Formulation and characteristics of bioactive compounds and antioxidant activity of moringa leaf herbal tea enriched with ginger (*Zingiber officinale*) and Javanese turmeric (*Curcuma xanthorrhiza*)

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Abstract

The study aims to evaluate an herbal tea formula made from *Moringa oleifera* leaves, combined with ginger and Javanese turmeric, and to analyze its effects on the tea's characteristics and antioxidant activities. Five herbal tea formulas were created using *M. oleifera* leaf powder and spice powder, with the spices consisting of 50% ginger (*Zingiber officinale*) and 50% Javanese turmeric (*Curcuma xanthorrhiza*). The formulations were as follows: F1 (100% *M. oleifera*), F2 (90% *M. oleifera* + 10% spices), F3 (85% *M. oleifera* + 15% spices), F4 (80% *M. oleifera* + 20% spices), and F5 (75% *M. oleifera* + 25% spices). These formulas were analyzed for changes in characteristics and antioxidant activity. The results showed that the inclusion of ginger and Javanese turmeric—especially in the F5 formulation—did not significantly alter the tea's carbohydrate, fat, moisture, antioxidant activity, total flavonoid, or total phenolic content. However, protein and ash contents decreased with higher spice concentrations. Sensory evaluation revealed that the F5 formulation was preferred by panelists because of its improved taste and aroma, suggesting that incorporating these spices enhances the sensory appeal of *M. oleifera* herbal tea without compromising its antioxidant and nutritional properties.

Keywords: Flavonoid, Flavor, Food, Phenolic, Taste

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Introduction

In recent years, functional food products have attracted considerable attention in the food industry. This surge in interest has led to increased consumer awareness of nutrient-rich functional foods, resulting in a growing demand for these products over the past decade (Todaro *et al*., 2023). Few of the many functional foods developed have gained popularity among consumers because of their health benefits and recognized nutritional value. Tea is one of the most widely consumed beverages, appreciated for both its flavor and health advantages (Viegas *et al.,* 2020). In addition, herbal teas made from various plant species are also popular. Among these herbal beverages, moringa leaf decoction is gaining traction for its health benefits, particularly its antioxidant properties and high phenolic content.

Moringa (*Moringa oleifera*) is well-known for its high nutritional value, containing essential proteins, vitamins, minerals, and antioxidants (Fguiri *et al.,* 2024; Saini *et al.,* 2016). Research has demonstrated that moringa leaves provide numerous health benefits, including lowering blood pressure, regulating blood sugar levels, and boosting the immune system (Mbikay, 2012; Munir *et al.,* 2025; Stohs & Hartman, 2015). In addition, Rockwood *et al.* (2013) discovered that moringa leaf extract exhibits strong antioxidant properties, which help combat free radicals in the body. One popular way to incorporate moringa leaves into the human diet is by making functional foods and beverages (Yang *et al.,* 2023). Adding *M. oleifera* enhances functional food products' nutritional value and antioxidant activity, such as avena sativa bread (Sánchez-Ortiz *et al.,* 2024) and chocolate (Gomes *et al.,* 2024). Furthermore, moringa leaves can be used to create healthy and refreshing herbal teas (Prabakaran & Krishnaswamy, 2020). Research on a functional drink made from a mixture of moringa leaves, pandanus leaves, and red ginger produced a product with potential as an anti-inflammatory agent (Widyaningsih *et al.* 2021). Pradanto *et al.* (2022) found the best composition of turmeric rhizome, moringa leaf, and brown seaweed with the highest total phenolic and total phlorotannin content for an antidiabetic functional beverage. Another research showed that formulated herbal fermented beverages utilizing fresh *Cymbopogon citratus, Zingiber officinale, M. oleifera*, *Mentha*, and *Curcuma longa*, resulted in notable enhancements in flavor and phytochemical composition (Don *et al.,* 2024). However, herbal teas made from moringa leaves often have an undesirable taste and flavor because of the low levels of essential oils and volatile compounds typically responsible for the flavor and fragrance of plants (Anwar *et al.,* 2007). To address this issue, natural ingredients such as spices can be a solution (Devi *et al.,* 2024; Shaikh *et al.,* 2025). *M. oleifera* tea, when combined with spices like cloves, cardamom, black pepper, and mint leaves, can produce a more aromatic, healthy, and nutritious herbal tea, though it may still have a bland taste (Panda & Singh, 2025). Therefore, additional spices are necessary to enhance the taste and flavor of moringa tea.

One spice ingredient known for its pungent flavor is ginger (*Zingiber officinale*). Ginger has been utilized both as a culinary spice and in traditional medicine. It is recognized for its potent anti-inflammatory and antioxidant properties, which help reduce inflammation and combat oxidative stress in the body (Wang *et al.,* 2021; Wang *et al.,* 2025). The main active compound in ginger is gingerol, a phenolic compound known for its significant antioxidant and anti-inflammatory effects (Sharifi-Rad *et al.,* 2017). Not only is ginger a nutritious spice, but it also offers numerous health benefits. It is widely recognized for reducing inflammation, alleviating nausea, and improving digestive health (Ali *et al.,* 2008; Rahmani *et al.,* 2014). The spicy flavor of ginger primarily comes from gingerol. In addition to gingerol, ginger contains other compounds such as shogaol and zingerone, which are degraded products of gingerol. These compounds contribute to ginger's distinctive taste and enhance its beneficial antioxidant properties (He *et al.,* 2019). In culinary uses, ginger enhances the flavor of food and beverages by adding a refreshing, tangy note while masking any bitter or unpleasant tastes from other ingredients (Pongpiriyadacha *et al.,* 2020). Incorporating ginger into MO herbal tea can boost its nutritional content and improve its flavor. Furthermore, Javanese turmeric (*Curcuma xanthorrhiza*) is another widely used herbal plant valued for its health benefits. It exhibits pharmacological effects, including antioxidant, antimicrobial, anti-inflammatory, antidiabetic, and anticancer properties (Rahmat *et al.,* 2021).

Combining *M. oleifera* leaves, ginger, and Javanese turmeric in herbal tea provides enhanced health benefits by leveraging these three plants' nutritional content and antioxidant properties. *M. oleifera* leaves are rich in essential nutrients, including vitamins A and C, calcium, and protein. Ginger contributes key compounds such as gingerol, shogaol, and zingerone, which not only boost health benefits but also enhance the flavor and aroma of the tea, making it fresher and more enjoyable (González & Navarro, 2020). Therefore, the formulation of tea using *M. oleifera* leaves, ginger, and turmeric improves its nutritional profile and offers consumers a more pleasant experience of tea drinking. This study aimed to investigate the effect of mixing Moringa leaves with Javanese ginger and turmeric powder on the volatile composition, antioxidant potential, and

organoleptic characteristics of the resulting herbal tea. While this analysis provides an insight into the potential functional value of the formulation, the findings are based on in vitro composition and antioxidant assays; thus, their physiological implications are only inferred and not directly proven.

Materials and Methods

Materials

Fresh *M. oleifera* leaves were obtained from the Indonesian Medicinal and Aromatic Crops Research Institute (Bogor, Indonesia, 6°34'35.402" S, 106°47'9.475" E). Ginger (*Zingiber officinale*) and Javanese turmeric (*Curcuma xanthorrhiza*) rhizomes were purchased from the local market (Bogor, Indonesia).

Herbal tea preparations and formulation

Fresh *M. oleifera* leaves were thoroughly washed and cut into small pieces. The leaves were oven-dried at 40°C for 24 hours and ground into a fine powder using a blender. The resulting powder was passed through a 40-mesh sieve. Meanwhile, fresh ginger (*Zingiber officinale*) and Javanese turmeric (*Curcuma xanthorrhiza*) rhizomes were peeled and sliced into thin pieces (1–2 mm thick), then oven-dried at 50°C for 8 hours. The dried rhizome slices were ground into powder and sieved through a 40-mesh sieve. All powdered *M. oleifera*, ginger, and Javanese turmeric samples were stored in airtight containers with silica gel at room temperature until further use for product formulation and analysis. For the preparation of herbal tea, 2 g of *M. oleifera* leaf powder were accurately weighed and placed in a tea bag, followed by the addition of spice powder according to the designated formulation ratio (Table 1). The spice blend consisted of equal proportions (1:1) of ginger (*Z. officinale*) and Javanese turmeric (*C. xanthorrhiza*) powders. Five formulation treatments were prepared (Table 1).

Organoleptic evaluation

An organoleptic analysis was conducted at the Research Center for Agroindustry, National Research and Innovation Agency, and approved by the Ethical Committee on Chemical Research—National Research and Innovation Agency, Indonesia. For the analysis, 2 g of herbal tea powder extract was brewed in 150 mL of boiling water (100°C) for 3 minutes. The resulting herbal tea beverage was served warm, at approximately 50°C, and its sensory properties were evaluated. Each panelist was randomly presented with samples of the herbal tea beverages, and water bottles were provided for rinsing between evaluations. They received 40–50 mL of each beverage in transparent glasses, which were randomly coded for identification. This evaluation involved 20 untrained panelists who assessed various sensory attributes, including taste, color, and aroma. Untrained panelists consisted of lay people selected based on the following criteria: gender (10 women and 10 men), age (20–40 years), health status, absence of sensory disorders (smell disorders (anosmia), taste disorders (ageusia), or food allergies), and not currently taking drugs or foods that affect the sensation of taste/aroma (antibiotics, cigarettes, alcohol). The analysis employed a 5-point hedonic scale, where 1 indicated "dislike extremely" and 5 indicated "like extremely". All participants were informed about the purpose of the study and provided their consent voluntarily prior to participation.

Sample extraction

Sample extraction was performed using the infusion (steeping) method following standard procedures for preparing herbal tea (Hernández-Fuentes *et al.,* 2025). One gram of dried *M. oleifera* leaf powder (or herbal tea blend) was placed into 100 mL of hot water (≈90°C) and steeped for 10 minutes. The infusion was then filtered using Whatman No. 1 filter paper and cooled to room temperature. The resulting filtrate was used directly to analyze antioxidant activity and chemical composition.

Table 1. Herbal tea formulation ratios.

Formulation Code	*M. oleifera* Leaf Powder (%)	Spice Powder (%)	
		Z. officinale	*C. xanthorrhiza*
F1	100	0.0	0.0
F2	90	5.0	5.0
F3	85	7.5	7.5
F4	80	10.0	10.0
F5	75	12.5	12.5

Proximate analyses

The nutritional contents of *M. oleifera* herbal tea were analyzed following the method previously described (Association of Official Analytical Chemists (AOAC), 2000). Analyses included moisture, ash, protein, fat, and carbohydrate contents.

Moisture content

The cups were dried in an oven at 105°C for 1 hour, then allowed to cool in a desiccator for 30 minutes before being weighed (A). This weighing process was repeated until a consistent weight was achieved. A sample, weighing between 1 and 2 g, was carefully placed into a cup and re-weighed (B). The cup containing the sample was then heated at 105°C for 5 hours or until a constant weight was reached. After heating, the cup and its contents were cooled in a desiccator. This entire procedure was repeated until a stable weight (C) was consistently obtained. The moisture content was calculated as follows (Equation 1):

$$\text{Moisture content}(\%) = \frac{(B - C)}{(B - A)} \text{x} 100\% \quad (1)$$

Where A is the weight of the empty cup (g), B is the weight of the cup and the initial sample (before dried), and C is the weight of the cup and dried samples (g).

Ash content

Initially, the crucible was heated to 550°C in a muffle furnace and maintained at that temperature for 2 hours. After this, the temperature was gradually lowered. Once the temperature reached room level, the samples were quickly transferred to a desiccator and weighed (B). A maximum of 3 g (C) of the sample was placed in the crucible and then incinerated in the furnace at a maximum temperature of 550°C for 1 hour, or until a light gray color was achieved, resulting in (A). The ash content was calculated using Equation 2:

$$\text{Ash content}(\%) = \frac{(A - B)}{C} \text{x} 100\% \quad (2)$$

Where A is the weight of the empty crucible and ash (g), B is the weight of the empty crucible (g), and C is the weight of the sample (g).

Fat content

A test portion weighing between 1 and 2 g was measured and placed into a thimble lined with filter paper and cotton. The cotton ball was carefully arranged inside the thimble to fully cover the sample, and then the thimble was folded to secure the sample in place. Next, the thimble was dried in an oven at a maximum temperature of 80°C for 1 hour and then transferred into a flat-bottom flask. The flask was positioned in a Soxhlet apparatus, which was connected to a fat flask containing previously dried and weighed boiling stones. The samples were extracted with N-hexane for approximately 6 hours. After the extraction process, the thimble was removed, and the leftover N-hexane was collected and evaporated. To eliminate moisture from the thimble and hexane, the flask was placed in an oven at 105°C for about 30 minutes. It was then allowed to cool in a desiccator before weighing. This drying process was repeated until a consistent weight was achieved. The fat content was calculated using Equation 3:

$$\text{Fat content}(\%) = \frac{(W - W_1)}{W_2} \text{x} 100\% \quad (3)$$

Where W is the weight of the flask with extracted fat (g), W_1 is the weight of the empty flask (g), and W_2 is the weight of the sample (g).

Protein content

A sample weighing 0.5 g was placed into a 100 mL Kjeldahl flask, along with 2 g of a selenium mixture and 25 mL of concentrated sulfuric acid (H_2SO_4). The mixture was then heated on an electric heater for approximately 2 hours. After heating, the solution was cooled to room temperature and diluted by transferring it into a 100 mL volumetric flask. Next, 5 mL of this solution was pipetted into the distillation unit, where 5 mL of 30% NaOH and a few drops of phenolphthalein indicator were added. The solutions were distilled for about 10 minutes. In a separate Erlenmeyer flask, a 10 mL solution of 2% boric acid mixed with indicators was prepared. The distilled solution was then titrated with a 0.01 N HCl solution. This procedure was also conducted for the blanks. The protein level was subsequently calculated using Equation 4:

$$\text{Protein content}(\%) = \frac{V_1 - V_2 \times N \times 0.014 \times Fk \times Fp}{W} \quad (4)$$

Where W is the weight (g) of the test portion/sample or standard, V_1 is the volume (mL) of 0.01 N HCl used to titrate a test, V_2 is the volume (mL) of 0.01 N HCl used to titrate reagent blank, N is the normality of HCl, F_k is the conversion factor (6.25), and F_p is the dilution factor.

Carbohydrate content

A 5-g sample was weighed and placed in a 500 mL Erlenmeyer flask. Then, 200 mL of a 3% hydrochloric acid (HCl) solution was added. The mixture was boiled for 3 hours under reflux. After cooling, it was neutralized with a 30% sodium hydroxide (NaOH) solution, using litmus paper or phenolphthalein as indicators. The solution was adjusted to slight acidity by adding 3% acetic acid (CH_3COOH). The resulting solution was transferred to a 500 mL volumetric flask and filtered. Subsequently, 10 mL of the filtered solution was pipetted into another 500 mL Erlenmeyer flask. To this, 25 mL of Luff solution, 15 mL of distilled water, and a few boiling stones were added. The mixture was brought to a boil within 3 minutes and allowed to boil for 10 minutes. After boiling, the solution was cooled in an ice bath. Next, 15 mL of a 20% potassium iodide (KI) solution and 25 mL of a 25% H_2SO_4 solution were slowly added. The solution was then titrated with a 0.1 N 0.5% sodium thiosulfate solution. After cooling the solution again, an additional 15 mL of 20% KI solution and 25 mL of 25% H_2SO_4 were added slowly. Titration continued with the 0.1 N 0.5% sodium thiosulfate solution, with 2–3 mL of starch indicator included for the final titration. The carbohydrate content was calculated using Equation 5:

$$\text{Glucose content}\,(\%) = \frac{w_1 \times fp}{w} \text{x}100\% \quad (5)$$

Where carbohydrate content is equal to 0.90 × glucose content, W_1 is the weight of samples (mg), W is the glucose (mg) contained for mL thiosulfate solution used, F_p is the correction factor.

Antioxidant Activity

The antioxidant activity was assessed using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical degradation method (Harish & Shivanandappa, 2006). A total of 9.858 mg of DPPH was dissolved in 100 mL of 95% ethanol to create a 0.25 mM DPPH solution. A parallel blank experiment was prepared using 0.5 mL of 95% ethanol. Subsequently, 200 µL of the leaf extract solution was added to test tubes, followed by the addition of 0.5 mL of the prepared DPPH solution. The contents of the tubes were thoroughly mixed and then incubated for 30 minutes in the dark before measuring the absorbance spectrophotometrically. Absorbance readings were taken using a Shimadzu UV-Vis 1800 at a wavelength of 517 nm. The inhibition rate of the DPPH free radical was calculated using the formula provided in Equation 6:

$$\text{DPPH Inhibition}\,(\%) = \frac{A_b - As}{Ab} \text{ x } 100\% \quad (6)$$

Where A_b represents the absorbance of the blank experiment, and A_s represents the absorbance of the sample.

Total phenolic content

The total phenolic content was determined using a modified version of the Folin–Ciocalteu method (Ainsworth & Gillespie, 2007). In this analysis, a sample solution with a concentration of 2000 ppm was used. Approximately 200 µL of the sample solution, standard and methanol (as a blank) was mixed with 1.5 mL of 10% (v/v) Folin–Ciocalteu reagent. The mixture was thoroughly mixed and then incubated for 5 minutes at room temperature. Afterward, 1.5 mL of 10% Na_2CO_3 was added, and the mixture was allowed to stand for 120 minutes in the dark room temperature. The absorbance of the resulting solution was measured spectrophotometrically using a Shimadzu UV-Vis 1800 at a wavelength of 752 nm. The total phenolic content was quantified and expressed in milligrams of gallic acid equivalent per gram of dry weight (mg GAE/g), with gallic acid used as the standard.

Total flavonoid content

The total flavonoid content was determined using a modified version of an established method (Sarker & Oba, 2018). The procedure involved preparing a sample solution with a concentration of 500 ppm. For the analysis, 500 µL of the sample was placed in a test tube and mixed with 1.5 mL of methanol, 0.1 mL of 10% aluminum chloride, 0.1 mL of 1 M potassium acetate, and 2.8 mL of distilled water. The mixture was then incubated at room temperature for 60 minutes. After incubation, the absorbance was measured spectrophotometrically using a Shimadzu UV-Vis 1800 at a wavelength of 432 nm. Quercetin served as the standard compound for this analysis. The total flavonoid content was quantified and expressed as milligrams of quercetin equivalents per gram of dry weight (mg QE/g).

Gas chromatography—mass spectrometry (GC-MS) analysis

Volatile compound analysis was conducted on samples using the Shimadzu GC-2010 system equipped with a GC-MS-QP2020 mass spectrometer. For sample preparation, approximately 2.0 g of each sample was placed into a headspace vial and incubated at 60°C for 30 minutes. The gas chromatography process began with an oven temperature set at 50°C, gradually increasing to 230°C. The injector temperature was maintained at 250°C, with a split ratio of 50:1. Compound separation

was achieved using an SH-I-5 column (30 m × 0.25 mm × 0.25 µm) at a flow rate of 0.99 mL/min. In the mass spectrometry analysis, the ion source temperature was set to 230°C, while the interface temperature was set to 280°C. Scanning occurred in the m/z range of 50–500 at a speed of 1666. Chromatographic data were processed using GC-MS software, and peaks were identified by comparing retention times with known standards (Manalu *et al.,* 2025; Maisaroh *et al.,* 2025).

Statistical analysis

Experiments were conducted using a randomized complete design with two biological replications. Data were statistically analyzed using MS Excel, Minitab 16, and SAS Portable 9.13 software. Both parametric and nonparametric analyses were applied according to the nature of the data. Nonparametric statistics were used to evaluate qualitative (hedonic) data, which were assessed using the Kruskal–Wallis test to determine the effect of formulation on the sensory attributes (flavor, taste, and color) of the herbal tea (Figure 1). Hedonic scale results were expressed as medians (Q_2). Quantitative data were analyzed using one-way analysis of variance (ANOVA), and mean comparisons were performed using Tukey's test at a 5% significance level ($P < 0.05$). All quantitative data are presented as mean ± standard deviation (SD).

Result and Discussion

Organoleptic test

The results of the organoleptic test of moringa leaf herbal tea enriched with ginger and Javanese turmeric are presented in Table 2. Color is critical in shaping the first impression of food quality and acceptance (Nielsen *et al.,* 1998). Sensory evaluation of herbal tea formulations composed of *M. oleifera* leaves, ginger (*Z. officinale*), and Javanese turmeric (*C. xanthorrhiza*) revealed significant differences ($P < 0.05$) in flavor and taste. At the same time, color remained statistically similar across formulations. This suggests that including spices can enhance sensory attributes without altering visual characteristics. Among all tested formulations, F5 (75% moringa, 12.5% ginger, and 12.5% Javanese turmeric) achieved the highest flavor and taste scores (score = 4), indicating optimal sensory balance. The improved acceptance of F5 is attributed to the masking effect of ginger's pungency (gingerols) and turmeric's earthy-warm notes (curcuminoids), which offset moringa's inherent bitterness and grassy flavor. In addition, volatile compounds from the spices contribute to enhanced aromatic complexity and overall palatability (Vázquez-Araújo *et al.,* 2010). The dominance of taste and flavor over color as determinants of consumer preference aligns with previous findings that sensory pleasure strongly influences food acceptance and purchasing behavior (Urala & Lähteenmäki, 2004). Moreover, cultural factors shape hedonic responses toward taste perception (Rozin *et al.,* 2006). Therefore, the F5 formulation represents the most favorable blend

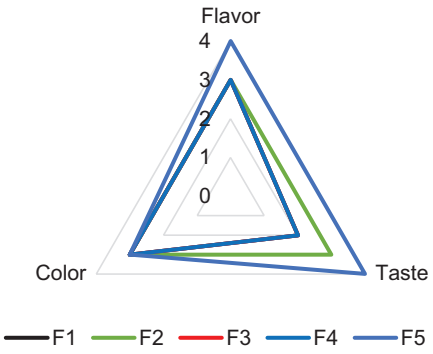

Figure 1. Hedonic sensory attributes.

Table 2. Scores for organoleptic attributes of moringa leaf herbal tea enriched with ginger and Javanese turmeric.

Formula	Flavor		Taste		Color	
	Median (Q2)	Average Ranking	Median (Q2)	Average Ranking	Median (Q2)	Average Ranking
F1	3	38.7	2	35.8	3	42.5
F2	3	45.2	3	48.8	3	51.3
F3	3	42.7	2	45.2	3	45.4
F4	3	47.9	2	43.1	3	47.0
F5	4	65.4	4	67.1	3	53.8
H-value	10.60		13.72		2.09	
P	0.031*		0.008*		0.719ns	

*indicates significant, ns=not significant by the Kruskal–Wallis test.

for consumer acceptance, primarily because of its balanced sensory profile and improved flavor harmony.

Nutritional content of *M. oleifera* herbal tea.

The nutritional composition of *M. oleifera* herbal tea was evaluated to determine the effects of substituting ginger and Javanese turmeric. The results of the proximate analysis for *M. oleifera* herbal tea made with these ingredients are presented in Table 3. This analysis shows variations in nutritional content across the samples tested. The ash content ranged from 8.29% to 9.35%, protein content varied between 21.76% and 28.12%, moisture content was between 8.52% and 9.34%, fat content ranged from 4.27% to 5.09%, and carbohydrate content varied from 49.75% to 55.54%. Significant differences ($p < 0.05$) were found in the protein and ash content among the different *M. oleifera* herbal tea samples made with ginger and Javanese turmeric. However, no significant differences ($p > 0.05$) were noted in moisture, fat, and carbohydrate content across the various formulations of *M. oleifera* herbal tea.

Reducing the concentration of *M. oleifera* leaves in herbal tea formulations by up to 20% was found to decrease the ash content; however, this difference was not statistically significant ($p > 0.05$). This observation may be because of the higher mineral content found in *M. oleifera* leaves compared to ginger and Javanese turmeric. *M. oleifera* leaves are rich in minerals, such as potassium, calcium, sulfur, zinc, magnesium, and copper, which contribute to the ash content identified in this study. A similar trend was observed for protein content, where reducing the concentration of *M. oleifera* leaves resulted in a significant decrease ($p < 0.05$). Numerous studies have reported that *M. oleifera* leaves contain substantial amounts of protein (20–30%) along with significant levels of essential amino acids (Kashyap *et al.,* 2022). Therefore, the reduction in the concentration of *M. oleifera* leaves is responsible for the decline in protein content observed in herbal tea. Furthermore, the addition of ginger and Javanese turmeric could not offset the protein loss because of their inherently low protein content.

Antioxidant activities of *M. oleifera* herbal tea.

Plants contain a variety of chemical compounds, such as phenolic acids, isothiocyanates, tannins, flavonoids, and saponins, which exhibit physiological activity and have applications in food. These compounds may offer therapeutic benefits, including antioxidant properties. The antioxidant activity of *M. oleifera* herbal tea was assessed using methods such as DPPH radical scavenging, total phenolic content, and total flavonoid content. The results of these antioxidant analyses are presented in Table 4.

Table 3. Proximate analysis results of *M. oleifera* leaves herbal tea.

Formula	Moisture (%)	Ash (%)	Protein (%)	Fat (%)	Carbohydrate (%)
F1	8.52 ± 3.98^{a}	9.35 ± 0.12^{a}	28.12 ± 0.04^{a}	4.27 ± 0.48^{a}	49.75 ± 3.66^{a}
F2	9.09 ± 3.16^{a}	8.89 ± 0.23^{ab}	25.62 ± 0.18^{ab}	4.37 ± 0.57^{a}	52.04 ± 2.99^{a}
F3	9.09 ± 3.12^{a}	8.66 ± 0.20^{ab}	25.50 ± 0.19^{b}	4.71 ± 0.45^{a}	52.04 ± 3.57^{a}
F4	9.30 ± 2.77^{a}	8.64 ± 0.22^{ab}	24.32 ± 1.27^{b}	4.80 ± 0.21^{a}	52.94 ± 4.06^{a}
F5	9.34 ± 2.87^{a}	8.29 ± 0.23^{b}	21.76 ± 0.57^{c}	5.09 ± 0.49^{a}	55.54 ± 2.44^{a}

Values are means ± standard deviation of two replications; means followed by different letters in the same column indicate significant differences by the Tukey's test ($p < 0.05$).

Table 4. Antioxidant activities of *M. oleifera* leaves herbal tea.

Formula	DPPH Inhibition (%)	Total Phenolic Content (mg GAE/g)	Total Flavonoid Content (mg QE/g)
F1	81.32 ± 0.15^{a}	1.23 ± 0.28^{a}	1.50 ± 0.37^{a}
F2	74.08 ± 0.00^{a}	1.21 ± 0.26^{a}	1.80 ± 0.50^{a}
F3	77.65 ± 0.15^{a}	1.21 ± 0.00^{a}	1.90 ± 0.25^{a}
F4	79.37 ± 0.15^{a}	1.23 ± 0.05^{a}	2.17 ± 0.51^{a}
F5	79.81 ± 0.15^{a}	1.31 ± 0.22^{a}	2.52 ± 0.30^{a}

Values are means ± standard deviation of two replications; means followed by different letters in the same column indicate significant differences by the Tukey's test ($p < 0.05$).

In the DPPH assay, the scavenging activity of *M. oleifera* leaves herbal tea formulated with ginger and Javanese turmeric was evaluated. The herbal tea with 100% *M. oleifera* leaves demonstrated the highest scavenging activity at 81.32 ± 0.153% (Table 4). When *M. oleifera* leaves powder was replaced with spice powders, no significant difference was observed among the tested formulations ($p \geq 0.05$). The findings indicated that incorporating up to 25% of spices decreased the radical scavenging activity to 79.81%. These results are consistent with previous studies, which reported that *M. oleifera* exhibits radical scavenging activities ranging from 33.80 to 98.43% (Al-Owaisi *et al.*, 2014). The minimal decrease in scavenging activity may be attributed to the addition of ginger and Javanese turmeric, suggesting that these spices could offset the loss of antioxidant compounds from the *M. oleifera* leaves.

Although the DPPH radical scavenging activity decreased from 81.32 ± 0.15% in F1 to 79.81 ± 0.15% in F5, this difference was not statistically significant ($p \geq 0.05$), suggesting that the incorporation of ginger and Javanese turmeric did not markedly impair antioxidant potential. The minimal decline (< 2%) indicates that phenolic and flavonoid compounds from the spices may have compensated for any loss of antioxidant compounds from *M. oleifera* leaves. This is consistent with prior studies reporting that gingerols, shogaols, and curcuminoids possess strong radical-scavenging capacity and can synergize with plant polyphenols to stabilize free radicals (Rahmat *et al.,* 2021; Sharifi-Rad *et al.,* 2017). The reduction in DPPH inhibition observed in some formulations is likely because of matrix effects rather than a loss of active constituents. Overall, the findings demonstrate that incorporating ginger and Javanese turmeric enhances the sensory quality of *M. oleifera* herbal tea while preserving its antioxidant and nutritional performance. Therefore, the F5 formulation can be considered optimal, balancing sensory acceptability with functional stability.

The radical scavenging activity in the herbal tea is closely associated with its phenolic and flavonoid constituents. Although the addition of spices did not significantly affect total phenolic and flavonoid contents ($p \geq 0.05$), the *M. oleifera*–based formulations exhibited phenolic levels of 1.21–1.31 mg GAE/g and flavonoid levels of 1.50–2.52 mg QE/g (Table 4). Previous studies have identified quercetin and kaempferol as the major flavonoids in *M. oleifera* leaves (Makita *et al.,* 2016). The presence of quercetin in ginger may have contributed to the higher flavonoid content in spiced blends. The relatively lower phenolic and flavonoid concentrations compared with previous reports (*Al-Owaisi et al., 2014*; Makita *et al.,* 2016) could be attributed to differences in extraction efficiency, geographical origin, environmental conditions, and postharvest handling. Mild ethanolic steeping typically yields fewer phenolic compounds than methanolic or hydroalcoholic extractions (Chemat *et al.,* 2012), while climatic and soil variations can strongly influence metabolite accumulation (Vázquez-León *et al.,* 2017; Pietrzak *et al.,* 2017). In spite of these lower values, the herbal tea demonstrated strong antioxidant potential, with DPPH inhibition ranging from 74% to 81%. This suggests that even modest phenolics and flavonoids can provide substantial antioxidant effects, possibly through synergistic interactions with other bioactive compounds such as terpenoids and volatile antioxidants (e.g., α-pinene, limonene, and eucalyptol). Similar findings were reported by Don *et al.* (2024) and Devi *et al.* (2024), who observed that combining *M. oleifera* with spices such as ginger and turmeric enhanced its antioxidant properties, in spite of having a moderate total phenolic content.

GC-MS composition of *M. oleifera* leaf herbal tea

The results of the GC-MS analysis are summarized in Table 4. The compounds identified in the analysis primarily belong to the terpene class. One vitamin (a D3 derivative) and one organosulfur compound related to isothiocyanate functionality were also confirmed. However, phenolic acids, flavonoids, tannins, alkaloids, and saponins were undetected. This absence is likely because of the limitations of GC-MS in analyzing nonvolatile, polar, or high-molecular-weight compounds.

The GC-MS analysis of five different formulations (F1 to F5) of *M. oleifera* herbal tea, enriched with ginger and Javanese turmeric, revealed a variety of volatile bioactive compounds. The formulation ratios ranged from 100% *M. oleifera* in F1 to a blend of 75% *M. oleifera* and 25% spices in F5, significantly affecting the quantity and diversity of key terpenoids, hydrocarbons, and aromatic compounds.

Each formulation possesses a unique phytochemical profile, influenced by the ratio of Moringa to spice mixtures (Table 5). Among the terpenes, alpha-pinene, camphene, and limonene are the most abundant, offering significant antioxidant and anti-inflammatory benefits. Notably, higher concentrations of p-cymene, eucalyptol, and γ-terpinene—known for their therapeutic and aromatic properties—are found in formulations containing moderate spice amounts (F2–F3).

The lack of limonene in F5, in spite of its presence in F2–F4, may be attributed to interference from the matrix at higher spice concentrations. In F5, the high levels of dominant terpenes, such as α-pinene, may overshadow or conceal the GC-MS signal of less common volatiles

Table 5. Bioactive compounds identified based on GC-MS analysis (% area).

Compound name	F1 (%)	F2 (%)	F3 (%)	F4 (%)	F5 (%)
Alpha-pinene	15.23	13.62	17.44	5.69	24.36
Camphene	18.57	14.58	10.08	3.80	8.76
Beta-pinene	1.58	1.75	2.01	1.28	2.82
Beta-myrcene	7.60	4.49	4.66	2.78	5.23
Limonene	8.01	11.47	11.50	4.56	0.00
p-Cymene	1.47	1.13	0.96	0.57	1.11
Eucalyptol (1,8-cineole)	0.00	0.00	1.52	2.34	0.00
γ-Terpinene	0.00	0.00	0.87	0.18	0.00
Diallyl disulfide	0.00	0.00	0.00	0.08	0.00
5-Hepten-2-one, 6-methyl-	2.22	2.76	2.89	2.02	4.99
Caryophyllene	0.00	0.00	0.00	0.17	0.17
3-Carene	0.17	0.14	0.00	0.29	0.29
Bicyclo [3.1.0] hexane, 4-methylene-1-(1-methylethyl)-	1.18	1.36	1.42	0.58	1.31
1,25-Dihydroxyvitamin D3, TMS derivative	0.00	0.17	0.00	0.00	0.00

like limonene. This phenomenon, known as ion suppression and co-elution, is well-documented in complex food volatile systems (Furey *et al.,* 2013). Kataoka *et al.* (2000) stated that food matrices rich in essential oil components can diminish detection sensitivity because of competitive ionization and overlapping chromatographic signals.

The concentration of α-pinene showed a significant decrease in F4, in spite of an increase in the spice percentage compared to F3. This reduction could be attributed to several factors, such as extraction saturation, thermal instability, or matrix suppression effects. In dense matrices, other volatile compounds or phenolic substances may interfere with the extraction of α-pinene, reducing its volatility or causing oxidative transformation into unidentified derivatives. Gheorghita *et al.* (2021) noted that α-pinene is susceptible to oxidative breakdown, which can form secondary compounds like verbenone or myrtenol. In addition, Chemat *et al.* (2012) highlight that in complex mixtures with high phenolic content, volatile terpenes may experience decreased extractability or detectability because of sorptive interactions, thermal sensitivity, or co-elution effects.

Adding ginger and Javanese turmeric significantly enhances the diversity and potency of bioactive volatile compounds. This includes introducing or enhancing compounds, such as eucalyptol, γ-terpinene, caryophyllene, and diallyl disulfide, which provide new antimicrobial, anti-inflammatory, and antioxidant properties. In addition, limonene and p-cymene, either absent or found in low concentrations in pure Moringa (F1), are markedly increased in samples F2–F4. This suggests an optimal synergy achieved with a 10–20% addition of spices. Furthermore, the presence of 1,25-dihydroxyvitamin D3 in F2 highlights the potential release of important micronutrients that are metabolically relevant.

Adding ginger and Javanese turmeric to *M. oleifera* leaf tea has significantly enhanced its volatile terpenoid profile, as GC-MS analysis shows. The monoterpene α-Pinene, a key component found in Moringa, was notably increased to 24.36% in formulation F5, representing a 25% rise with the added spices. New terpenes such as γ-terpinene, eucalyptol (1,8-cineole), and β-foreseen were also identified in the enriched formulations, particularly in F3 and F4. These compounds were either absent or present in only trace amounts in the 100% Moringa sample (F1). These compounds are recognized for their antioxidant, anti-inflammatory, and antimicrobial properties, thus boosting the functional benefits of the beverage (Estiasih *et al.,* 2025).

The addition of ginger and Javanese turmeric may lead to a synergistic increase in antioxidant volatiles, such as limonene, which is a key antioxidant terpene found abundantly in citrus and ginger oils. Limonene was detected in pure Moringa (F1) but peaked in formulations F2 and F3, reaching approximately 11.5%. Similarly, p-cymene, a monoterpene known for its potent antioxidant properties, was consistently present in all enriched formulations, with a concentration of 1.11% in F5. Including γ-terpinene and diallyl disulfide, compounds commonly associated with ginger and turmeric, contributes significantly to reactive oxygen species (ROS) scavenging capabilities. These findings suggest that a moderate addition

of spices (F2–F4) is optimal for enhancing antioxidant effectiveness, owing to the synergistic release of volatiles from these spices.

The enriched formulations introduce new compounds that are not typically abundant in Moringa. These include Eucalyptol (1,8-cineole) found in formulations F3 and F4, Caryophyllene present in F4 and F5, Diallyl disulfide present in F4, and a Vitamin D3 metabolite present in F2. Most of these compounds are primarily sourced from ginger and Javanese turmeric, indicating that the spice mix enhances the health benefits of the tea beyond what Moringa alone can provide.

Adding *Z. officinale* and *C. xanthorrhiza* optimizes the formulation. Based on the evidence of GC-MS, F2–F4 (10–20% spice enrichment) provides the most balanced profile, maximizing limonene, p-cymene, and eucalyptol without losing core moringa compounds. F5 maximizes α-pinene but shows reduced diversity in other antioxidants because of potential volatile suppression or matrix effects.

The addition of ginger and Javanese turmeric to *M. oleifera* herbal tea boosts the variety and concentration of bioactive volatile compounds; improves antioxidant, antimicrobial, and anti-inflammatory properties; and brings in unique pharmacologically significant molecules that are not found in pure moringa tea.

Therefore, ginger and Javanese turmeric not only enhance the phytochemical profile of *M. oleifera* but also work together to increase its health benefits, especially in blends containing 10–15% spice content. These modifications indicate that incorporating spices increases the chemical complexity and broadens the herbal tea's functional range, spanning from immune modulation to anti-inflammatory and respiratory assistance (Balasubramaniam *et al.,* 2024). Formulations F2 and F4 offer the optimal balance between the bioactivity of *M. oleifera* and the enhancements derived from spices, whereas F5 stands out in optimizing the concentration of alpha-pinene.

The results indicate that the concentrations of bioactive compounds vary significantly depending on the formulation strategy. Combining *M. oleifera* with ginger and Javanese turmeric alters the chemical profile and may enhance the bioactivity of the final tea product. This suggests that targeted formulation is an effective strategy for optimizing the tea's therapeutic and sensory qualities.

The GC–MS analysis demonstrated that blending moringa with ginger and Javanese turmeric significantly modified the profile of volatile and bioactive compounds in tea formulations. The primary volatiles identified included monoterpenes and sesquiterpenes, particularly α-pinene, camphene, limonene, and p-cymene, all of which have been reported to possess antioxidant, antimicrobial, and anti-inflammatory properties (Abdelmohsen *&* Elmaidomy, 2025; Baginska *et al.*, 2023; Silva *et al.*, 2021). The observed increase in α-pinene and limonene in F2–F3 indicates a synergistic interaction between *M. oleifera* and the spice volatiles, which likely enhances aroma and freshness perception, as these terpenes are recognized contributors to pleasant, citrus-like sensory characteristics in herbal beverages (Zhang *et al.*, 2024).

While the compounds detected are linked to biological activity, the current results are confined to in vitro antioxidant evaluations. The bioavailability and actual intake of these volatiles in a standard tea serving remain ambiguous. Many terpenes, while potent in vitro, are subject to rapid metabolism and demonstrate limited systemic absorption when ingested orally (Bakkali *et al.*, 2008). Consequently, the potential health benefits discussed herein are inferential, grounded in chemical composition rather than validated physiological effects.

The sensory perception of the *M. oleifera–based* herbal tea formulations was closely associated with their chemical composition, particularly the presence of phenolic and volatile compounds derived from both *M. oleifera* and the added spices, including Javanese turmeric, which notably influenced the perception of tea's color and bitterness. Curcuminoids and related diarylheptanoids in turmeric are responsible for its golden-yellow color and mild bitterness. This may have contributed to a more visually appealing but astringent sensory profile at higher inclusion levels (F4–F5). However, this bitterness was effectively masked by the pungent and warm notes of ginger, dominated by gingerols and shogaols, which provided balance and depth to the overall flavor. GC–MS results showing increased concentrations of terpenes such as α-pinene, limonene, and eucalyptol in the spice-enriched samples correlate with the panelists' higher ratings for flavor and aroma, as these volatiles impart fresh, citrusy, and herbal notes known to enhance palatability (Zhang *et al.,* 2024).

Interestingly, while the total phenolic and flavonoid contents did not differ significantly among formulations, the panelists' preferences (particularly for F5) suggest that sensory appeal was more influenced by volatile composition than by antioxidant potency per se. Nevertheless, a weak positive relationship between phenolic concentration and sensory scores can be inferred, since formulations with higher phenolic values (e.g., F5: 1.31 mg GAE/g) also showed better perceived body and flavor intensity. This may reflect the contribution of phenolics to mouthfeel and astringency, which, in moderate levels, can enhance the perception of "richness" in herbal

infusions (Vázquez-Araújo *et al.,* 2010). Therefore, the optimized formulation (F5) balanced the bioactive compound profile and sensory properties, where the interplay of terpenes, phenolics, and curcuminoids created a harmonious flavor while maintaining antioxidant capacity.

Overall, the study demonstrates that adding spices can enhance both the functional and sensory qualities of *M. oleifera*–based teas. However, several limitations should be acknowledged. The experiments were conducted with limited replication, which may affect the statistical robustness and generalizability of the results. Although GC–MS analysis provided helpful information on volatile compounds, the absence of complementary chromatographic techniques such as HPLC or LC–MS limited the identification and quantification of nonvolatile phenolic and flavonoid compounds. As a result, some bioactive constituents may have remained undetected. To strengthen the understanding of these findings, future studies should include in vivo evaluations and bioaccessibility assessments, and employ LC-MS-based profiling to comprehensively characterize phenolic and flavonoid compounds.

Conclusions

The findings of this study indicate that incorporating ginger and Javanese turmeric into *M. oleifera* herbal tea enhances its overall taste and flavor. Interestingly, the addition of these ingredients—particularly in the F5 formula—did not cause significant changes in carbohydrate, fat, moisture, antioxidant activity, total flavonoid, or phenolic content. However, there was a reduction in protein and ash content. Sensory evaluation results showed that the F5 formula was the most preferred by panelists because of its pleasant taste and aroma, while still maintaining desirable antioxidant and nutritional characteristics. Therefore, the F5 formulation represents a promising option for developing *M. oleifera* leaf herbal tea that combines improved sensory quality with preserved functional properties.

Authors Contributions

All authors contributed equally to this article.

Conflicts of Interests

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