

Tropical Journal of Phytochemistry & Pharmaceutical SciencesAvailable online at <https://www.tjpps.org>**Original Research Article****Microbial Quality and Sensory Evaluation of Infusions of a Polyherbal Tea Blend Formulated for Menstrual and Arthritic Pains**Victor U. Olugbue^{1*}, Segun S. Ogundapo², Stella E. Obasi¹, Ibukun C. Vining-Ogu¹, Chintua E. Igara¹, Joseph B. Suleman¹, Garba J. Danladi¹, Kerian C. Ngobidi¹, Onochie J. Nkama¹, Damian Uchendu¹, Fidelis I. Nsude³, Chibuike Kalu⁴¹Department of Science Laboratory Technology, Akanu Ibiam Federal Polytechnic Unwana Afikpo Ebonyi State, Nigeria²Department of Pharmaceutical Technology, Akanu Ibiam Federal Polytechnic Unwana Afikpo Ebonyi State, Nigeria³Department of Mathematics and Statistics, Akanu Ibiam Federal Polytechnic Unwana Afikpo Ebonyi State, Nigeria⁴Department of Food Science and Technology, Akanu Ibiam Federal Polytechnic Unwana Afikpo Ebonyi State, Nigeria**ABSTRACT**

Consuming herbal tea may promote health due to its antioxidant, anti-inflammatory, and antibacterial properties. However, contamination with objectionable microorganisms may pose health risks. This study evaluated the microbial quality and sensory properties of a polyherbal tea formulated from dried green and black tea (*Camellia sinensis*) leaves, ginger rhizomes (*Zingiber officinale*), and clove buds (*Syzygium aromaticum*) for the management of menstrual and arthritic pain. The plant materials were powdered, blended in different proportions, and used to prepare six formulations (F1–F6). The microbial safety of the infusions was evaluated in accordance with British and European Pharmacopoeia specifications, and moisture content was determined at 105°C using a drying oven. Sensory evaluation was conducted using a 7-point hedonic scale. All formulations contained bacteria and fungi within acceptable limits. Bacterial counts ranged from $0.33 \pm 0.08 \times 10^2$ in F6 to $4.70 \pm 1.11 \times 10^2$ CFU/g in F4, while total yeast and mould counts ranged from $2.00 \pm 0.35 \times 10^2$ in F1 to $6.17 \pm 1.33 \times 10^2$ CFU/g in F3. The infusions were free from *Salmonella* and *Escherichia coli*. Identified bacteria included *Staphylococcus aureus* (45.45%), *Bacillus* sp. (32.73%), and *Micrococcus* sp. (21.81%). Fungal isolates included *Penicillium* sp. (37.84%), *Aspergillus* sp. (22.97%), and *Cryptococcus* sp. (19.92%). Moisture content ranged from $2.84 \pm 0.02\%$ to $7.11 \pm 0.87\%$ w/w. Formulation 1 showed the best sensory attributes. The findings indicate that the plant materials are suitable for developing an acceptable herbal tea.

Keywords: Green tea, Clove, Anti-inflammatory, Microbial, Sensory quality.

Received 09 August 2026

Revised 17 August 2026

Accepted 18 August 2026

Published online: 13 September 2026

Copyright: © 2026 Olugbue *et al.* This is an open-access article distributed under the terms of the [Creative Commons Attribution License](https://creativecommons.org/licenses/by/4.0/), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.**Introduction**

The Consumption of tea made from dried leaves of *Camellia sinensis* has increased worldwide due to its natural origin and perceived health benefits. More than half of the world's population drinks herbal tea regularly.¹ Tea varieties can be differentiated by their processing methods: black tea is fully fermented, green tea is unfermented, while oolong tea is partially fermented.² Tea is rich in bioactive compounds like polyphenols, alkaloids, amino acids, polysaccharides, and saponins, among others.^{3,4} Some of the benefits of herbal tea consumption include antioxidant, anti-inflammatory, antimicrobial, immunoregulation, cardiovascular health promotion, obesity prevention, liver protection, diabetes prevention, and anticancer.^{3,5-7} Herbal tea may comprise one or more plant materials brewed as decoctions or infusions and drunk for their therapeutic, pharmacological, and prophylactic benefits.^{8,9}

In Northern Nigeria, green and black tea made from *Camellia sinensis*, ginger (*Zingiber officinale*), and clove (*Syzygium aromaticum*) are blended into a traditional beverage and consumed as a pain reliever for arthritic and menstrual pain and for general wellness, without scientific validation. Hence, there is a need to evaluate the microbial quality of polyherbal tea as an indicator of the quality and safety of consumers.¹⁰

The tea plant, *Camellia sinensis*, is a member of the family Theaceae and is believed to have originated in Southeast Asia. It is currently cultivated across more than 30 countries worldwide.¹¹ Tea is consumed in Africa, partly because of its natural origin and the numerous pharmacological benefits attributed to its bioactive constituents, particularly catechins and other polyphenolic compounds.^{7,12-14} Previous studies have also demonstrated the antibacterial potential of *C. sinensis* against several pathogenic microorganisms, including *Escherichia coli*, *Enterobacter faecalis*, *Staphylococcus aureus*, *Salmonella typhimurium*, and *Pseudomonas aeruginosa*.^{7,15}

Zingiber officinale, commonly known as ginger, is a medicinal plant belonging to the family Zingiberaceae and is indigenous to Southeast Asia.¹⁶ Its essential oil contains various bioactive compounds, including sesquiterpenes such as zingiberol, bisabolene, and zingiberene, as well as vanilloids, flavonoids, monoterpenes, and diterpenes. These constituents are thought to contribute substantially to the therapeutic properties of ginger.¹⁷ Reported pharmacological activities of ginger include antioxidant, anti-inflammatory, anticancer, anticholinergic, antispasmodic, hypotensive, rheumatologic, and cholesterol-regulating effects, as well as its potential role in promoting blood circulation.¹⁸ Furthermore, ginger has demonstrated antimicrobial activity against a range of microorganisms, including *E.*

*Corresponding author. mail: callvic2@yahoo.com
Tel: +2348060525961**Citation:** Olugbue VU, Ogundapo SS, Obasi SE, Vining-Ogu IC, Igara CE, Suleman JB, Danladi GJ, Ngobidi KC, Nkama OJ, Uchendu D, Nsude FI, Kalu C. Microbial Quality and Sensory Evaluation of Infusions of a Polyherbal Tea Blend Formulated for Menstrual and Arthritic Pains. Trop J Phytochem Pharm Sci, 2026, 5(5); 578 - 584
<http://www.doi.org/10.26538/tjpps/v5i5.3>

Official Journal of Natural Product Research Group, Faculty of Pharmacy, University of Benin, Benin City, Nigeria.

coli, *Bacillus subtilis*, *Salmonella* spp., *Staphylococcus* spp., *Proteus* spp., *Streptococcus* spp., and *P. aeruginosa*, as well as fungal species such as *Fusarium oxysporum* and *Aspergillus niger*.^{19,20}

Clove (*Syzygium aromaticum*) belongs to the family Myrtaceae and is indigenous to the Maluku Islands of Indonesia, although it is now cultivated in several regions worldwide.²¹ The plant contains a wide range of phytochemicals, including monoterpenes, sesquiterpenes, hydrocarbons, and phenolic compounds.²² Eugenol, a major bioactive constituent of clove oil, has been associated with several pharmacological activities, including analgesic, antiseptic, antioxidant, anticancer, antidepressant, anti-inflammatory, antispasmodic, antiviral, antifungal, and antibacterial effects. Its antimicrobial activity has been demonstrated against pathogenic bacteria like *Staphylococcus aureus* and methicillin-resistant *Staphylococcus epidermidis*.²² Kumar *et al.*²³ reported that clove essential oil had antibacterial effect against *Escherichia coli* and *Salmonella* spp.

Soil microbes that adhere to plant parts may contaminate herbal medicinal products during handling of plant materials or processing.²⁴ Often, herbal tea processing involves stages that include harvesting of plant materials, sorting and drying, grinding, mixing into different proportions, bagging and storage.²⁵ Microorganisms could be introduced inadvertently at any stage. Tea infusion is prepared by steeping the dried plant materials in hot water for about 10 minutes, which may reduce the level of microbial contamination. Hot water can inactivate some microbes, whereas most fungal and bacterial spores are not affected by hot water.^{26, 27} The contamination of tea with objectionable microorganisms can compromise efficacy and pose a risk to consumers' well-being.^{28, 29}

Considering the health benefits of herbal teas, their incorporation as functional food or beverage into food matrices will feature a variety of flavours, taste, aroma, visual appeal, texture and function as prebiotics that can support the growth of beneficial bacteria, and at the same time hinder some potentially harmful ones.^{30,31} The sensory quality of herbal tea depends on volatile compounds and tasty ingredients present in the tea.³² Some consumers avoid herbal tea due to microbial contamination, as well as its aroma, flavour, taste and colour.^{10,33} Hence, there is a need to evaluate the microbial quality of herbal tea to ensure its safety and quality, as well as the sensory properties, before introducing it into the market.³² This study aimed to evaluate the microbial quality and sensory attributes of polyherbal tea formulated

using green and black tea leaves, ginger rhizomes and clove buds for the management of menstrual and arthritic pain and general wellness.

Materials and Methods

Materials

Ethics consideration

Before the sensory evaluation commenced, an informed consent form was signed by each participant after being briefed about the study. The evaluation was conducted in accordance with the ethical guidelines outlined in the Declaration of Helsinki for research involving human participants. To protect participants' privacy, no information that could personally identify them was recorded or collected.

Plant material collection

Black and green tea leaves, ginger rhizomes, and clove buds were purchased in dried form from traders in Kano State, Nigeria, and transported to Akanu Ibiam Federal Polytechnic, Unwana, Afikpo, Ebonyi State, Nigeria. The black and green tea leaves were originally harvested from the Mambilla Plateau in Taraba State and subsequently transported to Kano State, where they were purchased from traders. The Mambilla Plateau is located between 7°20'N and 11°43'E.³⁴ The plant materials were collected in October 2025. Visible impurities were removed from the plant materials, which were subsequently pulverized using a mechanical blender and stored separately in airtight glass bottles until required for use.

The plant materials were botanically identified by Obinna Chukwunwike, a botanist in the Biology Unit, Department of Science Laboratory Technology, Akanu Ibiam Federal Polytechnic, Unwana, Afikpo, Ebonyi State, Nigeria. Voucher specimens, numbered SLT012 (*Camellia sinensis*), SLT053 (*Zingiber officinale*), and SLT038 (*Syzygium aromaticum*), were deposited and archived for future reference.

Formulation and bagging of plant materials

Six polyherbal tea formulations were produced by combining the pulverized plant materials in different proportions. The resulting mixtures were packed into tea bags, with each bag containing 2.5 g of the formulated blend of green tea, black tea, ginger rhizome, and clove buds, and subsequently heat-sealed (Table 1).

Table 1: Formulation ratios of polyherbal tea products

Formulation (F)	Percentage composition			
	Green tea	Black tea	Ginger	Clove
F1	40	40	15	5
F2	55	25	15	5
F3	25	55	15	5
F4	80	0	15	5
F5	0	80	15	5
F6	50	50	0	0

Methods

Preparation of tea infusion

A tea bag from each formulation was randomly chosen and immersed in 100 mL of boiling drinking water maintained at 95–100 °C. The tea was allowed to steep for approximately 15 minutes, with occasional stirring to facilitate extraction. The prepared infusion was cooled to room temperature before further analysis.³⁵

Microbiological analysis

The microbial safety of the polyherbal tea was evaluated in accordance with the microbiological quality requirements specified in the European and British Pharmacopoeias.^{36,37} The analysis was

performed to determine the levels of aerobic bacteria, yeasts, and moulds capable of growing in 1 g of the tea sample.

The tea infusions were serially diluted, and 0.1 mL of the first dilution (10⁻¹) was inoculated in triplicate using the spread-plate technique. Nutrient agar (NA) was used for determination of Total Aerobic Microbial Count (TAMC), while Sabouraud Dextrose Agar (SDA) was used for Total Yeast and Mould Count (TYMC). MacConkey agar (MCA) was employed to detect presence of *Escherichia coli*, Salmonella–Shigella agar (SSA) was used for detection of *Salmonella* spp., and Mannitol Salt Agar (MSA) was used for enumeration of *Staphylococcus aureus*. Plates designated for bacterial enumeration were incubated at 37 °C for 24 hours, whereas fungal cultures were incubated at 26 ± 2 °C for 3–5 days. After incubation, the culture plates were inspected for microbial growth. Visible colonies were

enumerated with the aid of a colony counter, and the results were expressed as colony-forming units per gram (CFU/g) of the tea sample.

Identification of microbial isolates

Standard microbiological procedures, including morphological and biochemical characteristics, were followed for the identification of bacterial isolates. These include: characteristic growth on selective medium, Gram staining, catalase test, coagulase test, indole utilization test, urease test, methyl red test, Voges-Proskauer test, and citrate test. Sugar fermentation tests carried out include: glucose, sucrose, lactose and fructose tests.³⁸ Fungal isolates were identified based on colonial and microscopic features with the aid of standard mycological texts and manuals.^{39, 40}

Determination of moisture content of powder of polyherbal tea

The moisture content of the polyherbal tea samples prepared for packaging was determined using the oven-drying method. The samples were dried at 105 °C to a constant weight, and the moisture content was expressed as the percentage loss in weight relative to the original sample weight. The analysis was performed in triplicate, and the mean moisture content was calculated.⁴¹ The following formula was used for moisture content (%) determination:

$$\text{Final weight} - \text{Initial weight} / \text{Initial weight} \times 100$$

Sensory evaluation

The sensory attributes of the tea samples, namely appearance, aroma, taste, and overall acceptability, were assessed using a 7-point hedonic scale. The evaluation involved 17 panelists, comprising 9 females and 8 males, with ages ranging from 22 to 36 years. It was evaluated at Akanu Ibiam Federal Polytechnic, Afikpo Local Government, Ebonyi State, Nigeria. The sensory testing was conducted in the panel room at a controlled temperature and relative humidity.

Statistical analysis

The total aerobic microbial count and total yeast and mould counts were expressed as mean $\pm$ standard deviation (Mean $\pm$ SD) based on three independent replicates. Moisture content was reported as mean $\pm$ standard error of the mean (Mean $\pm$ SEM) from three replicates. The data were subjected to one-way analysis of variance (ANOVA), while significant differences among the sensory evaluation means were determined using Duncan's multiple range test as a post hoc procedure. Statistical significance was established at $p < 0.05$.

Microbiological analysis

Herbal tea consumption has risen recently due to its health benefits, making it a reliable alternative to conventional medicine.² It is essential that herbal products comply with the standards set by

regulatory authorities to meet consumers' demands for food safety and high quality.⁴³ Herbal medicines are classified as non-sterile products and must have limited and acceptable microflora, as required by regulatory guidelines.⁴⁴

From this study, the polyherbal tea samples recorded total aerobic microbial count ranging from $0.33 \pm 0.08 \times 10^2$ in F6 to $4.70 \pm 1.11 \times 10^2$ CFU/g in F4 (Table 2). The total yeast and mould count ranged from $2.00 \pm 0.35 \times 10^2$ in F1 to $6.17 \pm 1.33 \times 10^2$ CFU/g in F3. The total aerobic microbial counts and total yeast and mould counts obtained in this study were below the acceptable limits of $\times 10^7$ CFU/g and $\times 10^5$ CFU/g, respectively, established by the European and British Pharmacopoeias.^{36,37} This finding indicates that the formulations met the specified microbiological quality requirements. The population of *Staphylococcus aureus* ranged from $1.07 \pm 0.44 \times 10^2$ CFU/g in F6 to $3.39 \pm 0.45 \times 10^2$ CFU/g in F1. Furthermore, *Escherichia coli* and *Salmonella* spp., which are commonly used as indicator organisms in microbiological quality assessment, were not detected in any of the tea formulations, suggesting satisfactory hygienic quality. These findings are consistent with the observations of Farouk *et al.*⁴⁵, who evaluated the microbiological quality of 32 herbal tea products and reported bacterial and fungal loads that complied with the limits specified by the United States Pharmacopoeia. Similarly, Anosike and Oranusi⁴⁶ recorded bacterial counts ranging from 1.7×10^2 to 4.5×10^2 CFU/g in their study. Carraturo *et al.*⁴⁷ also reported microbial loads ranging from 1.0×10^2 to 2.8×10^5 CFU/g in more than 80% of the samples examined during their comparative evaluation of commercial black and green teas. In a study conducted in Sri Lanka, Karunaratne *et al.*²⁵ recorded aerobic plate counts ranging from 3.4×10^3 to 2.0×10^4 CFU/g, while yeast and mould counts varied between 4.8×10^2 and 2.5×10^3 CFU/g. Similarly, Madhab *et al.*⁴⁸ reported a mould count of 8.5×10^3 CFU/g in tea samples. Ossowski *et al.*²⁹ investigated 18 commercially available black and green tea products and found considerable variation in fungal contamination. The highest fungal load, 9.3×10^3 CFU/g, was detected in green tea bags, whereas the lowest count, 1.7×10^2 CFU/g, occurred in green tea leaves. In addition, the highest concentration of Gram-positive bacteria, 1.8×10^3 CFU/g, was observed in deciduous black tea. Our study did not detect *E. coli* and *Salmonella* sp., which agrees with earlier findings of Karunaratne *et al.*²⁵ Conversely, Madhab *et al.*⁴⁸ reported *E. coli* and *Salmonella* counts of 7.0×10^2 CFU/g and 1.9×10^3 CFU/g, respectively. Kumadoh *et al.*¹⁰ reported the presence of aerobic bacteria and fungi beyond the acceptable limit, in addition to *E. coli* in some samples analyzed in their study. The presence of *E. coli* in tea samples is unacceptable because it causes urinary tract infection, pneumonia and diarrhoea.⁴⁹ De Sousa Lima *et al.*⁵⁰ and the Canadian Food Inspection Agency⁵¹ have reported the presence of *E. coli* and *Staphylococcus* sp in some homemade and commercial herbal medicines, dried herbs and tea. Whereas the present study found *S. aureus* in the tea samples, *E. coli* were absent in all the tested formulations.

Table 2: Microbial load of polyherbal tea formulated from green and black tea leaves, dried ginger rhizomes and clove buds (Mean $\pm$ Standard Deviation $\times 10^2$ CFU/g)

Parameters	F1	F2	F3	F4	F5	F6	GLG	LT	GTFF
TAMC	3.70 $\pm$ 0.83	3.53 $\pm$ 0.94	2.70 $\pm$ 1.24	4.70 $\pm$ 1.11	1.30 $\pm$ 0.31	0.33 $\pm$ 0.08	0.80 $\pm$ 0.29	0.17 $\pm$ 0.08	0.57 $\pm$ 0.42
TYMC	2.00 $\pm$ 0.35	3.17 $\pm$ 0.61	6.17 $\pm$ 1.33	2.73 $\pm$ 0.15	3.26 $\pm$ 0.77	2.57 $\pm$ 2.52	NT	NT	NT
<i>S. aureus</i>	3.37 $\pm$ 0.45	2.47 $\pm$ 0.61	1.23 $\pm$ 0.09	2.00 $\pm$ 0.56	1.07 $\pm$ 0.78	0.27 $\pm$ 0.44	0.20 $\pm$ 0.00	0.10 $\pm$ 0.00	0.20 $\pm$ 0.00
<i>Salmonella</i>	-	-	-	-	-	-	-	-	-
<i>E. coli</i>	-	-	-	-	-	-	-	-	-

Key: F1 to F6 = Different formulations of polyherbal tea, TAMC = Total aerobic microbial count, TYMC = Total yeast/mould count. GLG = Green Lemon Ginger tea (Control), LT= Lipton tea (control), GTFF= Green tea forest fruit (control), NT= Not tested, - = Absent.

Table 3 presents the results of the commonly identified bacteria in the polyherbal infusions. *S. aureus* (45.45 %) was the most common

bacterial contaminant in the tea infusion, followed by *Bacillus* sp (32.73 %) and *Micrococcus* sp (21.81 %). Our results confirmed

previous research that reported *S. aureus*, *Bacillus* sp and *Micrococcus* sp as being among contaminants of tea.^{29,47,50-53} *S. aureus* is associated with food poisoning and toxic-shock syndrome, among other diseases, whereas *Bacillus cereus* produces toxins.⁵³ Both bacteria could cause vomiting, diarrhoea, dehydration and systemic infection.^{53,54} The possible route of contamination by *S. aureus* is through harvesting of plant materials, processing and packaging.²⁵ The frequently identified fungi in the polyherbal tea are presented in Table 4. *Penicillium* sp (37.84 %) was the most frequently identified fungus in the tea samples, followed by *Aspergillus* sp (22.97 %), *Cryptococcus* sp (19.92%), *Mucor* sp (8.11 %), *Fusarium* sp (6.76 %),

and *Rhizopus* sp (5.41 %), with the latter being the least. Anie *et al.*⁵² isolated *Aspergillus niger* (20 %) and *Aspergillus flavus* (10 %) in a similar study on microbial quality of herbal products. In a study carried out by Karunaratne *et al.*²⁵, Ossowski *et al.*²⁹ and Carraturo *et al.*⁴⁷, the presence of similar fungal species was reported. The presence of some fungi as contaminants in tea may pose a significant threat to health if they are found to be mycotoxin-producing species. *Aspergillus* sp produces mycotoxins that are carcinogenic and associated with nephrotoxicity and hepatotoxicity.⁵⁵ The presence of *Cryptococcus* sp in tea samples could pose a serious risk to immunocompromised consumers.

Table 3: Frequently identified bacteria in the polyherbal tea infusions

Bacteria	Number	Percentage
<i>S. aureus</i>	25	45.45
<i>Bacillus</i> sp	18	32.73
<i>Micrococcus</i> sp	12	21.81
Total	55	

Table 4: Frequently identified fungi in the polyherbal tea infusions

Bacteria	Number	Percentage
<i>Penicillium</i> sp	28	37.84
<i>Aspergillus</i> sp	17	22.97
<i>Cryptococcus</i> sp	14	19.92
<i>Mucor</i> sp	6	8.11
<i>Fusarium</i> sp	5	6.76
<i>Rhizopus</i> sp	4	5.41
Total	74	

Moisture content

In the present study, moisture contents were analyzed as a critical parameter that determines the microbial stability, shelf-life, chemical stability, and post-processing deterioration of dried herbal pharmaceutical products.^{2,10,56} The analysis revealed that the moisture levels across the polyherbal tea formulations varied from $2.84 \pm 0.02\%$ (w/w) in F6 to $7.11 \pm 0.87\%$ (w/w) in F1 (Table 5). The highest percentage moisture content was possessed by F1 with an average value of $7.11 \pm 0.87\%$ w/w, followed by that of F4 and F5 with an average value of $6.45 \pm 0.22\%$ w/w and $6.44 \pm 0.68\%$ w/w, respectively. Formulation 6 recorded the lowest value of $2.84 \pm 0.02\%$ w/w. A one-way analysis of variance (ANOVA) revealed that the moisture content of F1 did not differ significantly from those of F2, F3, F4, and F5 ($p > 0.05$). A statistically significant difference ($p < 0.05$) was observed in the moisture content between formulations F1 and F6. Interestingly, all six formulations recorded moisture contents below the recommended limit of $\leq 12\%$ (w/w) for dried herbal medicinal products.⁴² This indicates that none of the formulations is at risk of microbial deterioration associated with high water activity and will have a longer storage life.⁵⁷ High moisture content ($> 12\%$) can

encourage the growth of microbes, which could lead to deterioration of various consumed products, like herbal teas.^{58,59} This finding correlates with the total aerobic microbial count recorded in this study, which was found to be within an acceptable limit. Abdullah *et al.*² reported a higher moisture content of 20 to 22 % in herbal tea. Kumadoh *et al.*¹⁰ reported moisture levels of 9.75–10.71% w/w in formulated tea bag samples. Earlier, Muhammad *et al.*⁶⁰ reported a moisture content ranging from 2.46 to 7.47 % in teas from Pakistan. Our finding corroborates this result. In 2006, Yao *et al.*⁶¹ reported that approximately 70% of commercially available teas from Australian supermarkets had a moisture content of about 6.6%, while the remaining 30% had moisture levels as high as 8.0%. Such elevated moisture content may negatively influence the stability and shelf life of tea products. According to Kumadoh *et al.*¹⁰, moisture contents of herbal plants depend on the nature and type of herb, postharvest practices, plant age and their chemical composition. The overall moisture data obtained in this study support the hygienic quality of all six formulations and are consistent with their microbiological safety profiles.

Table 5: Moisture contents of Polyherbal tea formulated from green and black tea leaves, dried ginger rhizomes and clove buds

Formulation	Moisture Content (%) (Mean $\pm$ SEM)	Required maximum limit by WHO. ⁴²
F1	7.11 ± 0.87^a	$\leq 12\%$ (w/w)
F2	5.11 ± 0.78^{ab}	-do-
F3	4.22 ± 0.73^{ab}	-do-
F4	6.45 ± 0.22^{ab}	-do-
F5	6.44 ± 0.68^{ab}	-do-
F6	2.84 ± 0.02^b	-do-

Key: %= Percentage; SEM= Standard error of mean; w/w= Weight/Weight. Values with different superscript letters in the column are statistically significant ($p < 0.05$).

Sensory evaluation

The polyherbal tea samples were served to each panelist who independently examined the contents and assigned scores for the test parameters: appearance, aroma, taste and overall acceptability (Table 6). The results revealed that F1 received the highest preference in the sensory attributes of all the formulations analyzed. Formulations F1 and F6 recorded the highest appearance preference scores, with mean values of 5.35 ± 1.46 and 5.23 ± 0.97 , respectively. These scores were not significantly different from those of the other formulations ($p > 0.05$); however, both differed significantly from the control sample (L. Tea) ($p < 0.05$). Researchers have suggested that the colour/appearance of tea is contributed by oxidation and fermentation processes during processing⁶⁰ and high concentration of anthocyanins.⁶² Similarly, F1 recorded the highest preference in aroma (5.29 ± 1.67); however, this value did not differ significantly ($p > 0.05$) from those of the other formulations and the control sample. However, the aroma of the control sample was significantly different ($p < 0.05$) from that of F2, F3, F4, F5 and F6. Aroma is an important sensory attribute used to distinguish and evaluate different types of tea. It is primarily associated with volatile compounds, which constitute approximately 0.01% of the dry weight of tea.⁶³ Anwar⁶⁴ noted that aroma plays a significant role in food assessment, as the

sense of smell can help identify freshness, rancidity, and potentially harmful food products. It can also detect characteristic aromas associated with various food qualities, including green, cheesy, nutty, spicy, and rancid notes. No statistically significant variation was observed in the taste scores among the formulations ($p > 0.05$). Taste plays an important role in the recognition, identification, and enjoyment of food products.⁶⁴ Mihafu *et al.*⁶⁵ noted that taste perception is primarily associated with four basic sensations: sweet, salty, sour, and bitter. The presence of bioactive constituents, particularly phenolic compounds and caffeine, may contribute to the taste characteristics of food products.^{60,66} Among the samples evaluated, F1 received the highest overall acceptability score (5.59 ± 1.80), followed by F5 (5.18 ± 1.42) and F3 (5.17 ± 0.88). However, the overall acceptability of F1 was not significantly different from that of the other formulations ($p > 0.05$). The overall acceptability of F1 was found to be similar to that of lipton tea control sample. Sensory evaluation is an important parameter for quality assurance, new product marketing and consumer acceptability. It can influence the customer's decision to purchase food products.⁶⁷ In food industries, sensory evaluation is used in production for new products and product modification for recipes.⁶⁸

Table 6: Sensory attributes of polyherbal tea formulated from green and black tea leaves, dried ginger rhizomes and clove buds.

Formulations	Parameters			
	Appearance	Aroma	Taste	Overall Acceptance
F1	5.35 ± 1.46^b	5.29 ± 1.67^{ab}	5.41 ± 1.42^a	5.59 ± 1.80^a
F2	4.53 ± 1.37^b	4.76 ± 1.52^b	4.53 ± 1.37^{ab}	4.88 ± 1.11^{ab}
F3	4.88 ± 1.05^b	4.94 ± 1.25^b	4.47 ± 1.23^{ab}	5.17 ± 0.88^{ab}
F4	4.53 ± 1.59^b	4.71 ± 1.16^b	4.41 ± 1.46^{ab}	4.81 ± 1.23^{ab}
F5	4.94 ± 1.34^b	4.71 ± 1.26^b	4.43 ± 1.38^{ab}	5.18 ± 1.42^{ab}
F6	5.23 ± 0.97^b	4.47 ± 1.28^b	4.76 ± 1.03^{ab}	4.88 ± 0.99^{ab}
L. Tea	6.29 ± 1.16^a	6.05 ± 1.30^a	4.82 ± 1.78^{ab}	5.59 ± 1.73^a
F-value	3.773	2.626	1.299	1.637
P-value	0.0002	0.020	0.263	0.143

Key: Mean $\pm$ SD (n=17), F1 to F6 = Different formulations of polyherbal tea, L. Tea= Lipton tea (control). Values followed by different letters in the same column are significantly different ($p < 0.05$) according to Duncan's test.

Conclusion

In this study, a polyherbal tea was formulated using black and green tea (*Camellia sinensis*), ginger rhizomes (*Zingiber officinale*), and clove buds (*Syzygium aromaticum*) for the management of pain, including arthritic pain and menstrual cramps. The microbial quality of the formulated tea was assessed. The total aerobic plate count and total yeast and mould counts were within the acceptable limits of $\times 10^7$ CFU/g and $\times 10^5$ CFU/g, respectively, as recommended by the British and European Pharmacopoeias. *Escherichia coli* and *Salmonella* sp., which are considered indicator organisms for coliform contamination, were not found in any of the formulations. *Staphylococcus aureus* and *Penicillium* sp. were the most frequently detected bacterium and fungus, respectively, in the tea samples. Formulation 1 had the highest moisture content, whereas Formulation 6 recorded the lowest. Sensory evaluation showed that F1 received the highest preference across the sensory attributes assessed, including appearance, aroma, taste, and overall acceptability. These findings indicate that the plant materials used in this study are promising sources of bioactive constituents for the development of herbal tea products with good consumer acceptability.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

Acknowledgement

The authors are appreciative of Tertiary Education Trust Fund (Tetfund) Institution-Based Research (IBR) grant (TETF/ES/DR&D/CE/POLY/UNWANA/IBR/2025/VOL.1) for funding the research.

References

1. FAO. FAO publications Catalogue 2022; FAO: Rome, Italy, 2020.
2. Abdullah R, Zaheer S, Kaleem A, Iqtedar M, Aftab M, Saleem F. Formulation of herbal tea using *Cymbopogon citratus*, *Foeniculum vulgare* and *Murraya koenigii* and its anti-obesity potential. J. King Saud Univ. Sci. 2023; 35: 1018-3647. <https://doi.org/10.1016/j.jksus.2023.102734>
3. Tang GY, Zhao CN, Xu XY, Gan RY, Cao SY, Liu Q, Shang A, Mao QQ, Li HB. Phytochemical composition and antioxidant capacity of 30 Chinese teas. Antioxidant. 2019; 8: 180.

4. Thomas TL, Umoyen AJ, Emeagi IL, Idu JI, Mshelmbula BP, Etukudo OM, Udensi UO. Bioactive Components of Tea Plant (<I>Camellia sinensis</I> (L.) O. Kuntze) From Mambilla Plateau, Taraba State - Trop. J. Phytochem. Pharm. Sci. 2025; 4(2): 50–54. <https://doi.org/10.26538/tjpps/v4i2.3>
5. Foster MT, Gentile CL, Cox-York K, Wei Y, Wang D, Estrada AL, Reese L, Miller T, Pagliassotti MJ, Weir TL. Fuzhuan tea consumption imparts hepatoprotective effects and alters intestinal microbiota in high saturated fat diet-fed rats. Mol. Nutr. Food Res. 2016; 60(5): 1213–1220. <https://doi.org/10.1002/mnfr.201500654>
6. Zhou J, Tang L, Shen CL, Wang J-S. Green tea polyphenols boost gut-microbiota-dependent mitochondrial TCA and urea cycles in Sprague–Dawley rats. J. Nutr. Biochem. 2020; 81: 108395.
7. Liu S, Zhang Q, Li H, Qiu Z, Yu Y. Comparative Assessment of the Antibacterial Efficacies and Mechanisms of Different Tea Extracts. Foods. 2022; 11: 620. <https://doi.org/10.3390/foods11040620>
8. Poswal FS, Russell G, Mackonochie M, MacLennan E, Adukwu EC, Rolfe V. Herbal Teas and their Health Benefits: A Scoping Review. Plant Foods Hum Nutr. 2019; 74(3): 266–276.
9. Liu Y, Guo C, Zang E, Shi R, Liu Q, Zhang M, Zhang K, Li M. Review on herbal tea as a functional food: classification, active compounds, biological activity, and industrial status. J. Future Foods. 2023; 3(3): 206–219.
10. Kumadiah D, Archer M-A, Kyene MO, Yeboah GN, Adi-Dako O, Osei-Asare C, Adase E, Mintah SO, Amekyeh H, Appiah AA. Approaches for the elimination of microbial contaminants from *Lippia multiflora* leaves intended for tea bagging and evaluation of formulation. Adv. Pharmacol. Pharm. Sci. 2022; Article 7235489. <https://doi.org/10.1155/2022/7235489>
11. Namita P, Mukesh R, Vijay KJ. *Camellia sinensis* (Green Tea): A Review. Glob J Pharmacol. 2012; 6 (2): 52–59.
12. Zhu Y, Luo Y, Wang P, Zhao M, Li L, Hu X, Chen F. Simultaneous determination of free amino acids in Pu-erh tea and their changes during fermentation. Food Chem. 2016; 194: 643–649.
13. Niu L, Li Z, Fa W, Zhong X, Peng M, Liu Z. Nano-Strategies for enhancing the bioavailability of tea polyphenols: Preparation. Appl. Chall. Foods. 2022; 11: 387.
14. Yan Z, Zhong Y, Duan Y, Chen Q, Li F. Antioxidant mechanism of tea polyphenols and its impact on health benefits. Anim. Nutr. 2020; 6: 115–123.
15. Boudou F, Belakredar A, Keziz A, Alsaeedi H, Cornu D, Bechelany M, Barhoum A (2025). *Camellia sinensis* phytochemical profiling, drug-likeness, and antibacterial activity against gram-positive and gram-negative bacteria: in vitro and in silico insights. Front. Chem. 2025; 13:1555574. doi: 10.3389/fchem.2025.1555574
16. Sharma S, Raj H, Gupya J. Herbal tea: a comprehensive review of health benefits and therapeutic potential. Int J. Res Publ Rev. 2025; 6(5):1376–1389
17. Zhang S, Kou X, Zhao H, Mak KK, Balijepalli MK, Pichika MR. *Zingiber officinale* var. rubrum: Red Ginger's Medicinal Uses. Molecules. 2022; 27: 775. <https://doi.org/10.3390/molecules27030775>
18. Paudel KR, Orent J and Penela OG (2025). Pharmacological properties of ginger (*Zingiber officinale*): what do meta-analyses say? A systematic review. Front. Pharmacol. 2025; 16:1619655. doi: 10.3389/fphar.2025.1619655
19. Shakir MSA, Marwan TJG. Study on antibacterial and antifungal activity of ginger (*Zingiber officinale*) on selected pathogenic microorganisms. Biochem. Cell. Arch. 2020; 2: 4629–4633. DocID: <https://connectjournals.com/03896.2020.20.4629>
20. Soumitra M, Rammohan B, Dijendra NR. Bactericidal action of ginger (*Zingiber officinale* Roscoe) extract against *Escherichia coli* through synergistic modulation of the AcrAB-TolC efflux pump and inhibition of peptidoglycan synthesis: *In vitro* and in silico approaches. Microb. Pathog. 2025; 204: 107624. <https://doi.org/10.1016/j.micpath.2025.107624>
21. Batiha GES, Beshbishy AA, Tayebwa DS, Shaheen MH, Yokoyama N, Igarashi I. Inhibitory effects of *Syzygium aromaticum* and *Camellia sinensis* methanolic extracts on the growth of Babesia and Theileria parasites. Ticks Tick. Borne Dis. 2019; 10: 949–958.
22. Batiha GES, Alkazmi LM, Wasef LG, Beshbishy AM, Nadwa EH, Rashwan EK. *Syzygium aromaticum* L. (Myrtaceae): Traditional Uses, Bioactive Chemical Constituents, Pharmacological and Toxicological Activities. Biomolecules. 2020; 10: 202. doi:10.3390/biom10020202 www.mdpi.com/journal/biomolecules
23. Kumar PV, Shams R, Singh R, Dar AH, Pandiselvam R, Rusu AV, Trif M. A comprehensive review on clove (*Caryophyllus aromaticus* L.) essential oil and its significance in the formulation of edible coatings for potential food applications. Front. Nutr. 2022; 9:987674. doi: 10.3389/fnut.2022.987674
25. Karunaratne SHS, Abeygunawardena GASI, Jayaratne DL, Premakumara GAS. Microbiological quality of different tea grades produced in diverse agro-climatic regions in Sri Lanka. Heliyon. 2024; 10(6): e27878.
26. Akduman G, Omurtag KI. Microbial evaluation of Turkish herbal teas before and after infusion. J. Food Qual. Hazard Control. 2020; 7: 170–174. DOI:10.18502/jfqhc.7.4.4844
27. Hussain F, Salam I, Farzana, Memon Z, Addullah M, Abbas G, Akbar M, Hussain A, Majeed M, Ali K, Moda HM. Occurrence of fungal microbial contamination in drinking water of megacity of Karachi (Pakistan) and their physicochemical control. Heliyon. 2024; 10 (7), e2896. Doi:10.1016/j.heliyo.2024.e28926
28. Agarwal S, Rai N, Pandey A, Kumar S, Anand J. Microbial contamination and its impact on tea quality. Environ. Conserv. J. 2025; 26 (1): 149–158. Doi:<https://doi.org/10.36953/ECJ.30213156>
29. Ossowski M, Nowakowicz-Dębek B, Wlazło L, Król J, Kasela M, Maksym P, Malm A. Evaluation of Microbiological Contamination of Black and Green Teas. Adv. J. Food Sci. Technol. 2019; 17(4): 65–71. doi:10.19026/ajfst.17.6016
30. Gibson GR, Hutkins R, Sanders ME, Prescott SL, Reimer RA, Salminen SJ, Scott K, Stanton C, Swanson KS, Cani PD, Verbeke K, Reid G. Expert consensus document: The International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of prebiotics. Nat. Rev. Gastroenterol. Hepatol. 2017; 14(8):491–502. doi:10.1038/nrgastro.2017.75
31. Trigo C, Castelló ML, Ortolá MD. “Potentiality of *Moringa oleifera* as a Nutritive Ingredient in Different Food Matrices,” Plant Foods Hum. Nutr. 2022; 1: 25–37. doi: <https://doi.org/10.1007/s11130-022-01023-9>
32. Dias I, Gayani P, Upul RA, Marapana J, Udaya RM, Rathnayaka SK. Production and Quality Evaluation of Herbal Tea Mixtures from *Phyllanthus Debilis*, *Osebeckia octandra*, and *Artocarpus heterophyllus* Leaves. Sumatera Med. J. 2023; 6 (3): 200 – 209.
33. Aderinola TA. “Nutritional, antioxidant and quality acceptability of smoothies supplemented with *Moringa oleifera* leaves,” Beverages. 2018; 4(4): 104. doi: <https://doi.org/10.3390/beverages4040104>
34. Salako G, Olalubi O, Sawyerr H, Howe G, Adebayo A, Adio A. Using multi techniques analysis in biogeoclimatic ecosystem classification and mapping of Mambilla plateau in Taraba State Nigeria. Open J Ecol. 2016; 6: 412–426.
35. Olugbue VU, Ogundapo SS, Obasi, SE, Vining-Ogu IC, Igara CE, Suleman JB, Danladi GJ, Ngobidi KC, Nkama OJ, Uchendu D, Nsude FI, Kalu C. Phytochemical Profile, Antibacterial Activity and Effect of Infusions from Polyherbal Tea on Gut Microbiota of Albino Rats. FUDMA J. Sci. 2026; 10(11): 343–352. <https://doi.org/10.33003/fjs-2026-1011-5268>
36. British Pharmacopoeia. Vol. V, British Pharmacopoeia Commission. Stationery Office, Great Britain. 2022. pp. V-A566-V-A5667.
37. European Pharmacopoeia. 10 ed. Vol. 1(Ph. Eur. 10.0), European directorate for the quality of medicines and healthcare of the

- council of Europe (EDQM), Council of Europe, Strasbourg, 2019. pp. 638.
38. Cheesbrough M. District Laboratory Practice in Tropical Countries. Part II, 5th edn., New York: Cambridge University Press. 2008.
 39. Larone DH. Medically important Fungi: A guide to identification. 3rd edn., Washington D. C., ASM press.1995.
 40. DeHoog GS, Guarro J, Figueras MJ. Atlas of Clinical Fungi. Atlas Version 2004.1, CD realization by Weniger, T. Computer Science II, University of Wurzburg, Germany. 2004.
 41. British Pharmacopoeia. *British Pharmacopoeia (BP)*, British Pharmacopoeia Commission, London, UK. 2021.
 42. WHO. Quality control methods for medicinal plant materials. World Health Organization. who.int. 2011.
 43. FSMA Rules & Guidance for Industry. FDA Food Safety Modernization Act (FSMA) rules and guidance pages. <https://www.fda.gov/food/food-safety-modernization-act-fsma/fsma-rules-guidance-industry>. 2024.
 44. Microbiological aspects of herbal medicinal products and traditional herbal medicinal products - Scientific guideline. (2015). European Medicines Agency. <https://www.ema.europa.eu/en/microbiological-aspects-herbal-medicinal-products-traditional-herbal-medicinal-products-scientific-guideline>. 2015.
 45. Farouk M, El-Ganiny A, Abdelmonem M, Serry F. Evaluation of microbial quality of herbal tea pharmaceutical products found in the Egyptian market. *Zagazig J. Pharm. Sci.* 2017; 26 (1): 32-38.
 46. Anosike SO, Oranusi S. Assessment of microbiological and chemical qualities of selected cocoa, tea and coffee brands in Nigerian market. *Afr. J. Clin. Exp. Microbiol.* 2018; 19(3). <https://doi.org/10.4314/ajcm.v19i3.5>
 47. Carraturo FO, De Castro J, Troisi A, De Luca A, Masucci A, Cennamo P, Trifuoggi M, Aliberti F, Guida M. Comparative assessment of the quality of commercial black and green tea using microbiology analyses. *BMC Microbiol.* 2018; 18(1): 4. Doi: 10.1186/s12866-017-1142-z
 48. Madhab M, Bhattacharyya PN, Begum R, Dutta P, Tanti AJ, Konwar M, Hazarika S, Sarmah SR. Microbial distribution and contamination in black tea. *Two Bud.* 2019; 64(1): 26-31.
 49. Makvana S, Krilov LR. "Escherichia coli infections," *Pediatr. Rev.* 2015; 36 (4): 167-171.
 50. De Sousa Lima CMS, Mayara ATF, Bruno PL, Patrícia CM, Francisco FOS, Jocivânia OS. "Microbial contamination in herbal medicines: a serious health hazard to elderly consumers," *BMC Complement. Med. Ther.* 2020; 20 (17): 1-9.
 51. Canadian Food Inspection Agency. Bacterial Pathogens in Dried Herbs and Dried Teas April 1, 2014 to March 31, 2018, Food Microbiology - Targeted Surveys - Final Report, Canadian Food Inspection Agency, Canada. 2018.
 52. Anie OC, Egbon OT, Enemchukwu CM, Adushoke EL. The Microbial Quality of Herbal Products. *J. Drug Deliv. Ther.* 2022; 12(5), 64-69. DOI: <http://dx.doi.org/10.22270/jddt.v12i5.5590>
 53. Onyewenjo C, Agbagwa OE, Frank-Peterside N, Onyewenjo SC. Microbial Quality and Antimicrobial Potential of some Herbal Remedies Marketed in Owerri-West Nigeria. *Afr. J. Health Sci.* 2022; 35(4): 537-549.
 54. Uhlig E, Megaelectra A, Molin G, Håkansson Å. Viable and Heat-Resistant Microbiota with Probiotic Potential in Fermented and Non-Fermented Tea Leaves and Brews. *Microorganisms.* 2025; 13(5), 964. <https://doi.org/10.3390/microorganisms13050964>
 55. Jin J, Lee YK, Wicks BL. Simple chemical extraction method for DNA isolation from *Aspergillus* species. *J. Clin. Microbiol.* 2004; 42: 4293- 4296.
 56. Eko SS, Indriwati SE, Suarsini E. "Preparation of various types of medicinal plants simplicia as material of Jamu herbal, education in the 21st century: responding to current issues," in Proceedings of the International Conference on Education, Hangzhou, China, April 2016.
 57. Awogbeni O, Ogunleye IO. Some selected vegetables. *Int. J. Eng. Technol.* 2009; 1: 1793-8236.
 58. Handbook of Pharmaceutical Excipients. Acacia, 5th edn., Pharmaceutical Press, London, pp. 1. 2006.
 59. Rashid R, Kurt A, Bern C. Effects of Deterioration parameters on storage of maize: A Review. *J. Nat. Sci. Res.* 2013; 3(9):147-165.
 60. Muhammad A, Asif A, Anwaar A, Nauman K, Imran H, Iftikhar A. Chemical Composition And Sensory Evaluation of Tea (*Camellia Sinensis*) Commercialized In Pakistan. *Pak. J. Bot.* 2013; 45(3): 901-907.
 61. Yao L, Liu X, Jiang Y, Caffin N, D'Arcy B, Singanusong R, Datta N, Xu Y. Compositional analysis of teas from Australian supermarkets. *Food Chem.* 2006; 94(1): 115-122.
 62. Pi-Jen T, McIntosh J, Pearce P, Camden B, Jordan BR. Anthocyanin and Antioxidant Capacity in Roselle (*Hibiscus sabdariffa* L.) Extract. *Food Res. Int.* 2002; 35: 351-356.
 63. Moreira J, Aryal J, Guidry L, Adhikari A, Chen Y, Sriwattana S, Prinyawiwatkul W. Tea Quality: An Overview of the Analytical Methods and Sensory Analyses Used in the Most Recent Studies. *Foods.* 2024; 13(22):3580. <https://doi.org/10.3390/foods13223580>
 64. Anwar S, Syed QA, Asmat U, Arshad M, Rehman MA, Ahmad W, Muzammil HS. Antioxidants quantification, minerals profile, colour properties and sensorial quality of *Laurus nobilis* composite tea infused with ginger and stevia. *J. Food Meas. Charact.* 2023; 17(5): 5144-5153.
 65. Mihafu FD, Issa JY, Kamiyango MW. Implication of Sensory Evaluation and Quality Assessment in Food Product Development: A Review. *Curr. Res. Nutr. Food Sci.* 2020; 8(3): 75-83.
 66. Zhang L, Cao QQ, Granato D, Xu YQ, Ho CT. Association between chemistry and taste of tea: A review. *Trends Food Sci. Technol.* 2020; 101: 139-149.
 67. Rathee R, Rajain P. "Role colour plays in influencing consumer behaviour," *Int. Res. J. Bus. Stud.* 2019; 12(3): 209-22. doi: <https://doi.org/10.21632/irjbs.12.3.209-222>
 68. Calanche JB, Beltran JA, Hernandez Arias AJ. Aquaculture and sensorimetrics: The need to evaluate sensory attributes and the consumers' preferences. *Rev. Aquac.* 2020; 12(2): 805-821.