interact with each other to form the unique aroma characteristics of HT.

Content and contribution value of taste compounds in tea

The taste of tea is the comprehensive perception of its taste components by taste. The main taste substances of tea are tea polyphenols, amino acids, caffeine, soluble sugar, and the content and proportion of these substances have a profound impact on the taste of tea soup²⁰. Through the detection and analysis of taste components in HT samples, we can further explore the taste characteristics and causes of HT.

Content of taste compounds in tea: As can be seen in table 2, only three taste substances were significantly different between HS and LS, namely SS, Phe and GCG, while six were found in Ganlu, namely Ca, SS, Gln, Phe, Arg and EGC. This

phenomenon has a great relationship with the two processing ways as well as the source of raw materials. First, the raw materials (one shoot and one leaf) of Ganlu have a longer growth and development time, which is more influenced by the environment, and second, the processing process of Ganlu is more complex, which further amplifies the gap between the two.

The main different taste substances between HT and LT are bitter and sweet substances. For example, CA, Phe, Arg as well as EGC of bitter components in HG were significantly lower than those in LG, while ss of sweet components were significantly higher than those in LG ($P < 0.05$). The differences in these taste substances led to the different sensory profiles of the taste (Fig. 1A).

Contribution of taste compounds in tea: As the main taste substances of tea such as tea polyphenols and soluble sugar are a complex, there is no unified threshold, so the taste activity value (TAV) can not be determined. Therefore, we use the standardized coefficient of partial least squares regression (PLSR) to analyze the contribution of 20 taste substances to five taste attributes of tea, and the results are shown in Figure 2. The color depth represents the contribution, red is the positive contribution, and blue is the negative contribution. It can be seen from the figure that polyphenols, caffeine, ester catechins (EGCG, ECG, GCG) and some bitter amino acids (Phe, Arg) contribute to the bitterness and puckery of tea. Soluble sugars and some amino acids (ASP, Gln, Ser) contribute to the fresh sweet of tea. The contribution of theanine to fresh is weaker than that of some free amino acids, such as aspartic acid and glutamine, mainly because of its high threshold. There are few substances related to sour taste, mainly because green tea has almost no sour taste.

Safety quality characteristics of HT

Besides the flavor quality of tea, the quality of tea also includes the safety quality of tea. The main control factors of tea safety quality include the types and contents of pesticide residues, harmful heavy metals and microorganisms, among which pesticide residues and harmful heavy metals are the main factors. In this study, five harmful heavy metals and 50 kinds of pesticide residues in different tea samples of high mountain and low mountain were detected, and the results are shown in Table 3.

From the content of heavy metals in tea, the average content of five heavy metals in HT was far lower than the maximum limit value of the corresponding standard. The contents of Cr, as, Cd, Pb and Cu in HS were 67.0 %, 96.3 %, 93.5 %, 85.8 % and 76.5 % lower than the limit value respectively, and the contents of Cr, as, Cd, Pb and Cu in HG were 61.2 %, 98.35 %, 96.5 %, 88.9 % and 76.9 % lower than the limit value respectively. This indicated that all HTs were not polluted by heavy metals, and the risk of heavy metal pollution in HT was very low. Compared with LT, HT contained more Cr, as and Cd. Among the three heavy metal elements, the average pollution index of Cr is the largest, reaching 0.388, while the pollution index of the other two heavy metals is smaller (< 0.1). Therefore, we think that CR is the most dangerous heavy metal element in HT at present, and we should strengthen the control of Cr in the production and processing of HT in the future.

In terms of the pesticide residues of tea, a total of seven pesticide residues were detected in all HT samples, four of them were detected in Shihua and seven in Ganlu, and all of them were found to be well below the corresponding minimum limit values (Table 3). Taking the pesticide chloropyrifos, for example, which has a relatively large pollution index in HT, its pollution index in HS and HG is only 0.086 and 0.130, respectively, which is much lower than its limit pollution index of 1, which illustrates the HT's high safety factor. The agroforestry species and contents of the HT samples were all lower than those of the LT samples when compared with the LT samples. For example, only 4 agroforestry residues were detected in HS, whereas 8 were detected in LS. The main reason for this is that pesticides use is much less in the HT area with a rich ecosystem and a cold winter season where infestation occurs less frequently.

Discussion

Analysis on the formation of flavor quality characteristics of HT

*** How do the content and proportion of tea flavor components affect the flavor of HT?**

The flavor components of tea mainly refer to the aroma components and taste components of tea, which are characterized by many kinds, complex composition, great difference in the contents of major and trace components, and complex leaching, mixing and interaction of each component. These characteristics and their interaction will affect the sensory quality and flavor of tea to varying degrees²¹⁻²⁴.

Effects of content and proportion of components substances on tea flavor: The sensory evaluation showed that the aroma of high mountain green tea was mainly tender

and green, accompanied by a certain amount of sweet and weak chestnut, while the aroma of low mountain green tea was described as chestnut, accompanied by a certain amount of tender, green and sweet. GC-MS was used to detect related components and their contents, showing consistent component content and dose-effect relationship (Table 1). The measured data showed that the content of alcohols with fruity and green flavor in HT was indeed higher than that in Low Mountain Tea, which was the main reason for its unique aroma. At the same time, tea aroma is not only related to the content and ratio of tea components, but also shows synergistic, masking and tone-changing effects among the components²⁵⁻²⁹, which all reflect the difference of tea flavor characteristics and sensory experience. As the previous research of our team has shown, under the same OAV, the aroma components of floral fragrance are easily masked by the aroma of other fragrances³⁰. Based on the detection results of aroma components, we speculated that another main reason for the different aroma characteristics of HT and Low Mountain Tea might be the difference in the ratio (G/C) of "green substances" and "chestnut substances". Taking Ganlu as an example, the G/C value of HG is relatively large, a large number of Green and fruit-flavorings such as (Z)-3-hexen-1-ol, Benzyl alcohol, Linalool and Nerolidol will cover up the expression of chestnut substances, thus making it present the characteristics of high green and low chestnut. However, the G/C value of LG is relatively small, the green substances with low content cannot cover up the chestnut substances, so it finally presents a high chestnut.

In addition, we also found that the content of hydrocarbons in LT is relatively high, and most of these hydrocarbons are "Wood". These wood substance may have an offsetting effect with green substance and a promoting effect with chestnut, thus further promoting the expression of chestnut substance. In addition to the aroma substance composition and proportion relationship, the aroma components of the interaction between (masking / offset / enhancement) is also the important factors that affect HT aroma (Fig.3), to further explore the mechanism of formation of the high mountain green tea scent, still need to study the interaction between the aroma components.

Effects of content and proportion of tasting components on tea flavor: Taste characteristics of HT, from the taste profile reflects that the sweet taste is high, bitter taste is low. It is generally believed that one of the main reasons for the high quality of HT is that "HT has lower tea polyphenols and higher total amino acids³¹", which determine the quality of HT³². In this study, high mountain green tea did contain lower tea polyphenols and higher total amino acids, but the difference did not reach a significant level. Therefore, we speculate that the reason why HT has high flavor, sweet and low bitter taste is not the action of a certain kind or several substances, but the formation of a variety of substances. These substances include: high content of soluble sugar, non-ester catechins, low threshold sweet free amino acids, and low content of caffeine and tea polyphenols.

*** How does the environment affect the content and proportion of components in HT?**

The flavor quality of tea is directly determined by the content and ratio of the substances contained in tea, which in turn is mainly determined by the environmental

conditions of tea tree growth³³⁻³⁵.

The accumulation of inclusion in HT is related to the high mountain environment of tea tree, such as high altitude, low temperature, cloudy and diffuse light, etc. Under such conditions, the late germination of tea bud and the slow growth of tender leaves are conducive to the accumulation of inclusion and optimization of the ratio³⁶. The results showed that the annual average temperature of HT tea garden was 2-3.5 °C lower than that of LT tea garden, the annual sunshine time was 400-550h less, and the bud and leaf production period was 15-36 days more (the data came from the internal investigation of the local meteorological bureau and the research group). These conditions were conducive to the nitrogen metabolism and accumulation of free amino acids; at the same time, they could inhibit carbon metabolism to a certain extent, and reduce the synthesis of astringent substances such as tea polyphenols and caffeine³⁷; The germination of HT buds began last autumn and winter and under the effects of low temperature stress in winter and temperature difference between day and night, soluble sugars such as sucrose and glucose will accumulate in tea leaves³⁸. Soluble sugar is an important contribution to the sweet of tea, and "sweet" is also one of the biggest characteristics of HT compared with LT (Fig.1B). In addition, low temperature stress can also promote the accumulation of glycosides in tea³⁹, and when tea tree leaf bites and damage by pests, these indican substance could hydrolysis to release the fragrance, for example, nerolidol, geraniol, linalool, benzyl alcohol and phenylethyl alcohol⁴⁰⁻⁴². Most of the floral aroma compounds with low boiling point volatilize in the process of tea processing, and the rest are mostly green and fruit aroma compounds, which give high mountain tea unique aroma characteristics.

Analysis on the formation of safety quality of HT

*** Causes of safety quality characteristics of heavy metals in HT**

In this study, the average contents of Cr, As, Cd, Pb and Cu in all HTs were lower than the limited value, indicating high quality of heavy metal safety and quality. Heavy metals in tea mainly come from soil^{43; 44}, according to the content determination results of 5 major heavy metals in the soil of the tea garden tested by the research group before (Fig. 4), We found that under the influence of soil heavy metal ion activity, the content of heavy metals in tea was not completely positively correlated with the content of corresponding elements in soil, and it was also significantly related to the enrichment ability of different heavy metals in the growth of tea trees. For example, the Pb content of HT is lower than that of LT (Table 3), but the Pb content of HT soil is higher than that of LT soil (Fig. 4). We preliminarily speculated that the climatic environment of high mountain changed the absorption and enrichment capacity of different heavy metals in HT trees, and this inference still needs to be further explored in subsequent experiments.

*** Causes of pesticide residues in HT and potential safety hazards**

The direct factor affecting pesticide residues is the prevention and control methods of pests and diseases in tea gardens, and excessive chemical prevention and control is the main reason for the risk of pesticide residues⁴⁵⁻⁴⁷. This study sites of HT areas, vegetation coverage rate is high, the rich ecosystem around tea garden, tea garden

beneficial insects population and diversity index is larger, this natural advantage will directly or indirectly, inhibition of plant diseases and insect pests, reduce the HT garden management reliance on chemical pesticides to control plant diseases and insect pests. The proportion and frequency of pesticide use in HT gardens were lower than those in LT gardens (Table 4), which reduced the risk of pesticide residue in HT to a large extent.

In addition, it should be noted that due to the low investment in tea pest control (only 16.67% HT garden has plant protection personnel). As a result, the damage index of fresh shoots of tea gardens in High Mountain reached 0.23, more than twice that of tea gardens in Low Mountains (Table 4). The damage degree of new shoots of tea leaves in High Mountain was greater than that in Low Mountain, which would reduce the yield of fresh leaves in HT gardens. At present, the yield of fresh leaves in the HT gardens in this region exceeds the demand. Under such supply and demand relationship, the managers of the tea gardens are willing to sacrifice part of the tea output in exchange for high safety quality and low management cost of tea. However, once this supply and demand relationship is broken, when the demand for fresh leaves of HT increases, investment in pest and disease prevention and control of tea plantation will increase. If improper operation, such as the large use of pesticides, will increase the risk of pesticide residue pollution of HT, which should be paid attention to.

Materials and methods

Study area

The experimental area is located in Mengding Mountain, Ya'an City, Sichuan Province, China (Fig. 5A). According to the local minimum altitude (600m) and the local actual tea production situation, the tea area with an altitude of more than 1000m and the characteristics of typical HT area (superior ecological environment, far away from the city, extensive management, little use of chemical fertilizers and pesticides, and low utilization rate of tea Garden Development) were defined as HT area; the area with altitude less than 1000m and typical LT characteristics (tea garden development and utilization) was defined The area with high rate, a large amount of pesticides and chemical fertilizers will be invested every year, and it is close to the city) is defined as LT area. There are 6 HT areas and 7 LT areas.

Sample collection and processing

From the middle of March to the beginning of May, 2018, bud and one bud with one leaf were collected respectively in the above 13 areas, and different green tea was made according to the local technology of Shihua and Ganlu (Fig. 5B). They were numbered, packaged in composite aluminum bags and frozen at - 5 °C.

Quantitative description evaluation

The sensory evaluation panel included trained assessors (7 males, 5 females: age 23–27 years). The solution was sniffed and graded (5-point scoring method) by the evaluation team which trained in systematic sniffing (specific training methods refer to the previous reports of our group³⁰, including two parts, strength training of aroma

monomer and recognition training of mixed solution). Using distilled water as CK, room temperature 23 ± 1 °C, all experiments were repeated 3 times.

The flavor profile analysis was carried out according to the method established by our group. The chosen taste descriptors were fresh (reference standard components are glutamate), sweet (sucrose), bitter (quinine), puckery (tannins) and acid (citric). The chosen aroma descriptors were green (reference standard aroma components are trans-3-hexenol), tender (nonanal), chestnut (3-Methylbutyraldehyde), sweet (phenylethanol) and fresh (trans-2-hexenal). 5-point scoring method was selected for evaluation, which were 0 (odorless)- 5 (strong).

Determination of aroma compounds

Extraction of aroma-active components: the HS-SPME method was chosen to isolate the aroma active components; a tea sample (3.0 g) with 30 µg ethyldecanoate as an internal standard (10 mg/kg tea sample) was placed in an extraction bottle (15 mL volume). The sample was equilibrated in a thermostatic water bath for 10 min at 50 °C, and then sampled for 30 min in the head space. After that, SPME fiber was withdrawn and directly introduced to the GC-MS, and the process was repeated three times.

GC-MS instrument setup and analytical conditions: GC-MS analyses were performed on an Agilent 7890A/5975C-GC/MSD inert detector operating in EI mode in a 69.9 eV chromatographic column: capillary-column chromatography DB-5 ms (30 m × 250 µm × 0.25 µm). The sampling was manual, no shunt was used, the sample was at a constant temperature, and the temperature of the injection port and GC-MS direct interface were 250 °C and 280 °C, respectively. Temperature programming: the column temperature was 50 ~ 250 °C; the starting column temperature was 50 °C which was held for 3 min, and then increased to 150 °C at a rate of 2 °C/min, held for 2 min, then increased to 250 °C at a rate of 2.5 °C/min and held for 4 min. Helium was used as a carrier gas at a flow rate of 1.0 mL/min. The ion source temperature was 230 °C, the quadrupole temperature was 150 °C and the scanning quality range was set at 20 ~ 700 amu.

Qualitative and quantitative analysis of aroma compounds: Qualitative analysis: the NIST standard spectral library (<https://www.nist.gov/>) and retention indices (RIs) from other literatures were used to match ion mass spectra, and then completed the qualitative analysis of aroma components. Quantitative analysis: the internal standard method was used to quantify the volatile flavour compounds in the tea aroma. Ethyl decanoate was selected as the internal standard (10 mg/kg tea sample).

Determination of taste compounds

Determination methods of total tea polyphenols, total amino acids, total caffeine and total soluble sugar: weigh 0.6 g (± 0.001 g) of tea sample into a 150 ml conical flask, add 60 ml of boiling distilled water, immediately move it into a boiling water bath, and extract for 45 min (shake every 10 min). After the extraction, it was immediately filtered under the condition of hot decompression. The filtrate is transferred into a 100ml volumetric flask, the residue is washed with a small amount of hot distilled water for 2-3 times, and the filtrate is filtered into the volumetric flask. After cooling, the filtrate

is diluted to 100 ml with distilled water. Operate according to the methods specified in the following determination methods, and calculate the content of each component in the sample according to the mass of dry tea sample. The total amount of tea polyphenols was determined by Folin phenol method, the total amount of free amino acids was determined by Ninhydrin colorimetry, the content of caffeine was determined by ultraviolet spectrophotometry, and the total amount of soluble sugar was determined by anthrone colorimetry.

Determination of catechin components: weigh 1.5 g (± 0.0001 g) of uniformly ground sample, add 125 ml boiling water, extract in boiling water bath for 45 min, pump and filter while hot, finally fix the volume to 250 ml, filter with 0.45 μ m water phase filter head, and collect 2 ml filtrate for standby. Detector: waters 600 double pump HPLC, watersempower chromatographic management system, waters2489 UV detector. Methods: the chromatographic column was phenomenex Gemini C₁₈ (250 mm \times 4.6 mm, 5 μ m), the elution temperature was 20 °C, the injection volume was 5 μ L, the flow rate was 1 ml / min, and the detection wavelength was 278 nm. Phase A is 0.2 % acetic acid solution, and phase B is pure acetonitrile.

Determination of amino acid composition: accurately weigh 0.25 g (± 0.0001 g) uniformly ground sample in 50 ml centrifuge tube, weigh, add 25 ml boiling water, extract in boiling water bath for 45 min, quickly cool to room temperature, make up water loss, centrifuge at 4000 R / min for 10 min, and draw the supernatant for testing. Take 5 ml of the supernatant, add 5 ml of 5 % trifluoroacetic acid solution, centrifugate at 4 °C 7000 r / min for 20 min, pass through 0.45 μ m inorganic filter membrane, suck 20 μ l of the test solution, inject it into the automatic amino acid analyzer, and use external standard method for quantitative analysis.

Each sample was measured three times in parallel and the average value was taken.

Determination of heavy metals in tea

Tea pretreatment: nitric acid perchloric acid wet digestion method was adopted, that is, 1.0 g (± 0.001 g) of sample was weighed into a Teflon cup, 25 ml of mixed acid with volume ratio of 5:1 (HNO₃ : HClO₄) was added, placed in the fume hood, covered and soaked for 16h, heated with electric heating plate, the temperature was 150 \pm 2 °C, slowly digested until the residual 2-5 ml was colorless and transparent liquid, then it was removed and cooled The deionized water was washed, filtered, and the volume was set to 25 ml.

Cr was detected by graphite furnace atomic absorption spectrometry, As was detected by Liquid Chromatography Atomic Fluorescence Spectrometry, Cd was detected by graphite furnace atomic absorption spectrometry, Pb was detected by Graphite Furnace Atomic absorption spectrometry and Cu was detected by Flame atomic absorption spectrometry.

Detection parameters: the absorption wavelengths are 357.9 nm for Cr, 228.8 nm for Cd, 283.2 nm for Pb, and 324.7 nm for Cu respectively. The lamp current is 10.0 mA, the spectral bandwidth is 0.7 nm, the high pressure is 207.00 v, the gas flow rate is 2.2 l / min, and the burner height is 7.0 mm.

Determination of pesticide residues in tea

Tea sample pretreatment: acetonitrile was used as extraction solvent, ultrasonic extraction, and carb / NH₂ solid phase extraction column was used to purify or activated carbon was used to remove impurities such as pigment. The pesticide residues, such as organophosphorus, pyrethroid and organochlorine, which are thermally stable, weakly polar and volatile, were detected by gas chromatography (GC); insecticides with thermal instability or low vapor pressure, such as carbamates and N-methyl carbamates, were detected by high performance liquid chromatography (HPLC); imidacloprid and acetamiprid were detected by ultra-performance liquid chromatography tandem mass spectrometry (LC-MS); seven kinds of pyrethroids were detected The pyrethroids were detected by GC-MS.

GC-MS detection conditions: Agilent 7890b-7000c gas chromatography tandem mass spectrometry was used, Agilent hp-5ms 30 m × 250 μ m × 0.25 μ m capillary column was used; injection port temperature: 270 °C; injection volume was 1 μL, no split flow injection; gradient heating program of column temperature box: after holding 80 °C for 2 min, 15 °C/min was raised to 310 °C and kept for 5 min, with a total of 22.33 min; carrier gas N₂ ≥ 99.999 %, 1 ml/min constant current mode; separation of nitrogen and nitrogen was carried out Sub source temperature, 230 °C; quadrupole temperature, 150 °C; electron bombardment source voltage, 70 ev.

LC-MS detection conditions: Agilent 1290-6470 ultra-high performance liquid chromatography tandem mass spectrometer, Agilent c1850 mm × 0.30 mm × 1.8 μ m column. The injection volume was 1.0 μL, the mobile phase a was methanol, and the mobile phase B was water (containing 0.1 % formic acid). Gradient elution process: 0~1 min, 10 %A; 1~6.6 min; 10%~80 %A; 6.6~7.0 min, 80%~90 %A; 7.0~7.10 min, 90%~98 %A; 7.10~11.60 min, 98 %A; 11.60~15.00 min; 98 %A; flow rate, column temperature 30 °C, electrospray ionization (ionization) source, positive ion mode, dry gas temperature 350 °C, dry gas flow rate, nebulizer pressure, sheath gas temperature 300 °C, sheath gas flow rate /The capillary voltage was 4000 V, scanning mode was DMRM.

Statistical analysis

The experimental results are expressed as the means ± standard deviations (SD) of three parallel measurements. Correlations were calculated using Partial least squares regression (PLSR). Statistical analyses were performed using SPSS25.

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Author contributions

Conceptualization, Cong-ming Wang and Xiao Du; formal analysis, Cong-ming Wang and Cong-ning Nie.; experiment and investigation, Cong-ming Wang, Cong-ning Nie and Xiang Zhang; funding acquisition, Xiao Du; writing and original draft, Cong-ming Wang; review and editing, Xiao-qin Tan and Qian Li. All of the authors read and approved the final manuscript.

Competing interests

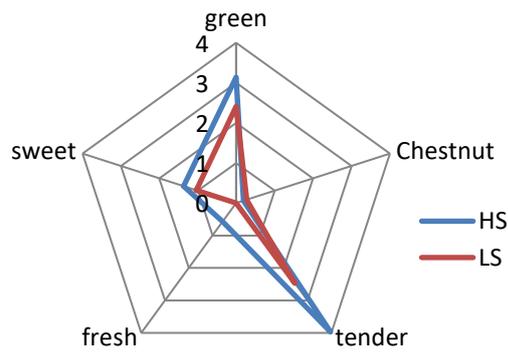
The authors declare no competing interests.

Ethics requirements.

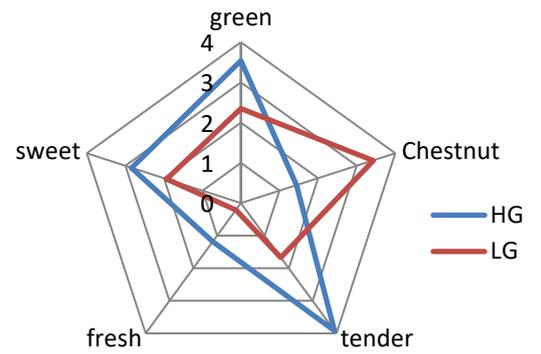
In the sensory evaluation part of this study, trained evaluators were used to conduct necessary sensory evaluation on tea samples. Except for the above parts, this study does

not include other work with human or animal. This study was approved by the local ethics committee of China: "Sichuan Regional ethics review committee of traditional Chinese Medicine", and all methods were carried out in accordance with relevant guidelines and regulations, informed consent was obtained from all subjects before their participation in the study.

A
Aroma profile

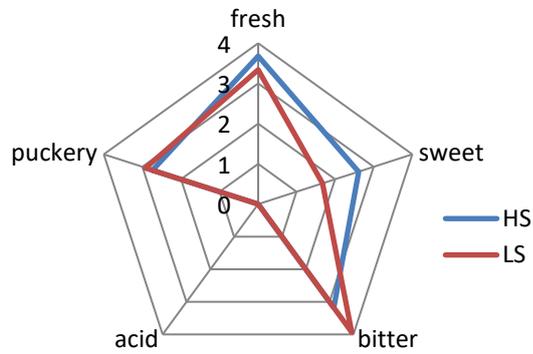


(c) Aroma profile of Shihua

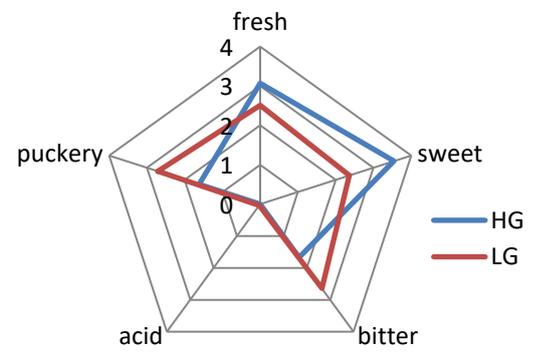


(d) Aroma profile of Ganlu

B
Taste profile



(a) Taste profile of Shihua



(b) Taste profile of Ganlu

Fig 1. Flavor profile of different tea. “HS”,High mountain Shihua,“LS”,Low mountain Shihua,“HG”,High mountain Ganlu,“LG”,Low mountain Ganlu.

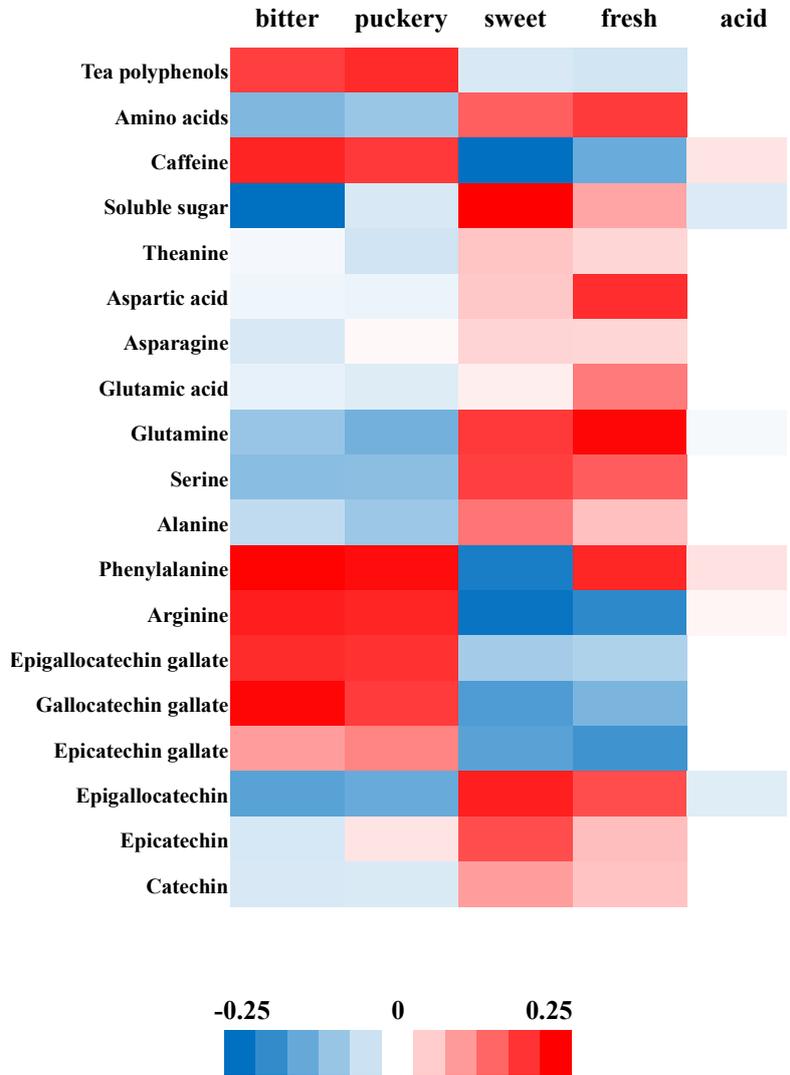
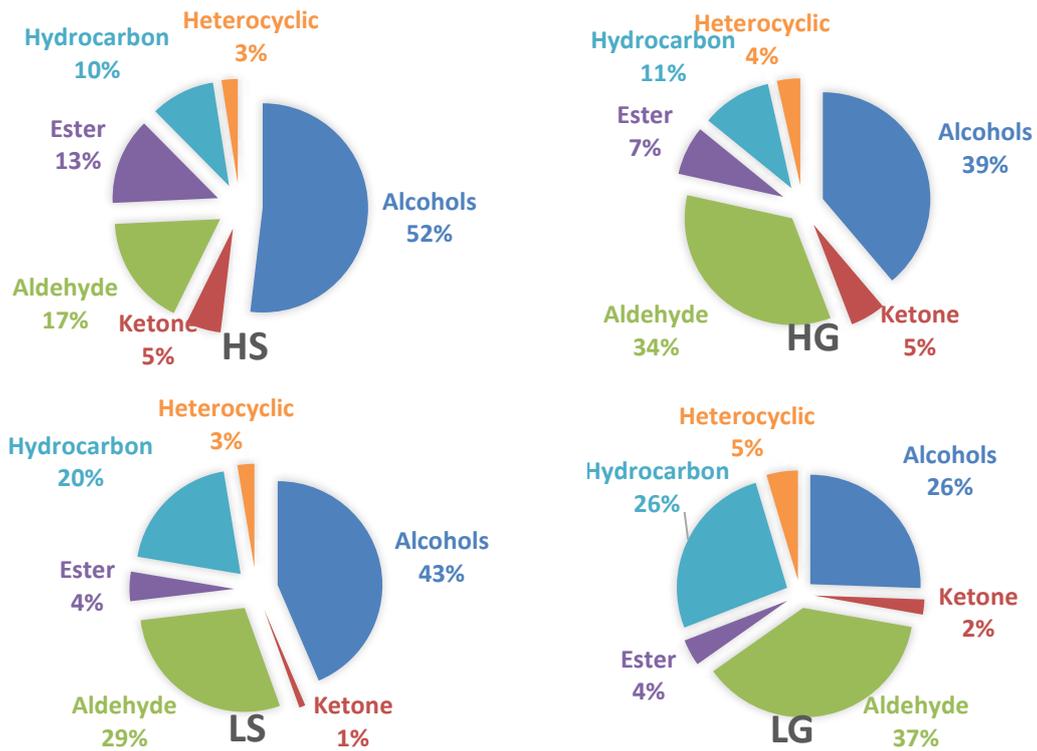
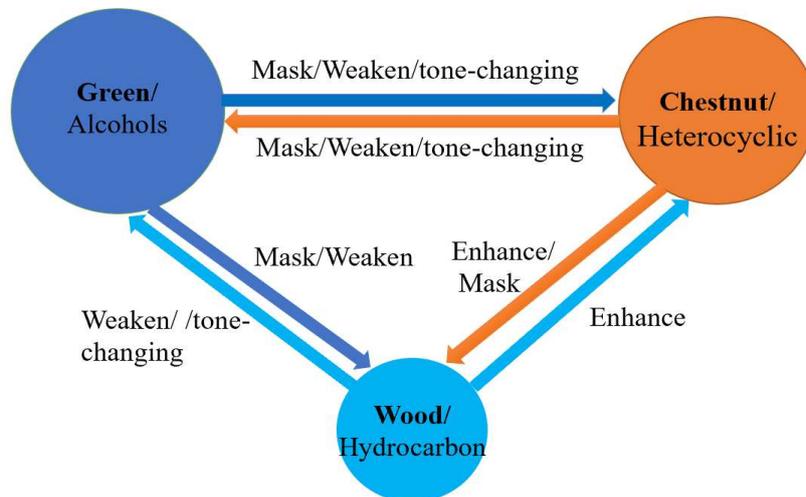


Fig 2. Correlation between main taste substances and sensory characteristics of HT. Here, we use the standardized coefficient of partial least squares regression (PLSR) to analyze the contribution of 20 tea flavor substances to 5 tea flavor attributes



A: Types and proportions of aroma compounds in different types of tea.



B: Possible relationship between aroma compounds in tea

Fig 3. The proportion of aroma compounds in HT and LT and the possible relationship between them

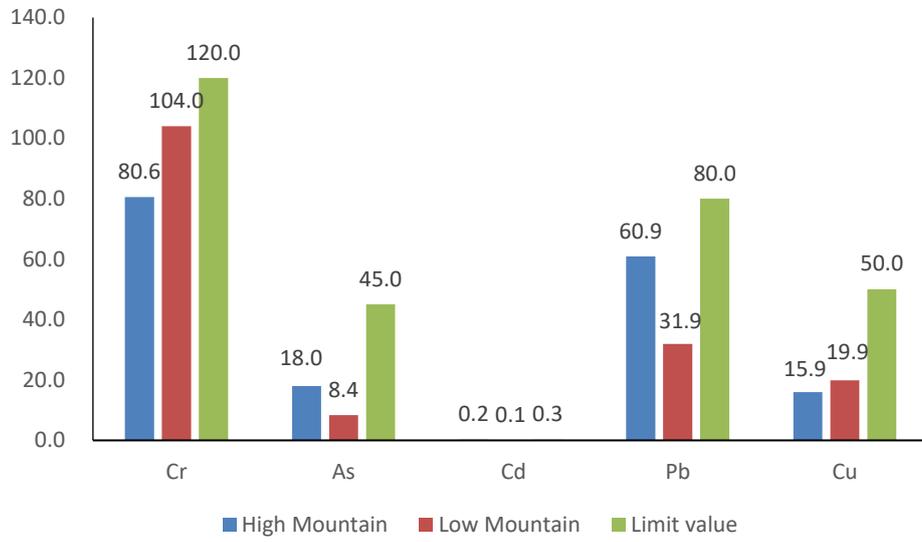
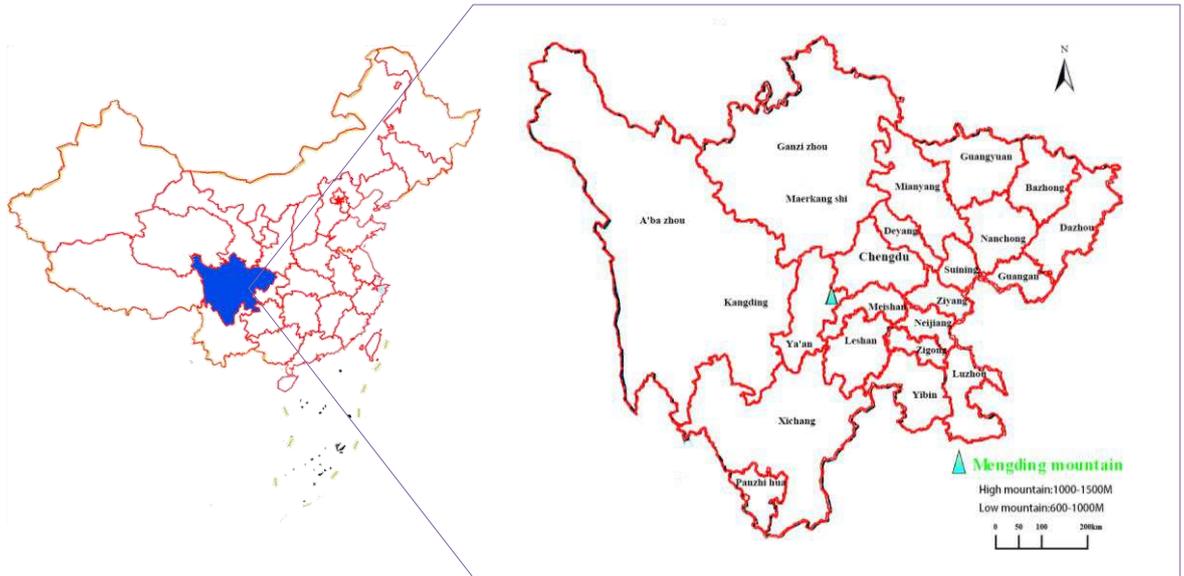


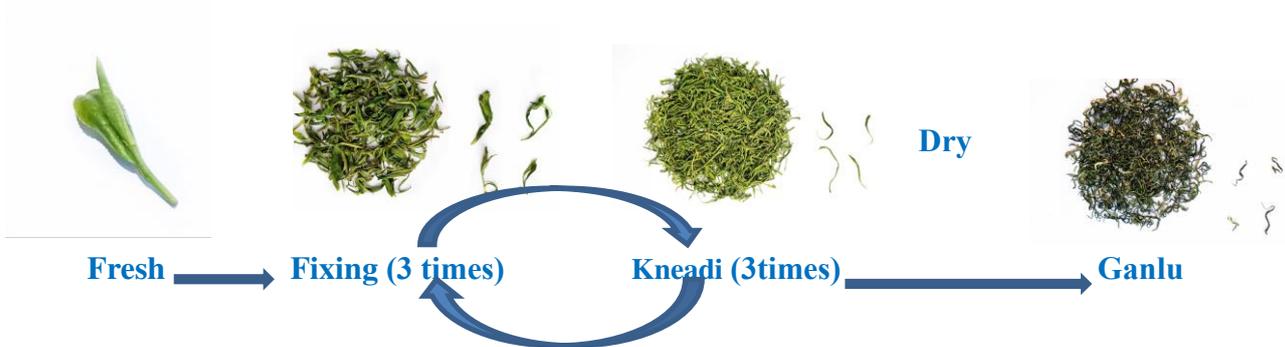
Fig. 4. Heavy metal content of tea garden soil in different areas. The data come from our research group. The limit value comes from the Chinese agricultural standard: Environmental conditions of producing areas of pollution free agricultural products (NY5010-2016).

A



(a) The making process of Shihua

B



(b) The making process of the stone flower of Ganlu

Figure 5. Location map of study area and treatment method of study materials. A: Location map of study area, the sampling site is located in Mengding Mountain, Ya'an City, Sichuan Province, China. B: The making process of Shihua and Ganlu

Table 1. Content of aroma components, OAVs and flavor description of different tea samples

No.	Category	Aroma components	Concentration(mg/g) ^a				OAV ^b				Aroma description ^c	
			HS	LS	HG	LG	HS	LS	HG	LG		
1	Alcohols	1-Penten-3-ol	0.97	0.30	2.77	2.41	0.39	0.12	1.11	0.96	Butter, Fish, Green	
2		1-Pentanol	0.19	0.17	0.08	0.08	0.04	0.03	0.02	0.02	Balsamic, Fruit, Green	
3		(Z)-3-Hexen-1-ol	4.01	3.36	1.75	0.34	20.04	16.80	8.75	1.70	Grass, Green Fruit, Green Leaf	
4		1-Hexanol	-	-	0.23	0.04	Null	Null	0.90	0.17	Banana, Flower, Grass	
5		2-Ethylhexanol	2.62	1.48	2.28	1.14	Null	Null	Null	Null	Green, Rose	
6		Benzyl alcohol	3.46	2.95	2.09	1.58	0.63	0.54	0.38	0.29	Boiled Cherries, Moss, Roasted Bread,	
7		Linalool oxide I	0.75	0.65	1.30	0.58	125.60	108.80	216.00	96.80	Floral	
8		Linalool	1.37	1.51	2.29	1.06	912.00	1004.80	1529.60	704.00	Coriander, Floral, Lavender, Lemon,	
9		Linalool oxide II	1.00	0.58	1.92	0.62	166.40	96.80	320.00	104.00	Floral	
10		Phenethyl alcohol	1.94	1.15	1.55	1.99	1.94	1.15	1.55	1.99	Fruit, Honey, Lilac, Rose, Wine	
11		DL-Menthol	0.73	0.75	0.75	0.60	2.44	2.50	2.50	2.02	Mint, Cool	
12		Nerolidol	0.75	-	1.97	-	50.30	Null	131.04	Null	Rose, Keiskei, Apple	
13		Ketone	Geraniol	0.94	-	1.05	0.94	125.18	Null	139.78	125.95	Geranium, Lemon Peel, Passion Fruit,
14	6-Methyl-5-hepten-2-one		0.96	-	0.43	-	9.62	Null	4.30	Null	Fruit, Fresh	
15	Alpha-Ionone		0.26	0.28	0.55	0.74	639.36	695.52	1373.76	1844.64	Violet, Fruit, Wood	
16	Aldehyde	Geranylacetone	0.72	-	1.72	0.25	12.01	Null	28.63	4.09	Apple, Banana	
17		Valeraldehyde	-	-	2.45	5.14	Null	Null	24.47	51.42	Almond, Bitter, Malt, Oil, Pungent	
18		Hexanal	1.59	2.38	4.53	3.85	317.95	476.93	905.47	770.00	Apple, Fat, Fresh, Green	
19		Heptaldehyde	0.88	2.45	4.53	2.31	28.32	78.93	146.04	74.47	Citrus, Fat, Green, Nut	
20		Benzaldehyde	1.28	1.02	1.45	1.09	4.26	3.41	4.84	3.62	Bitter Almond, Burnt Sugar, Cherry,	
21		2,4-Heptadienal	-	-	0.73	0.56	Null	Null	207.36	159.96	Fat, Nut	
22		Nonanal	1.65	1.89	2.85	1.87	471.99	539.14	813.64	535.19	Fat, Floral, Green, Lemon	
23		2,2-Dimethyl-6-	0.74	0.72	0.86	0.82	147.92	143.77	171.42	164.51	Fresh fruit, camphor	
24		Ester	Hexyl acetate	1.16	-	1.18	0.82	29.03	Null	29.55	20.39	Apple, Banana, Grass, Herb, Pear
25			Methyl salicylate	1.58	0.39	0.72	-	26.41	6.45	11.95	Null	Almond, Caramel, Peppermint, Sharp
26	(Z)-3-Hexenyl hexanoate		1.30	0.28	1.06	0.24	13.03	2.78	10.61	2.40	Fruit, Prune	

27		Benzyl benzoate	0.79	0.68	0.81	0.68	0.79	0.68	0.81	0.68	Balsamic, Herb, Oil
28	Hydrocarbon	DL-Limonene	0.59	-	-	0.48	1.19	Null	Null	0.95	Orange
29		Alpha-Pinene	-	-	0.20	1.09	Null	Null	1.66	9.10	Cedarwood, Pine, Sharp
30		Alpha-Farnesene	-	-	0.31	0.61	Null	Null	0.62	1.22	Boiled Vegetable, Floral, Wood
31		Alpha-Caryophyllene	-	-	0.57	2.38	Null	Null	3.56	14.88	Wood
32		3-Carene	-	-	0.44	1.79	Null	Null	3.38	13.76	Pine, Wood
33		Myrcene	0.63	1.11	0.73	1.30	44.93	79.49	51.84	92.82	Balsamic, Fruit, Geranium, Herb, Mus
34		Longifolene	0.35	1.73	2.99	5.26	Null	Null	Null	Null	Pine
35		N-Heptadecane	-	-	0.10	0.80	Null	Null	Null	Null	Sweet, Foral, Preserves
36	Heterocyclic	2-Acetylfuran	-	-	0.60	0.73	Null	Null	0.01	0.01	Butter, Floral, Fruit, Green Bean
37		2-Pentylfuran	-	-	0.52	0.59	Null	Null	108.00	122.40	Butter, Caramel
38		2-Acetyl pyrrole	0.89	0.77	0.66	0.73	0.01	0.01	0.01	0.01	Bread, Cocoa, Hazelnut, Licorice,

^aIn order to better explain the aroma characteristics of different tea samples, the representative tea samples with the most suitable aroma profile were selected for GC-MS detection. ^bOAV= C/OT, C is the concentration of a single volatile component, OT is the odour threshold of the same volatile component. The references of OT: Leffingwell & Associates Database (<http://www.leffingwell.com/>), and compilations of odour threshold values in air, water and other media and compilations of flavour threshold values in water & other media (2011 editions)—by van Gemert (<http://www.thresholdcompilation.com/>). ^cThe source of aroma description: FEMA GRAS data base (<https://www.femaflavor.org/>). “-” means not detected or below the detection limit. “Null” means there is no relevant threshold or OAV cannot be calculated.

Table 2. Contents of main taste substances in different teas

Taste substances ^a	Concentration(mg/g)		Significance	Concentration(mg/g)		Significance ^b	Taste description ^c
	HS	LS		HG	LG		
TPs	240.42±16.13	243.4±9.50	NS	200.65±13.17	220.28±10.86	NS	Bitter, astringent
AA	52.55±11.28	50.9±6.93	NS	47.43±9.15	42.75±6.83	NS	Fresh, sweet
CA	46.67±1.97	49.8±3.01	NS	22.84±2.64	43.85±1.12	*	Bitter
SS	40.16±2.64	25.63±1.75	*	53.44±1.73	29.10±2.23	*	Sweet
The	22.45±0.72	21.97±2.41	NS	15.59±1.1	15.32±0.82	NS	Fresh, sweet
Asp	5.42±0.24	5.16±0.17	NS	3.1±0.15	2.01±0.08	NS	Fresh, slightly sour
Asn	2.45±0.27	2.33±0.14	NS	0.43±0.08	0.49±0.02	NS	Fresh, slightly sour
Glu	6.05±0.19	5.77±0.42	NS	3.04±0.27	2.422±0.08	NS	Fresh, slightly sour
Gln	2.03±0.08	2.43±0.06	NS	2.15±0.19	1.30±0.07	*	Fresh, slightly sour
Ser	1.05±0.03	0.96±0.08	NS	0.77±0.27	0.53±0.11	NS	Sweet
Ala	0.36±0.01	0.35±0.03	NS	0.32±0.18	0.20±0.05	NS	Sweet
Phe	0.82±0.01	2.2±0.04	*	0.14±0.03	0.73±0.04	**	Bitter
Arg	1.07±0.04	1.12±0.02	NS	0.71±0.02	1.88±0.19	*	Bitter
EGCG	113.72±10.84	121.73±8.24	NS	91.6±9.68	109.92±11.26	NS	Bitter, astringent
GCG	4.04±0.17	7.58±0.98	*	12.69±1.37	14.47±0.81	NS	Bitter
ECG	22.02±5.57	28.38±2.34	NS	28.96±3.39	31.16±1.99	NS	Bitter, astringent
EGC	8.82±1.58	6.83±1.17	NS	27.59±1.68	15.62±2.64	*	Bitter, slightly sweet
EC	6.12±0.95	5.99±0.57	NS	8.85±0.87	10.51±0.57	NS	Bitter, slightly sweet
C	1.41±0.06	1.4±0.03	NS	1.38±0.15	1.12±0.24	NS	Bitter, slightly sweet

^a TPs, Tea polyphenols; AA, Amino acids; CA, caffeine; SS, Soluble sugar; The, Theanine; Asp, Aspartic acid; Asn, Asparagine; Glu, Glutamic acid; Gln, Glutamine; Ser, Serine; Ala, Alanine; Phe, Phenylalanine; Arg, Arginine; EGCG, Epigallocatechin gallate; GCG, Gallic catechin gallate; ECG, Epicatechin gallate; EGC, Epigallocatechin; EC, Epicatechin; C, Catechin.

^b Significance: NS means no significant difference between samples; *Significantly different at $p \leq 0.05$; **Significantly different at $p \leq 0.01$.

^cThe source of taste description: FEMA GRAS data base (<https://www.femaflavor.org>).

Table 3. Contents and pollution index of heavy metals and pesticide residues in tea

Risk elements	Contents (mg / kg)				Limit value ^a (mg / kg)	Pollution index ^b			
	HS	LS	HG	LG		HS	LS	HG	LG
heavy metal									
Cr	1.650±0.712ab	1.724±0.294a	1.940±0.481a	1.331±0.093b	5.000	0.330	0.345	0.388	0.266
As	0.074±0.017b	0.070±0.024b	0.033±0.007a	0.060±0.017b	2.000	0.037	0.035	0.017	0.030
Cd	0.065±0.005c	0.026±0.005a	0.035±0.001b	0.057±0.008c	1.000	0.065	0.026	0.035	0.057
Pb	0.708±0.015a	1.835±0.501b	0.554±0.008a	0.716±0.247a	5.000	0.142	0.367	0.111	0.143
Cu	14.083±1.547a	17.989±0.271b	13.881±0.462a	17.731±1.385b	60.000	0.235	0.300	0.231	0.296
Pesticide residues									
Imidacloprid	0.001±0a	0.005±0.002b	0.001±0a	0.008±0.004b	0.500	0.001	0.005	0.002	0.012
Acetamiprid	-	0.002±0a	-	0.015±0.001b	10.000	Null	Null	Null	0.002
Carbendazim	-	0.006±0a	0.009±0b	0.021±0.003c	5.000	Null	0.001	0.004	Null
chlorfenapyr	-	-	-	0.470±0.094	20.000	Null	Null	Null	0.024
Buprofezin	-	-	-	0.500±0.137	10.000	Null	Null	Null	0.050
Chlorpyrifos	0.086±0.015b	0.170±0.021d	0.130±0.009c	0.042±0.011a	1.000	0.086	0.170	0.130	0.042
Cypermethrin	0.046±0.002a	0.045±0.009a	0.047±0.009a	0.072±0.010b	20.000	0.002	0.002	0.002	0.004
lamda-cyhalothrin	0.044±0.006a	0.041±0.012a	0.080±0.024b	0.130±0.032c	15.000	0.003	0.003	0.005	0.009
pyridaben	-	0.010±0.003a	0.009±0.002a	0.890±0.271b	5.000	Null	0.002	0.002	0.178
Bifenthrin	-	0.039±0.006a	0.026±0.005b	0.904±0.316c	5.000	Null	0.008	0.005	0.380

^aLimit value: The limit value of heavy metals in tea comes from China National Standard: limit of pollutants in food (GB 2762-2017); The pesticide residue limit value of tea comes from the Chinese national standard: maximum residue limit of pesticide in food (GB 2763-2019).

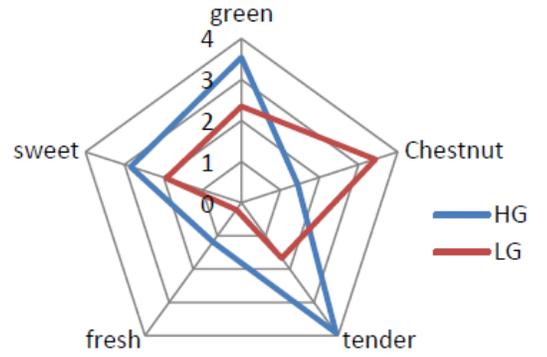
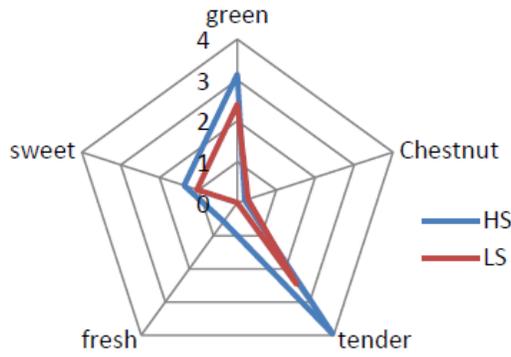
^bPollution index=w/ws, w is the content of element, ws is the limit value of element.

Table 4. Investment in prevention and control of diseases and pests in tea garden in experimental area

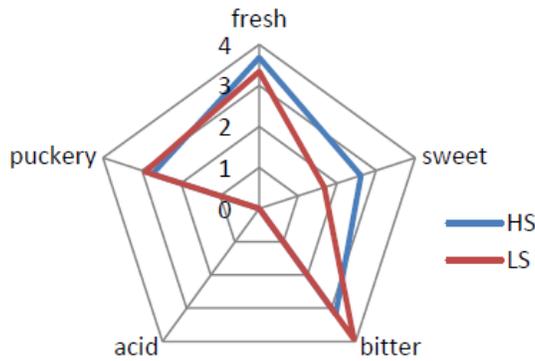
Tea district	Are pesticides used here (%)		Frequency of pesticide (times / year)	Is there any plant protection personnel (%)		Damage rate of tea shoots (%)
	Yes	No		Yes	No	
High mountain	33.33	66.67	2~5	16.67	83.33	17.54
Low mountain	100	0	3~8	85.71	14.29	6.27

Figures

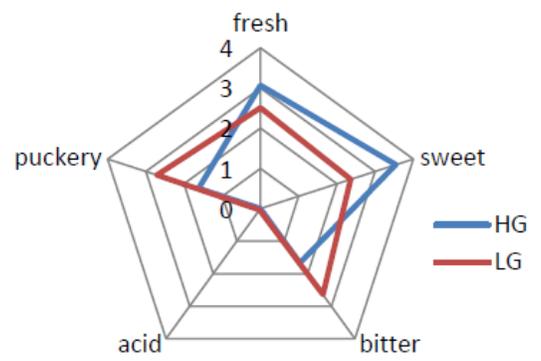
A Aroma profile



(c) Aroma profile of Shihua



(d) Aroma profile of Ganlu



B Taste profile

(a) Taste profile of Shihua

(b) Taste profile of Ganlu

Figure 1

Flavor profile of different tea. "HS", High mountain Shihua, "LS", Low mountain Shihua, "HG", High mountain Ganlu, "LG", Low mountain Ganlu.

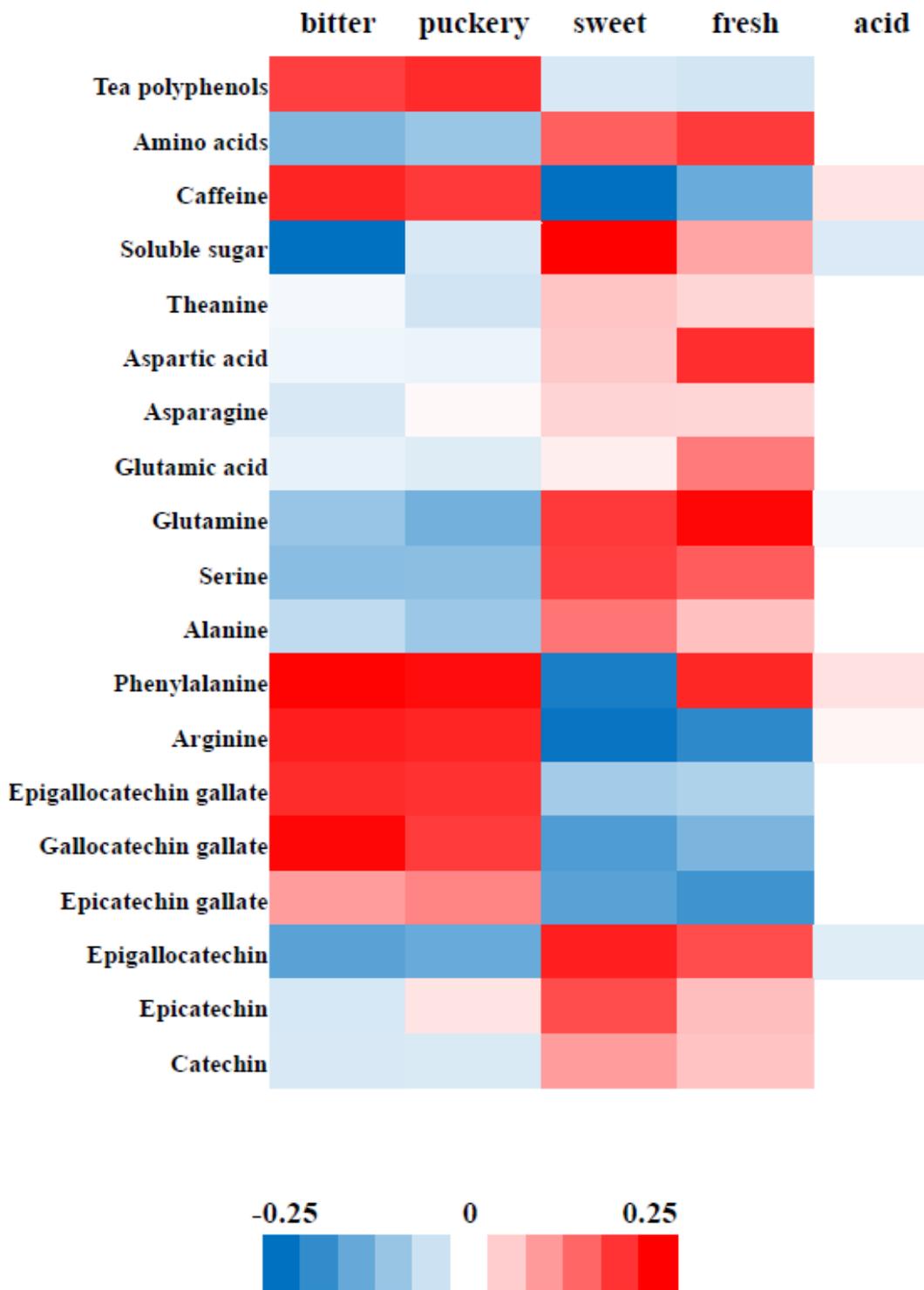
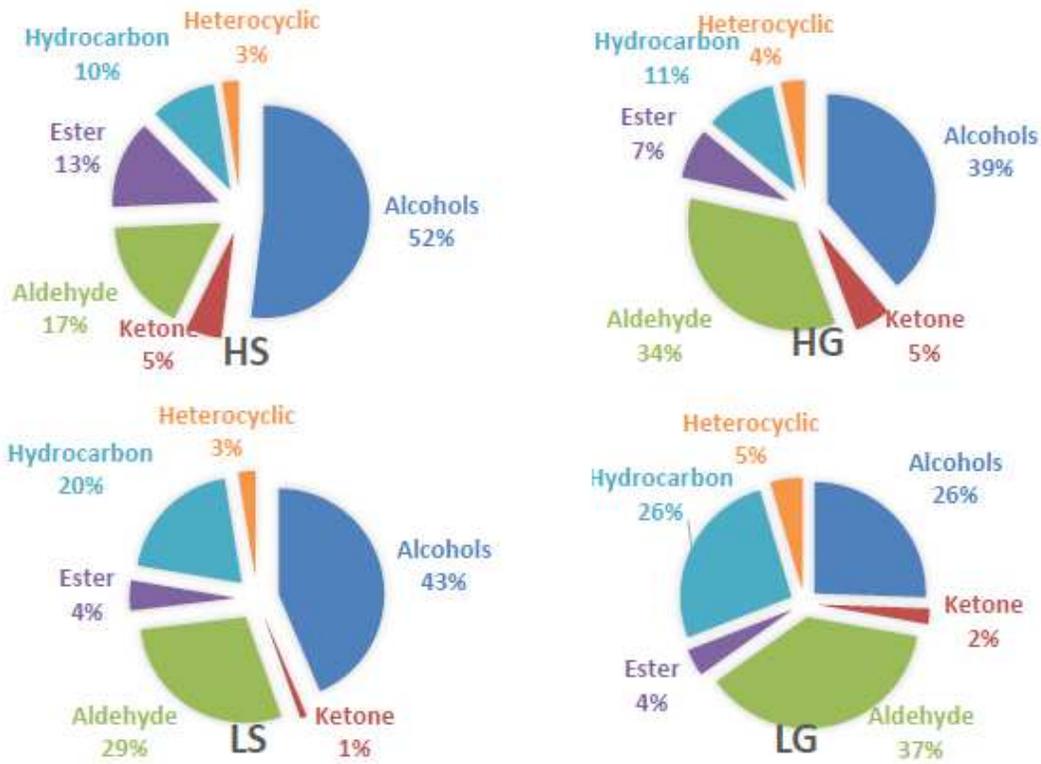
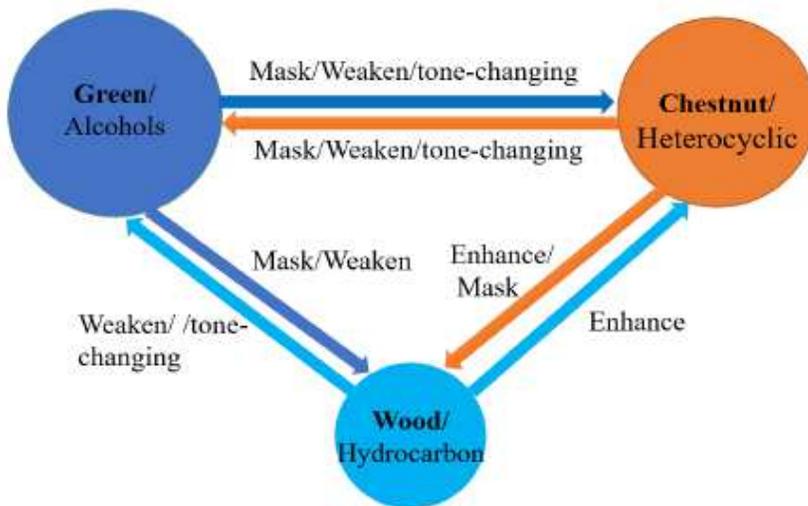


Figure 2

Correlation between main taste substances and sensory characteristics of HT. Here, we use the standardized coefficient of partial least squares regression (PLSR) to analyze the contribution of 20 tea flavor substances to 5 tea flavor attributes



A: Types and proportions of aroma compounds in different types of tea.



B: Possible relationship between aroma compounds in tea

Figure 3

The proportion of aroma compounds in HT and LT and the possible relationship between them

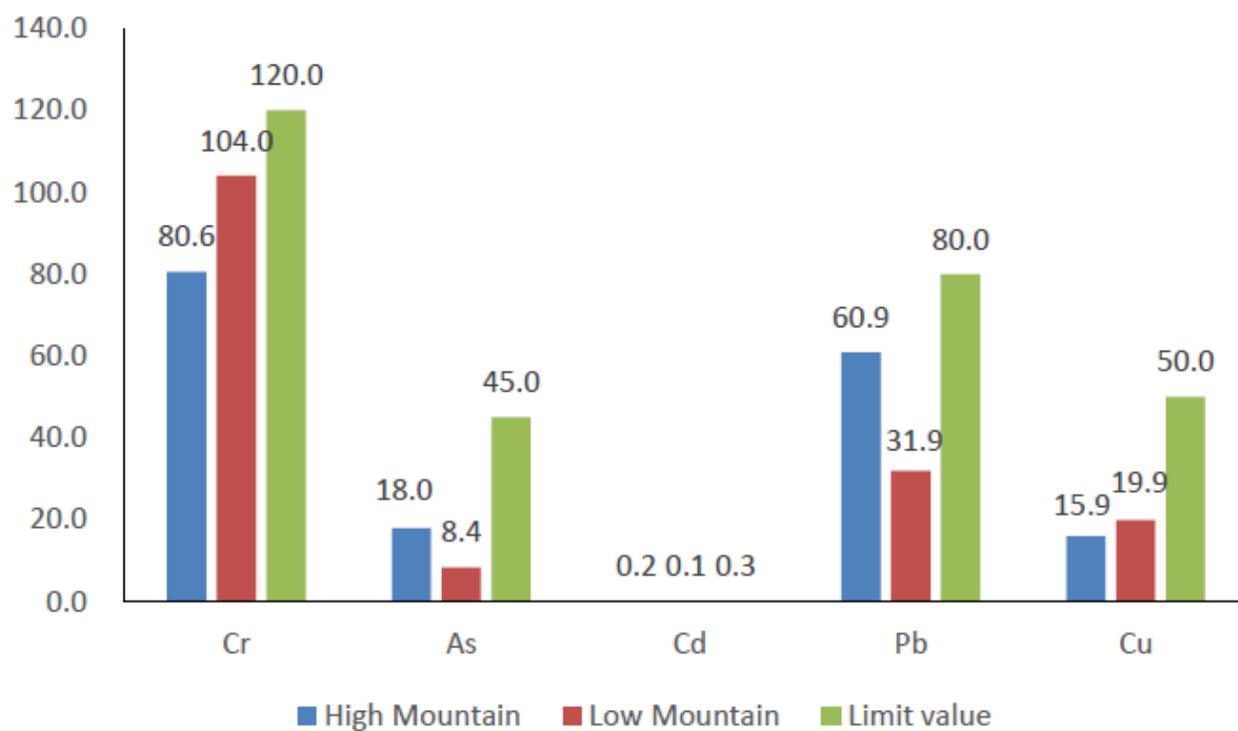


Figure 4

Heavy metal content of tea garden soil in different areas. The data come from our research group. The limit value comes from the Chinese agricultural standard: Environmental conditions of producing areas of pollution free agricultural products (NY5010-2016).

