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Enhancing the Smell of Green Tea with the Use of both Natural and Tea Fertilisers

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Abstract

One of the best vital agricultural performances toward boosting Tea Plant (TP) yield is the use of natural fertilizer. Unfortunately, each nutritional desire of the TP could not be met by a single organic fertilizer. Organic fertilizer and tea fertilizer with an appropriate nutrient ratio are the best ways to meet tea plant nutritional needs. The physical excellence of Green Tea (GT) will be tested over four years using four fertilizations: tea-specific fertilizer such as + urea + colza cake, tea-specific fertilizer + urea + livestock waste compost, and tea-specific fertilizer + urea, combined tea-specific fertilizer. By increasing the amount of EGCG, ECG, caffeine, tea polyphenols, along with the water extract in GT, the fertilizer made just for tea could make it taste better. The levels of" D-limonene, cis-jasmone, nonanal, linalool, cis-3-hexenyl hexanoate, along with cis-3-hexenyl benzoate, which give GT its smell, increased significantly after organic fertilizer and tea-specific fertilizer were employed. This combined fertilization, however, needed a weaker consequence on growing the quality of GT's aroma. By combining fertilizer designed specifically for tea with an organic fertilizer in tea orchards, this study offers a theoretical foundation for prudent fertilizing.

Keywords: Aroma, Sensory Quality, Green Tea (GT),Tea-Specific Fertilizer

Full length article **Corresponding Author*, e-mail: v.ruby@jainuniversity.ac.in

1. Introduction

The greatest consumed drinks worldwide are tea, yet little is known about how sustainable it is for the environment. Throughout the past few decades, economic growth has been accompanied by a steady increase in the consumption of tea worldwide. With 35 l, tea consumption per person is currently second only to packaged water, and it is anticipated to increase considerably more shortly, especially in big countries like China and India. The tea industry has the potential to be an energy along with emission-intensive one owing to the significant usage of agrochemical inputs during tea growing and the use of traditional energy (such as coal and firewood) during tea processing [1].Few studies have looked at the benefits of smelling Green Tea (GT), even though some tea varieties with particularly strong flavors—like black tea and herbal tea—have been studied about their physical and psychological benefits of odor. Some smelly parts of GT have been shown to make people feel less stressed. For instance, an investigation into the impact of the fragrance of green on the brain discovered that the components of the fragrance assisted the brain's production of dopamine, then regulates just how well the brain functions, mood, as well as consideration.(E)-2-hexenal, (z)-3-hexanol are some of the major things that give GT its smell [2].The environment can also affect tea plants of the same cultivar. Since mountainous regions are where tea plants are primarily produced, variations in elevation can have a substantial impact on tea quality. The growth of tea leaves would be sped up by higher temperatures and less mist at lower altitudes. Yet, tea growers and tasters frequently link heavier mist at higher altitudes with higher-quality tea. There are conflicting reports regarding how cultivation altitudes affect catechin content, which calls for a deeper, more thorough investigation using more potent techniques [3].In 2016, India had about 2.90 million ha of tea plantations, and the country made more than 2.4 million tonnes of tea. Tens of thousands more tea farmers will immediately see a decrease in income due to changes in crop growth quantity and quality. Traditionally, higher fertilization rates along with pesticide use have been consumed to improve these factors. Research based on interviews with tea agriculturalists has exposed that the normal quantity of nitrogen fertilizer used on GT plantations in India is about 521 kg/ha. This is a lot more than what is needed [4]. Volatile substances that the olfactory epithelium detects make up an aroma. Only roughly 300 of the approximately 700 volatile chemicals found in tea—with a concentration of 0.012-0.13% GT—were found. The amount of GT is directly affected by the way it is grown and when it is picked. One of the most significant ways for farmers to boost crop yields has been through the usage of fertilizer. Nonetheless, growers have gradually started using chemical fertilizers due to the rising market demand for tea. Long-term tea farming has been linked to several problems with land degradation, such as soil acidity, structural damage, loss of nutrients, and lack of good bacteria [5].

The study [6] looked at how adding organic fertilizer affects the tea plant's metabolites, especially changes in volatile organic compounds (VOCs), and how that affects the tea plant's microbiome. Through the investigation of real-time aromatic features occurring during tea brewing, it also sought to develop a hypothetical substance designed for the parameter of GT aromatic value. The study [7] looked at the polyphenols, caffeine, and antioxidants, along with the cup quality of clones grown with various amounts of nitrogen. The goal was to find the best clones and processing methods for making antioxidant-rich GT that can be drunk every day. The article [8] discussed potential markers that could be used to choose the best tea cultivars to grow and the raw components to make oolong tea. It is described how to quickly and precisely screen potential cultivars using distinctive scent components. In a two-year field experiment, the study [9] examined the possible impacts of topically applying nano selenium to tea plants that had been exposed to oxidative stress brought on by pesticides. After adding 10 mg/L of nano-Se, the quantity of protein, soluble sugar, carotene, polyphenols in tea, and catechins was all much higher than they were before. Theanine, glutamic acid, and proline, coupled with arginine production are anticipated to increase as a result of the altered GS-GOGAT cycle. During a two-year field research, soil samples from tea plantations with a pH of 5.09 and significant levels of lead (Pb) and cadmium were taken (Cd). Additional soil amendments included shellfish and organic fertilizers. This means that in the future, tea farms whose soils have become too acidic could use the best shellfish amendment as a technological fix to improve the soil and tea quality [10].This study looked at how agroecological tea management approaches affected the quantity and quality of tea, soil health indices, and tea producers' net income. By employing agroecological management strategies, the paper [11] showed that soil natural substance rose by 0.9% and soil pH by an average of 0.5 units. In Fengcheng, a rural region of India with a concentration on the tea business, the article [12] examined how local farmer knowledge is developed and how this aids some people and rural places in maintaining their agricultural resilience. By examining the actual aromatic properties of GT in the present work, a theory-based reference technique is presented [13].The first step in understanding how the volatiles in the headspace interacts with the tea distillation, which is their counterpart, was determining the optimal time to manage the aroma's quality. The study [14] investigated how applied magnesium (Mg) affected the leaf quality and aroma of Wuyi Rougui tea seedlings in hydroponic environments. Throughout a five-year field experiment in which N, P, and K fertilizers were sprayed collectively, soil as well as plant samples were collected to learn more about how various combinations of these fertilizers affect soil fungus and tea. Compared to NPK, NK or NPK, NP, NPK, and PK had a significantly lower yield and inferior quality [15].

2. Materials and methods

2.1Experimental Design

The investigational tea garden which is in Jinjiapu town, Huanggang City. The 131-teavariability has grown for five years. It was adapted for processing GT and developed from a population of the Mingshan highlands in India. The four fertilizer treatments used in the experiment, which took place between 2016 and 2020, are displayed in Table 1. The grade for the colza cake was 4.6-2.48-1.4, for the tea-specific fertilizer was 18-8-12 (N-$P_2O_5K_2O$), and for the animal, waste compost was 1.68-2.38-1.3. The control plots (CK) were left unfertilized. Plots had a 30 m^2 plot size, were repeated three times, and were random illilitersy pooled. The tea garden was evenly managed throughout the region with routine ploughing, weeding, and the application of deep fertilizer. Beginning in November of each year in 2016 through 2020, the base fertilizer was applied, and beginning in March of each year, the top dressing was done. Colza cake, composted animal waste, a fertilizer specifically for tea, and urea were all employed as the foundation fertilizers. In April 2020, single buds in conjunction with dual leaves of spring shoots, each weighing one kilogram, were chosen at random and processed utilizing the conventional methods of withering, fixing, and progressing, along with the ventilation in the three repeating plots of each treatment.

2.2 Sensory Evaluation

Exactly, 5.1 g of GT were combined by250millilitres of steaming purified water following the Tea Sensory Assessment Method in GB/T 23776-2018, as well as the tea infusion was drained after 5 minutes. Five experts used a 100-point scale to evaluate the look, colour, smell, taste, and infusion quality of the leaves. Out of a possible 100 points, 25% were given for the colour of the dry tea, 10% for the colour of the brewed tea, 25% for the smell, 10% for the brewed leaves but also 30% for the flavour.

2.3 Analysis of the Main Parts

Using this 120 °C-drying procedure, the amount of tea water extract in the sample was determined (GBT 8305-2013).After 45 minutes of boiling 250 millilitres of water at 100 °C, each tea powder (2 g) was taken out by a suction filtration system (while shaking every 10 minutes). The tea powder was cooked for 1 hour at 120 °C, cooled for 45 minutes, along with the then baked for another hour. For the GB/T 8314-2013 ninhydrin colorimetric procedure, the filter was put into 500 millilitres of water, along with volumetric flask was enhanced to the flask to make it the right size. In a 25-milliliter colorimetric tube, 1millilitreof the test substance, 0.4millilitres of phosphate bufferas well as 0.4milliliters of a 2% w/v ninhydrin solution were located. The tube was tested for colour at 570 nm after being heated in a bath of boiling water for 15 minutes with 25 milliliters of water added.

The Folin-Ciocalteu reagent and the colorimetric technique (GB/T 8314-2013) were used to determine the overall quantity of polyphenols throughout the tea. The actions were as follows: Twice, 6millilitres of 90% v/v methanol were used to extract 0.2 g of tea powder for 10 min each time at 70 °C. After that, the tube was spun at 3500 rpm for 10 mins. When the extracts were mixed, 10 millilitres of 70% v/v methanol were added. 100 millilitres of purified water were improved to 1millilitre of the solution to make the test liquid. 2 milliliters of the experimental liquid, 6 milliliters of 20% v/v Folin-Ciocalteu reagent, and 5 milliliters of 8.5% w/v Na2CO3 were combined and let react for 60 minutes to determine the amount of tea polyphenols present. The value of the absorbance was calculated using a 765 nm wavelength A separate procedure involved adding 2.0 milliliters of the specimen's extract to a 10 milliliter volumetric flask, diluting it to volume with a stabilized solution, and then filtering it through a 0.45 millimeter membrane. High-Performance Liquid Chromatography (HPLC) systems were utilized toward look at each catechins and caffeine. A 4.6 mm by 150 mm Waters C18 column was used to do HPLC at $50^{\circ}c$. The examplesbe eluted by a gradient of solvents A and B, going from 0% to 100% A to 0% to 10% to 68% A to 32% B to 0% to 30% to 100% A. Catechins were discovered using a 280 nm UV detector.

2.4 Analysis of the volatile substances in tea

This approach for evaluating the volatile chemicals in tea was carried out following this study. Before each extraction, the SPME fibre was put in the GC instrument's injection port for 10 minutes at 280°C to get rid of any volatile chemicals that were left over. The dried tea sample and 30 millilitres of boiling water were put into a 100 millilitres vial, which was then sealed with a silicone septum. Then, Add 20 liters of an inner standard solution right away. After settling for 15 minutes in a water bath, the sample was kept at 50 °C with the SPME fiber exposed to the headspace for 45 minutes. Once the fiber has been inserted into the GC injector port, it was heated for three minutes at 240 °C to remove the chemicals. For the study, an Agilent 6785A GC was linked to an Agilent 6589 C MSD ion Trap MS. The signal-to-noise ratio had to be three for detection. Constructed onan internal consensus solution, the volatile chemical concentration was determined in$\frac{\mu g}{l}$.

2.5 Statistical analysis

Three tea samples were taken from the repeated plots and averaged out to produce the findings. To find significant differences between means, SAS JMP version 9.4 was used to run Duncan's multiple range tests, which are a type of one-way ANOVA. The statistical significance was judged by whether or not the p was less than 0.05. GraphPad Prism 9.0 was used to do a principal component analysis (PCA). Origin 9.0 was used to make the figures (Demo version, Northampton, Massachusetts, USA). TB tools were used to create the heat map.

3. Results and Discussions

3.1Analysis of Sensory Qualities

Table 2 gives an example. The characteristics of M1 and M2 that received 97 uses for appearance and quality were fine, tight, bending, slightly tippy, and green bloom. The unfertilized (CK) samples scored the lowest, receiving only 83 points for their "dull green," dry tea-colored appearance. "Green, vivid" was mostly used to describe the brew's hue. The remarks on smell were "high aroma" in M1 in addition to M2, with M1 receiving the highest overall score and the flavor being described as "heavy, mellow, and brisk". The taste ratings for the unfertilized (CK) samples were lower and they were considered to be brisker and fresher. The four samples' total scores fell within the range of 74.25 to 97.20. Overall, the GT made from M1 had the best quality and score, with more than 97 points. M2 and M3 were second and third, respectively.

3.2 Main Quality Components Flavor

Table 3 provides the levels of the primary flavor-enhancing ingredients, such as total catechins, acids, caffeine, as well as tea polyphenols. All soluble compounds, comprising polyphenols, pectin, sugars, alkaloids, or others, are present in tea leaf extracts. Compared to M2 (49.93%) and CK, M1 (52.92%) and M3 (53.52%) had considerably greater water extract levels (51.10).The building blocks of flavor are amino acids, one of GT's most significant chemical constituents but a key element in the beverage's crisp and snappy flavor. M1 had the highest free amino acid content (3.97%), followed by M3 (4.74%), M2 (4.55%), and CK (4.09%), with no discernible modification among M3 with the other two fertilization methods like M1 and M2. Between the fertilized and non-fertilized treatments, there was a significant variation in caffeine concentration, but not between the fertilizations three techniques M1, M2, and M3.

Tea quality and Tea polyphenols were tightly connected. These are the primary ingredients that make up the tea; they serve a purpose and are also the substances that most closely resemble the bitterness of thetea. Mathematical analysis showed that while fertilization treatments M1,M3, as well as M1,M2, were not statistically different from those of tea cultivated without fertilization, they were not significantly different either (CK).

Catechins, which make up between 70% and 80% of all tea polyphenols in tea plants, dominate the category of tea polyphenols. This fertilization treatment had a significant impact on both the catechin composition and content. Total catechin content was distributed as follows: M3 (25.18%) > M1 (23.90%) > M2 (24.90%) > CK (24.66%).M3 was considerably different from the other treatments ($p<0.05$), although M1 and M2 showed no difference. The fertilization methods had a big impact on catechin content. The findings (Figure 1) showed that the majority of catechins, including (+)-catechin, (_)-epigallocatechin gallate (EGCG), (_)-gallocatechin 3-O-gallate (GCG), and (_)-epigallocatechin (EGC), did not change significantly between M1, M2, and M3. (C). However, in the fertilizations M1, M2, and M3, EGCG, (_)-epicatechin gallate (ECG), GCG, along with the EC were all considerably greater than in CK. M1 and M2 did not appear to differ much in the ECG's contents, but M2 were suggestively lesser than M3. Moreover, compared to the other three treatments, fertilization treatment M1 had significantly less ()-catechin gallate (CG).

3.3 Fertilization treatments' effects on volatile components

Tea flavor, quality assessment, and widespread consumption are all significantly influenced by the aroma. Four tea samples from various fertilization methods were analyzed using HS-SPME-GC-MS to determine the fragrance components (Table 4). The 37 components in all the GTs were the same, according to a comparison investigation. The major volatile substances were D-limonene (6.13~20.86 g/L), cis-jasmone (1.28~32.09 g/L), nonanal (4.93-09.05 g/L), linalool (2.46-09.73 g/L), cis-3-hexenyl hexanoate (1.37-9.56 g/L), -cadinene (2.56-6.66 g/L), indole (0.57-7.88 g/L), cis-3-hexenyl benzoate (2.67-5.60 g/L), eremophilene (2.50-5.46 g/L), cis-geraniol (2.90-4.83 g/L), methyl salicylate (2.36-6.86 g/L), nerolidol (1.45-5.66 g/L), and 2,4-di-tert-butylphenol (1.02-3.15 g/L). M1 (263.05 g/L), M2 (250.80 g/L), M3 (69.77 g/L), and CK (55.00 g/L) were the next highest concentrations along with total volatile components. Each fertilization treatment had a substantially greater overall odor than the no-fertilization condition. This was especially evident for M1 and M2, where the overall odor was approximately 4.0 and 5.5 times stronger than in CK. Compared to CK, M1, M2, and M3 also had more volatile substances. In contrast, neither M1 nor M2 included 1-octen-3-ol, cis-linalool oxide, 2-octenal, trans-ocimene, acetic acid, or phenylmethyl ester, and neither M3 contained copaene, 2-octen-1-ol, 2-heptenal, or 2-octenal. Additionally, neither M1 nor M2 contained trans-ocimene.

3.4 Treatments for Fertilization's Impact on Certain Classes of Volatile Substances

The detailed classification of the volatile compounds is displayed in Table 5 in addition to Figure 2. The 52 fragrance mechanisms discovered in all these examined models included ten distinct types of alcohols, thirteen different aldehydes, six different ketones, ten different alkenes, eight different esters, also eight other compounds. The amounts of the various types of aromata that were included in the overall substances of volatile compounds varied, but the order of the contents of each class of volatile compounds was M1 > M2 > M3 > CK (Table 5). (Figure 2). Alkenes accounted for the majority, with alcohols and esters coming in second and third. The monoterpene alcohols that are produced during the processing of tea (such as geraniol, linalool, and their oxides) usually contribute to a fruity, floral, and wine-like flavor. Linalool but also geraniols are considered to be important ways to judge the smell of tea. 15.20% of M3 and 16.18 % of M1 were made up of alcohol molecules.

3.5 PCA of Important Criteria in Leaf Extracts with Various Fertilization Procedures

A statistical technique called principal component analysis (PCA) can display an total data as a wide, qualitative visual that highlights the shared characteristics between within the samples. An examination using principal component analysis was carried out on the aspects of tea quality that were deemed to be the most significant (Figure 3).The findings indicated that the first principal component (PC1), which accounted for 73.81% of the difference, and the second principal component (PC2), which accounted for 16.04%, were both significant. The first two major variables together accounted for 89.85% of the difference. There was a clear divide between the three groups of the four tea samples (Figure 3).

Table 1: Different treatments' fertilizer types and dosages

Fertilization Model	N, P_2O_5,K_2O	Fertilizer Combination
M3	324-84-126 kg/ha	Tea–specific fertilizer 1050 kg/ha + urea 300 kg/ha
M2	320-131-129 kg/ha	Tea-specific fertilizer 750 kg/ha + livestock waste compost 3000 kg/ha + urea 300 kg/ha
M1	366-109-129 kg/ha	Tea-specific fertilizer 900 kg/ha + colza cake 1500 kg/ha + urea 300 kg/ha
CK		Unfertilized

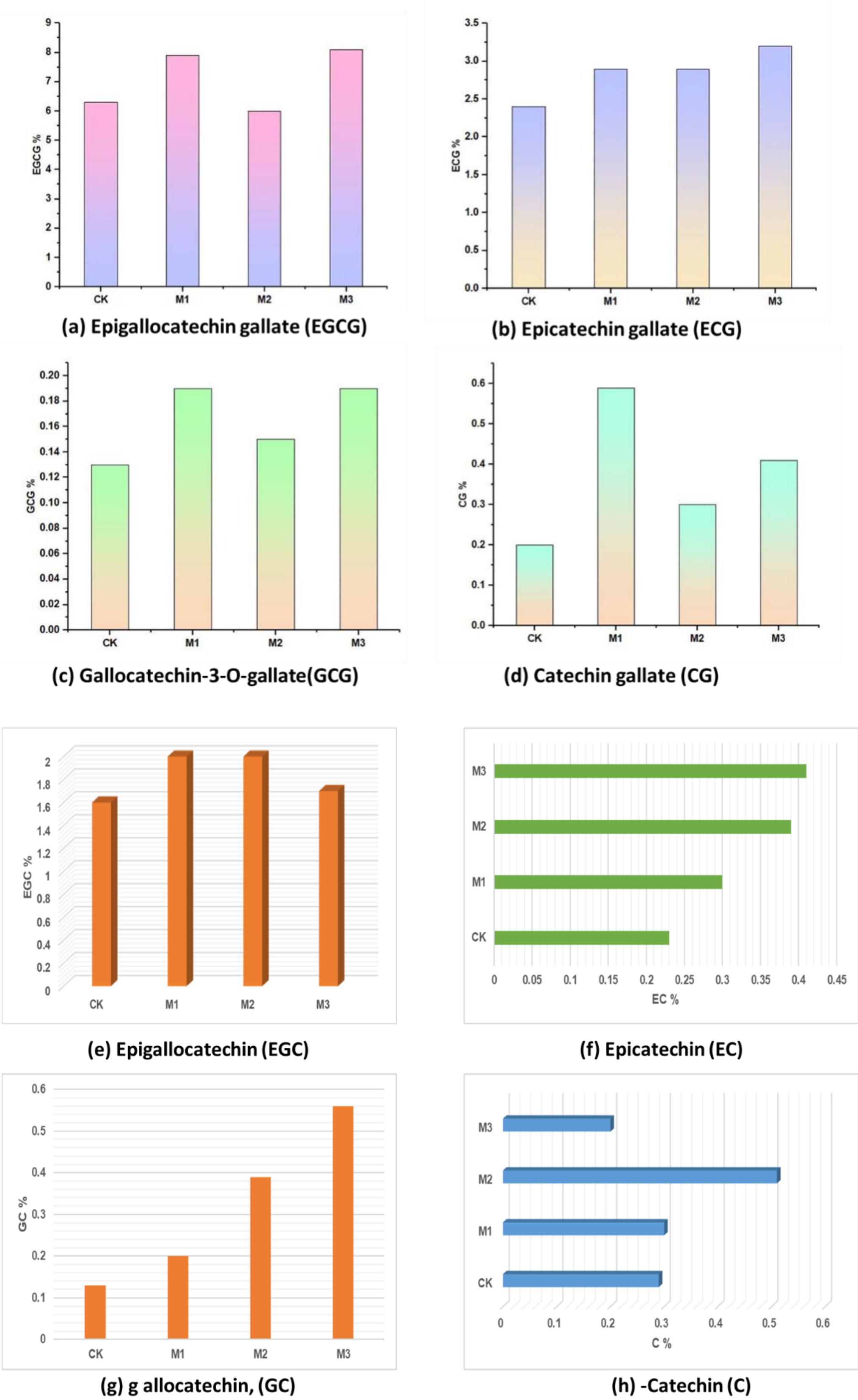

Figure 1: The impact of various fertilization methods on catechin content.Each value is the mean standard deviation of three samples of tea. The (a-c) above the bars are statistically different.

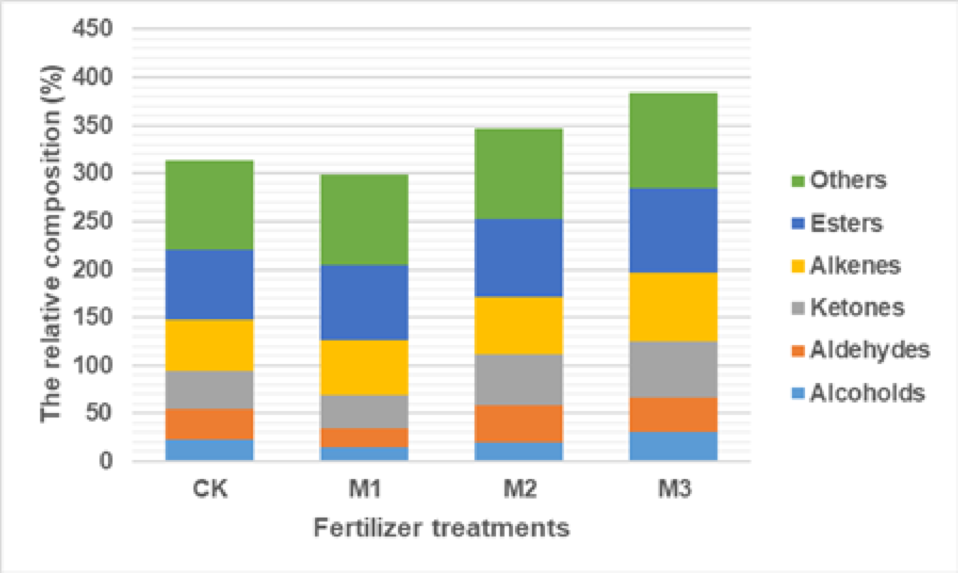

Figure 2: This overall volatile component concentration of tea samples grown with four different types of fertilizers took into account how much of each group of volatile compounds was present. Each number is the mean standard deviation of 3 tea samples.

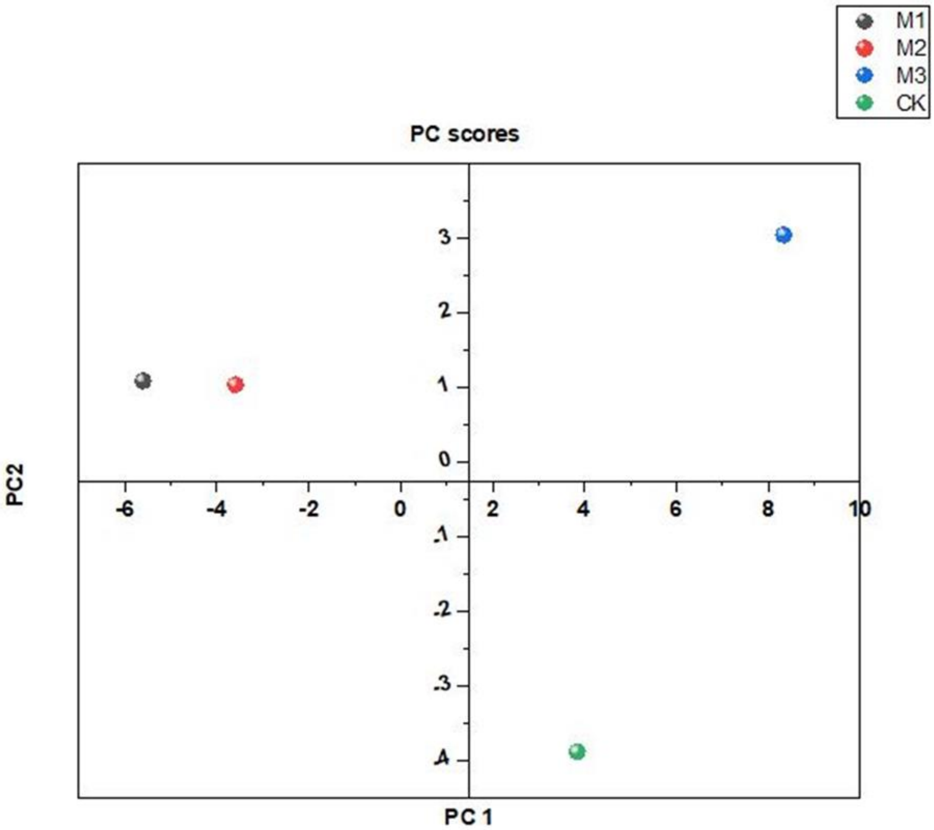

Figure 3: The score plot of components based on PCA 1 and 2 and the loading plot of PC1 vs PC2

Table 2: Evaluation of the taste and smell of tea samples made with different fertilization

Fertilization Treatment	Appearance (25%)		Infusion Color (10%)		Aroma (25%)		Taste (30%)		Infused Lest (10%)		Total Score
	Comment	Score	Comment	Score	Comment	Score	Comment	Score	Comment	Score	
CK	tight, bent, fairly tippy, dull green	83	light yellow	82	Pure and normal	83	Relatively fresh and brisk	85	Soft	83	83.50
M1	fine, tight, bent, slightly tippy, green bloom	88	clear green, bright	82	High aroma	90	Heavy,mellow,and brisk	89	Soft and even	89	88.80
M2	fine, tight, bent, slightly tippy, green bloom	88	green, bright	85	High aroma	89	Sweet and brisk	88	Soft and bright	88	87.95
M3	fine, tight, bent, fairly tippy, green bloom	85	clear green, bright	87	Clean and pure	88	Sweet and brisk	88	Soft and bright	88	87.15

Table 3: Impact of various fertilization methods on the percentages of total catechins, polyphenols, caffeine, free amino acids, and water extract in GT

Mode	Water Extract	Free Amino Arid	Caffeine	Tea Polyphenols	Total Catechins
CK	49.0±0.66 b	3.10±0.20 d	3.02±0.20 b	17.38±0.19 c	12.57±0.28 c
M1	52.92±0.08 a	3.97±0.14 ab	3.35±0.04 a	18.70±0.19 ab	13.89±0.18 b
M2	49.93±0.32 b	3.66±0.09 c	3.35±0.09 c	18.27±0.52 b	13.81±0.49 b
M3	53.52±0.06 a	3.83±0.09 bc	3.83±0.09 bc	19.46±0.50 a	15.17±0.38 a

Table 4: Impact of various fertilization methods on the fragrance compound content ($\mu g/L$)

No	Aroma Component	CK	M1	M2	M3
1	D-Limonene	7.22±0.54 d	30.97±1.32 a	24.52±1.21 b	12.21±0.66 c
2	Cis-jasmone	2.18±0.05 d	21.10±2.43 a	17.09±2.12 b	6.88±0.22 c
3	Nonanal	3.82±0.32 c	13.04±1.22 a	12.01±1.10 a	6.76±0.33 b
4	Limalool	3.37±0.12 c	11.62±1.03 a	10.76±1.13 a	6.55±0.43 b
5	Cls-3-Hexenyl hexanote	2.26±0.23 d	8.47±0.97 a	6.59±0.74 b	4.01±0.56 c
6	δ-Cadinene	3.67±0.36 c	7.39±1.32 a	7.78±1.21 a	5.39±0.91 b
7	Indole	0.79±0.01 c	6.91±0.98 a	7.06±0.74 a	1.29±0.03 b
8	cis-3-Hexenyl benzoate	1.78±0.4 c	6.70±1.11 a	6.54±0.62 a	2.97±0.03 b
9	Eremophilene	1.40±0.10 c	6.15±1.21 a	6.34±0.0.97 a	2.21±0.83 b
10	cis-Geraniol	1.89±0.19 c	5.51±0.35 a	5.76±0.56 a	4.83±0.74 b

Table 5: The total amount of different smell types present in GT samples following four fertilizer applications $(\mu g/l)$

Volatile Compounds	CK	M1	M2	M3
Alcohols	10.36±1.23 c	24.67±2.31 a	24.84±1.69 a	19.29±1.53 b
Aldehydes	6.41±0.26 c	23.09±1.48 a	20.25±1.24 a	10.19±0.33 b
Ketones	3.90±0.11 c	27.58±2.16 a	23.57±1.59 a	8.71±0.44 b
Alkness	13.92±0.96 d	52.19±3.11 a	45.73±2.96 b	23.37±1.83 c
Esters	6.70±0.13 c	26.11±1.67 a	22.45±1.38 a	12.66±0.81 b
Others	2.71±0.14 c	18.41±1.15 a	16.06±0.94 a	5.33±0.06 b

4. Conclusions

This study found that tea-specific organic fertilizer improved the taste of water extract, caffeine, EGCG, and ECG. When tea-specific fertilizer was mixed with organic fertilizer, GT tea scent components such D-limonene, cis-jasmone, nonanal, linalool, cis-3-hexenyl hexanoate, and benzoate increased significantly. By applying organic and tea-specific fertilizer to tea plantation soils, GTs was valued more. Plantations produced more GTs.

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