



Physicochemical Analysis, Caffeine Quantification, and Sensory Profiling of *Oenanthe javanica* Tea

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Abstract

Herbal tea infusions have gained increasing attention due to their potential health-promoting and therapeutic properties. *Oenanthe javanica* has been recognised for its antioxidant and detoxifying attributes; however, its sensory and physicochemical characteristics remain insufficiently explored. This study aimed to determine caffeine concentrations across different formulations, evaluate key physicochemical properties, and assess consumer sensory acceptance. Leaf samples were analysed through moisture content determination, controlled brewing conditions (water volume, temperature, and storage), Fourier transform infrared spectroscopy (FTIR), spectrophotometric caffeine quantification, and sensory evaluation involving 30 panellists. Physicochemical analysis showed that dried *O. javanica* leaves exhibited slightly higher moisture content than *Camellia sinensis* (black tea). FTIR analysis confirmed the presence of key functional groups, including C–H, C=O, C=C, and C–N bonds, which contribute to its chemical profile. Caffeine quantification confirmed its presence in *O. javanica*, while black tea consistently exhibited the highest caffeine content, and the 1:1 blended formulation showed the lowest concentration due to dilution effects. The calibration model based on the Beer–Lambert law demonstrated strong linearity for caffeine quantification. In addition, brewing conditions and storage significantly influenced the physicochemical properties of the infusions, where lower solvent volumes produced more concentrated, acidic, and caffeine-rich extracts. Sensory evaluation revealed that pure *O. javanica* tea received lower overall hedonic acceptance compared with black tea, while the blended formulation showed improved acceptability. Overall, the findings provide new insights into the physicochemical and sensory characteristics of *O. javanica*, supporting its potential application as a functional ingredient in herbal tea product development.

Keywords: *Oenanthe javanica*, herbal infusion, ATR-FTIR, caffeine, physicochemical analysis, sensory evaluation

1. Introduction

Herbal teas, also known as tisanes, represent a rapidly growing segment of the global functional beverage market. Unlike traditional tea, which is derived exclusively from the leaves of *Camellia sinensis*, herbal teas are prepared from various plant materials, including leaves, seeds, bark, fruits, flowers, and grasses obtained from medicinal plants [1]. These beverages have long been incorporated into traditional diets and are increasingly consumed because of their potential health-promoting properties, including antioxidant, anti-inflammatory, antibacterial, antidiabetic, and anticancer activities [2,3]. Their biological activities are primarily attributed to the presence of diverse phytochemicals, such as polyphenols, flavonoids, terpenoids, alkaloids, and polysaccharides [4].

Oenanthe javanica (Blume) DC., commonly known as water dropwort, is an aromatic perennial herb belonging to the Apiaceae family. It is widely distributed throughout tropical and temperate regions of Asia and is known locally as *Shui qin* in China, *Seri* in Japan, *Minari* in Korea, *Phak-chilom* in Thailand, and *Selom* in Malaysia [5,6]. Traditionally, *O. javanica* has been

consumed as both a vegetable and a medicinal herb. Recent studies have demonstrated a wide range of biological activities, including antioxidant, anti-quorum sensing, antidiabetic, neuroprotective, hepatoprotective, alcohol-detoxifying, cardiovascular protective, memory-enhancing, and anti-hepatitis B virus activities [7–10].

Functional beverages are defined as drinks containing bioactive ingredients that provide physiological benefits beyond basic nutrition and hydration [11]. Increasing consumer awareness of health and wellness has driven demand for beverages with reduced or no caffeine content while maintaining desirable functional properties. Caffeine (Figure 1), chemically known as 1,3,7-trimethylxanthine, is a naturally occurring alkaloid widely consumed for its stimulant effects on the central nervous system [12]. Although moderate caffeine intake enhances alertness and reduces fatigue, excessive consumption has been associated with adverse health effects, including restlessness, insomnia, gastrointestinal disturbances, tachycardia, and cardiovascular complications [13–15]. Consequently, there is growing interest in identifying plant-based ingredients that offer

functional health benefits while naturally containing lower levels of caffeine.

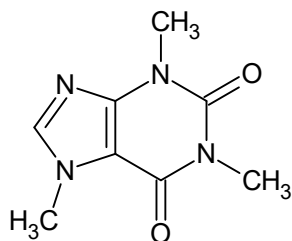


Figure 1. Chemical structure of caffeine.

Despite the long-standing traditional use and recognised medicinal value of *O. javanica*, information regarding its physicochemical properties, caffeine content, and sensory characteristics as a herbal tea infusion remains limited. To address this knowledge gap, the present study comprehensively evaluated the effects of different *O. javanica* formulations on moisture content, physicochemical properties, molecular functional groups characterised by ATR-FTIR spectroscopy, caffeine concentration, and pH under different brewing conditions, including variations in water volume, brewing temperature, and storage duration. In addition, consumer acceptability and sensory characteristics of the tea formulations were assessed through sensory evaluation involving 30 untrained panellists.

2. Research Methods

2.1 Plant Material Processing

Fresh *Oenanthe javanica* leaves were purchased from a local market in Tapah, Perak, Malaysia. Diseased, damaged, and wilted leaves were discarded prior to processing. The selected leaves were thoroughly washed, cut into small pieces, and dried in a hot-air oven at 50 °C until a constant weight was achieved. The dried leaves were then ground into a fine powder and stored in airtight containers until further analysis.

2.2 Moisture Content Determination

Approximately 3 g of each tea sample was weighed into pre-weighed Petri dishes and dried in a hot-air oven at 50 °C until a constant weight was obtained. All measurements were performed in triplicate. The moisture content (%) was calculated using the following equation:

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

2.3 Characterization of Molecular Functional Groups by ATR-FTIR

Attenuated Total Reflectance-Fourier Transform Infrared (ATR-FTIR) spectroscopy was employed to characterise the molecular functional groups of the tea

samples. Infrared spectra were recorded over the mid-infrared region within the wavenumber range of 4000–400 cm⁻¹ to identify the characteristic absorption bands corresponding to the functional groups present in the samples.

2.4 Quantitative Determination of Caffeine by Solvent Extraction and UV-Vis Spectrophotometry

2.4.1 Preparation of tea samples

Tea samples were prepared by blending powdered *O. javanica* and *Camellia sinensis* (black tea) in different proportions: 100% *O. javanica* (OC1), 50:50 (OC2), and 100% black tea (OC3). Each formulation was prepared to a constant total weight of 2 g. The samples were infused in 200 mL of hot distilled water, gently stirred, and allowed to steep for 5 min.

Following filtration, 2 g of anhydrous sodium carbonate (Na₂CO₃) was added to the filtrate to reduce interference from polyphenolic compounds. The mixture was stirred thoroughly and allowed to stand for 5 min before being filtered again. Subsequently, a 10 mL aliquot of the filtrate was transferred to a separatory funnel and extracted with 10 mL of chloroform by vigorous shaking for 5 min. The mixture was then allowed to stand for 10 min to achieve complete phase separation. The lower chloroform layer containing the extracted caffeine was carefully collected for subsequent UV-Vis analysis.

2.4.2 Preparation of standard caffeine solutions

A 100 ppm caffeine stock solution was prepared by dissolving 0.01 g of analytical-grade caffeine in 100 mL of chloroform. Working standard solutions (10–40 ppm) were prepared by appropriate dilution of the stock solution. Absorbance measurements were recorded using a UV-Vis spectrophotometer over the wavelength range of 190–400 nm, and the maximum absorption wavelength (λ_{max}) was determined at 274 nm.

2.5 Effect of Brewing Conditions and Storage on pH

The pH of the tea infusions was measured using a calibrated digital pH meter.

2.5.1 Effect of water volume

To evaluate the effect of brewing water volume, 1 g of each tea formulation was infused in 50, 100, and 150 mL of boiling water for 5 min. The infusions were cooled to approximately 50 °C, filtered through Whatman No. 1 filter paper, and their pH values were measured.

2.5.2 Effect of water temperature

The effect of brewing temperature was investigated by infusing 1 g of each tea formulation in 100 mL of water under three different conditions: hot water (100 °C),

room temperature (25 °C), and cold water (ice bath) for 30 min.

2.5.3 Effect of storage

To evaluate the effect of storage, 1 g of each tea formulation was brewed in 100 mL of boiling water. The infusions were cooled to room temperature (25 °C), transferred into sealed containers, and stored at 5 °C for four weeks. The pH of each sample was measured at two-week intervals throughout the storage period.

2.6 Sensory Evaluation Procedure

Sensory evaluation was conducted to assess the consumer acceptability of the prepared tea formulations. Thirty untrained panellists were recruited from the student population of Universiti Teknologi MARA (UiTM) Perak Branch, Tapah Campus. The identity and composition of the tea samples were concealed throughout the evaluation to minimise bias.

Three tea formulations, namely *O. javanica* tea, black tea, and a 1:1 blend of *O. javanica* and black tea, were served in identical paper cups at approximately 60 °C, with each panellist receiving 15 mL of each sample. Panellists were instructed to hold each sample in the mouth for approximately 5 s before swallowing. Between samples, they rinsed their mouths with warm water and rested for 1–2 min to minimise carry-over effects.

The tea samples were evaluated based on appearance, visual consistency, aroma, taste, overall satisfaction, enjoyment, aftertaste, and overall quality using a 5-point hedonic scale, where 1 represented "dislike very much" and 5 represented "like very much". Mean sensory scores were calculated and presented as radar plots for comparison among the tea formulations.

3. Results and Discussion

3.1 Moisture Content Analysis

Moisture content is an important quality parameter that influences the stability, shelf life, and susceptibility of food products to microbial deterioration. Excess moisture promotes the growth of bacteria, yeasts, and moulds during storage, resulting in quality deterioration, undesirable odours, and losses in nutritional and functional properties [16]. Consequently, moisture determination is essential for evaluating drying efficiency and establishing appropriate storage and packaging conditions for dried herbal products [17].

The recommended moisture content for dried herbal teas is generally below 11%, as higher moisture levels increase the risk of microbial spoilage during storage [18]. In the present study, the fresh *O. javanica* leaves exhibited a moisture content of 22.48%, whereas commercial black tea contained approximately 22.00%. The slightly higher moisture content of *O. javanica* may be attributed to differences in leaf structure and

processing. Commercial black tea undergoes withering, rolling, enzymatic oxidation, and final drying, processes that disrupt cellular structures and facilitate moisture removal [19,20]. In contrast, *O. javanica* leaves undergo less extensive processing prior to drying, allowing greater retention of inherent moisture within the plant tissues [21,22]. Therefore, adequate drying was performed to reduce the moisture content to below the recommended limit, thereby enhancing the storage stability of the tea formulations.

3.2 ATR-FTIR Characterisation of Tea Formulations

ATR-FTIR spectroscopy was employed to identify the characteristic functional groups present in the tea formulations based on their infrared absorption bands. The spectra obtained exhibited distinct absorption patterns corresponding to the major chemical constituents of each tea sample (Figure 2) [23].

A broad absorption band in the region of 3200–3500 cm^{-1} was observed in all tea formulations and was attributed to O–H stretching vibrations of hydroxyl groups present in polyphenols, polysaccharides, and carbohydrates [24–26]. Black tea exhibited a more intense O–H stretching band centred at 3287.69 cm^{-1} , indicating a relatively higher abundance of hydroxyl-containing compounds, particularly phenolic constituents. In contrast, the intensity of this band decreased in the blended formulation, which may reflect interactions between phytochemical constituents or differences in the relative abundance of hydroxyl-containing compounds following blending [27].

Absorption bands within the region of 2920–2854 cm^{-1} were assigned to the C–H stretching vibrations of aliphatic methylene groups, indicating the presence of saturated hydrocarbon chains. A distinct absorption band at 2921.34 cm^{-1} was observed for *O. javanica*, suggesting the presence of aliphatic constituents within the leaf matrix. The absorption bands between 1640 and 1746 cm^{-1} corresponded to C=O stretching vibrations, which are commonly associated with carbonyl-containing compounds such as polyphenols, fatty acids, and amino acids [26]. Furthermore, characteristic absorption bands observed at approximately 1615 and 1450 cm^{-1} were attributed to aromatic C=C stretching vibrations, confirming the presence of aromatic compounds in all tea formulations. Compared with the individual tea samples, the blended formulation exhibited a slight reduction in the intensity of the bands at approximately 1613.55 and 1605.92 cm^{-1} , which may be attributed to interactions between phytochemical constituents or changes in their relative composition following blending rather than structural degradation.

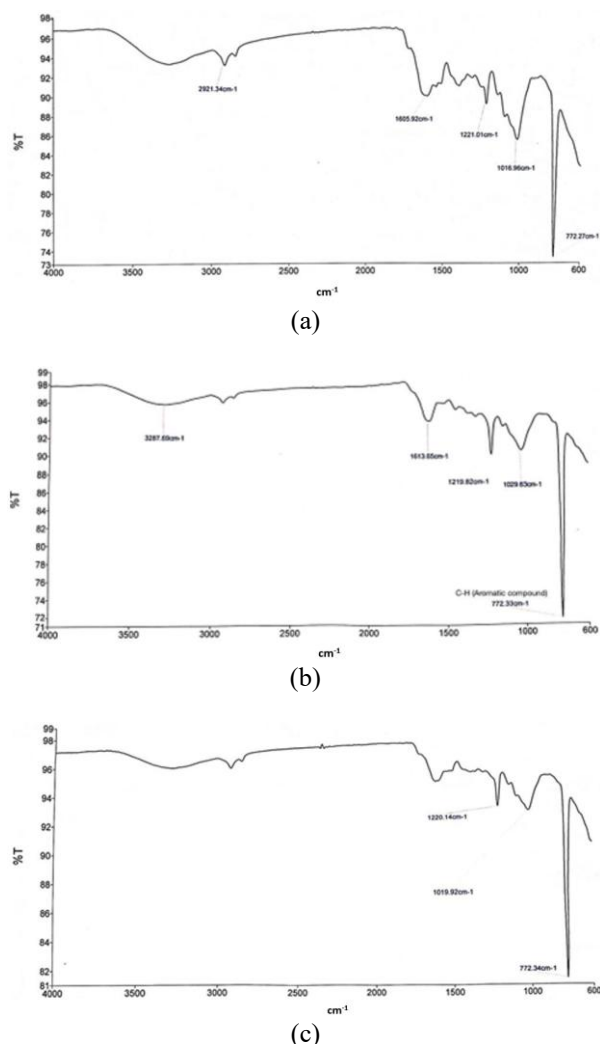


Figure 2. FTIR spectra for various types of tea formulations: (a) *O. javanica*, (b) black tea, and (c) a mixture of *O. javanica* and black tea.

3.3 Spectrophotometric Quantification of Caffeine

The calibration curve constructed using standard caffeine solutions exhibited excellent linearity over the concentration range investigated, consistent with the Beer–Lambert law. The regression equation obtained was $y = 0.0483x + 0.0269$, with a correlation coefficient (R^2) of 0.9929 (Figure 3), indicating a strong linear relationship between caffeine concentration and absorbance and confirming the suitability of the method for caffeine quantification [28].

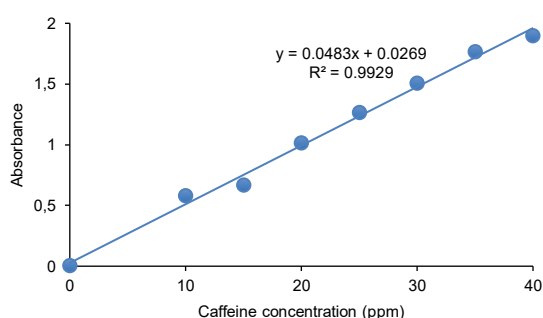


Figure 3. The standard curve of caffeine.

Caffeine was detected in all three tea formulations. Among the crude aqueous extracts, commercial black tea (OC3) exhibited the highest caffeine concentration (9.01 ppm), followed by *O. javanica* (OC1) at 6.36 ppm. Although *O. javanica* contained less caffeine than black tea, it is recognised as a rich source of bioactive phytochemicals, including chlorogenic acid, persicarin, flavonoids, and caffeic acid derivatives, which contribute to its reported health-promoting properties [29]. The blended formulation (OC2) recorded the lowest caffeine concentration (5.72 ppm), which may be attributed to the dilution of caffeine-containing constituents resulting from the combination of *O. javanica* and black tea, thereby reducing the relative caffeine concentration in the final infusion [30].

The relatively high caffeine concentrations observed in the crude aqueous extracts can be attributed to the increased solubility of caffeine in water at elevated extraction temperatures, which enhances its extraction efficiency [31]. However, lower caffeine concentrations were detected following liquid–liquid extraction with chloroform. This observation may reflect the partitioning behaviour of caffeine between the aqueous and organic phases. Because caffeine distribution is governed by partition equilibrium, a proportion of the compound remains in the aqueous phase at equilibrium, thereby reducing the amount recovered in the chloroform layer [32].

3.4 Effect of Extraction Conditions and Storage on pH

The pH, which reflects the acidity or alkalinity of a solution, was measured using a calibrated pH meter. The results demonstrated that brewing conditions and storage significantly influenced the pH of the tea formulations.

3.4.1 Effect of water volume on the pH of tea infusions

The influence of brewing water volume on pH is presented in Figure 4. For black tea (OC3), the pH increased from 5.29 at 50 mL to 5.32 and 5.47 at 100 and 150 mL, respectively. A similar trend was observed for *O. javanica* tea (OC1), where the pH increased from 5.91 to 6.03 and 6.07, while the blended formulation (OC2) exhibited corresponding increases from 5.54 to 5.65 and 5.73. These findings indicate that increasing the brewing water volume generally produced less acidic tea infusions.

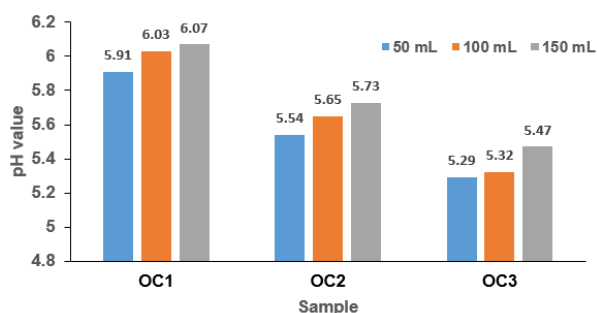


Figure 4. The pH value of each tea infusion with brewing-volume variation.

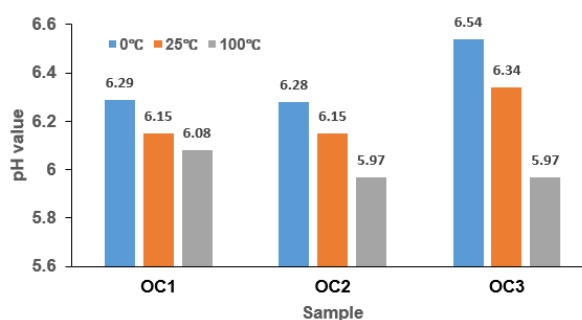


Figure 5. The pH value of each tea infusion with brewing-temperature variation.

The increase in pH can be attributed primarily to the dilution of water-soluble acidic constituents extracted during brewing. Tea infusions contain a complex mixture of polyphenols, organic acids, amino acids, caffeine, and mineral constituents that collectively determine their physicochemical properties [33]. As the amount of water increased while the quantity of tea remained constant, the concentration of these dissolved compounds decreased, resulting in a gradual increase in pH. Similar observations have been reported in studies demonstrating that extraction conditions, particularly solvent volume and extraction parameters, significantly affect the concentration of bioactive compounds released into tea infusions [34,35].

Black tea naturally contains caffeine because it is produced from *Camellia sinensis*, whereas *O. javanica* is not recognised as a natural source of caffeine [36]. Consequently, the blended formulation was expected to contain less caffeine than pure black tea. Nevertheless, the higher pH observed at larger brewing volumes is more likely attributable to dilution of acidic phytochemicals than to changes in caffeine concentration. Overall, increasing the brewing water volume produced more dilute infusions with slightly higher pH values.

3.4.2 Effect of water temperature on the pH of tea infusions

The effect of brewing temperature on pH is illustrated in Figure 5. Black tea exhibited pH values of 5.97, 6.54, and 6.34 when brewed with hot (100 °C), cold, and room-temperature water, respectively. Similar trends were observed for *O. javanica* tea and the blended formulation, with hot-water extraction consistently producing lower pH values than cold or room-temperature brewing.

Brewing temperature is a critical factor governing the extraction of water-soluble constituents from tea leaves. Elevated temperatures enhance both the diffusion and solubility of polyphenols, organic acids, caffeine, and other phytochemicals, thereby increasing their extraction efficiency [37,38]. Consequently, hot-water extraction produces more concentrated infusions containing greater amounts of acidic constituents, resulting in lower pH values. Conversely, cold-water extraction proceeds more slowly and extracts fewer soluble compounds, yielding less acidic infusions with comparatively higher pH values. These findings are consistent with previous studies demonstrating that brewing temperature substantially influences the chemical composition and physicochemical properties of tea infusions.

3.4.3 Effect of storage on the pH of tea infusions

The influence of refrigerated storage on pH is presented in Figure 6. The initial pH values of black tea, *O. javanica* tea, and the blended formulation were 5.75, 6.06, and 5.85, respectively. After two weeks of storage at 5 °C, the pH of all formulations decreased slightly. However, after four weeks, the pH increased modestly, with black tea exhibiting the greatest recovery. These observations indicate that refrigerated storage induced gradual physicochemical changes in the tea formulations over time.

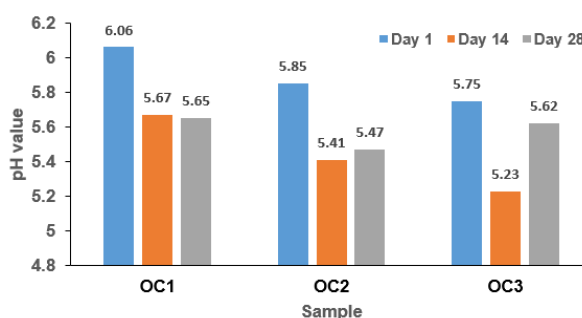


Figure 6. The pH value of each tea infusion stored in a refrigerator at approximately 5 °C for 28 days.

The observed pH fluctuations may be associated with gradual physicochemical transformations occurring during storage. Tea polyphenols are susceptible to

oxidation and other structural modifications, and their stability is influenced by storage conditions such as temperature, oxygen availability, and light exposure [39,40]. Although refrigeration slows these reactions, it does not completely inhibit changes in phytochemical composition. Previous studies have also reported that storage alters the chemical profile and quality characteristics of tea through continuous changes in its bioactive constituents [41]. Therefore, the slight increase in pH after four weeks may reflect the gradual stabilisation of dissolved phytochemicals within the tea infusions.

3.5 Sensory Evaluation and Consumer Acceptability

Sensory evaluation was conducted with 30 untrained panellists to assess the consumer acceptability of the three tea formulations. The evaluation comprised seven sensory attributes, namely appearance, visual consistency, aroma, taste, overall satisfaction, enjoyment during tasting, and aftertaste.

For *O. javanica* tea, the sensory profile is presented in Figure 7. The infusion was generally well accepted in terms of appearance and visual consistency, with only a small proportion of panellists expressing negative responses. However, lower acceptance was observed for aroma and taste, each receiving 14 negative responses. Likewise, overall satisfaction, enjoyment during tasting, and aftertaste were rated less favourably than the visual attributes. These findings suggest that although *O. javanica* tea possessed acceptable visual characteristics, its flavour profile was less preferred by consumers.

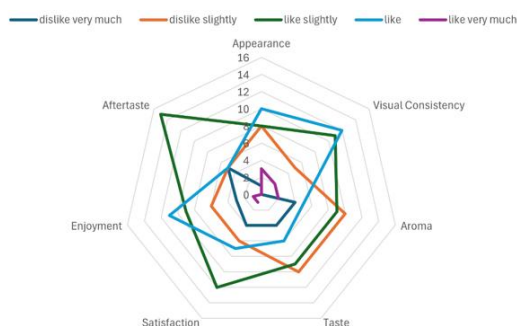


Figure 7. Radar plot of the sensory attributes of *Oenanthe javanica* tea.

Black tea demonstrated the highest sensory acceptance among all formulations (Figure 8). Most panellists rated its appearance, visual consistency, aroma, taste, and overall sensory quality favourably, with relatively few negative responses recorded across all evaluated attributes. The consistently high scores indicate that black tea possessed the most acceptable sensory profile among the three formulations.

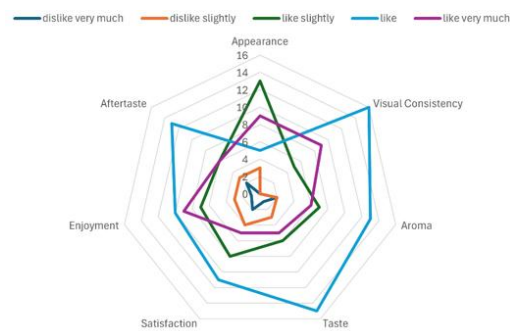


Figure 8. Radar plot of the sensory attributes of black tea.

The blended formulation exhibited intermediate sensory performance between pure *O. javanica* tea and black tea (Figure 9). Compared with *O. javanica* tea, the blend received fewer negative responses for aroma, taste, overall satisfaction, enjoyment during tasting, and aftertaste, indicating that the incorporation of black tea improved the overall sensory characteristics of the herbal infusion.

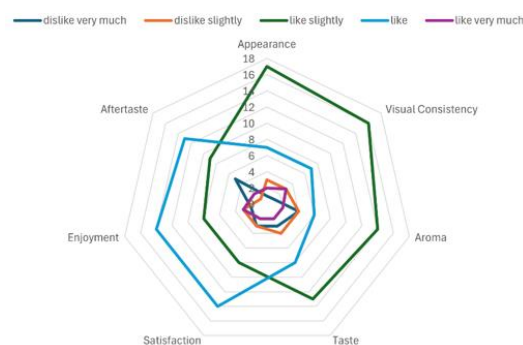


Figure 9. Radar plot of the sensory attributes of the blended tea formulation.

Overall, the radar plots demonstrated that black tea produced the largest polygon area, reflecting the highest level of consumer acceptance, whereas *O. javanica* tea generated the smallest polygon, indicating the lowest overall preference. The blended formulation showed intermediate acceptability, suggesting that blending *O. javanica* with black tea effectively improved its sensory quality. Consumer preference for tea is strongly influenced by key sensory attributes, particularly aroma, taste, appearance, and aftertaste, which collectively determine overall acceptability [42]. The comparatively lower acceptance of *O. javanica* tea may be associated with its characteristic herbal flavour and slight bitterness, attributes that are frequently reported to reduce consumer preference for herbal beverages [43]. Nevertheless, the improved sensory performance of the blended formulation suggests that incorporating conventional black tea can effectively enhance consumer acceptability while maintaining the potential functional benefits of *O. javanica*.

4. Conclusion

This study demonstrated that *Oenanthe javanica* possesses favourable physicochemical characteristics and a lower caffeine content than *Camellia sinensis* (black tea), highlighting its potential as a naturally low-caffeine herbal tea ingredient. Brewing conditions, including water volume, temperature, and storage, significantly influenced the pH and physicochemical properties of the tea infusions. Sensory evaluation further showed that blending *O. javanica* with black tea improved consumer acceptance compared with pure *O. javanica* tea, although black tea remained the most preferred formulation. Overall, these findings support the potential application of *O. javanica* in the development of functional herbal tea products. Future studies should focus on optimizing formulation strategies and conducting comprehensive phytochemical characterisation to further enhance product quality and commercial potential.

Acknowledgement

The authors would like to express their sincere appreciation to the Faculty of Applied Sciences, Universiti Teknologi MARA (UiTM) Perak Branch, Tapah Campus, for providing the laboratory facilities, equipment, and technical assistance necessary for the successful completion of this research.

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