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ORIGINAL RESEARCH ARTICLE

Fortification of Cakes with Guava (*Psidium guajava*) Fruit and Leaf Powders: Effects on Nutritional Quality, Bioactive Compounds, Antioxidant Activity, and α-Amylase Inhibition

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ABSTRACT

This study examined the impact of fortifying cakes with guava (*Psidium guajava*) fruit and leaf powder on their nutritional content, bioactive substances, antioxidant activity, antidiabetic potential, and sensory characteristics. Cakes were produced using varying concentrations of guava fruit and leaf powders, thereafter subjected to proximate, mineral, phytochemical, antioxidant, α-amylase inhibition, and sensory evaluations. Fortified cakes exhibited enhanced levels of protein, dietary fiber, and minerals in comparison to the control group. Leaf-fortified formulations displayed elevated levels of total phenolics, flavonoids, anthocyanins, and antioxidant activities, whereas fruit-fortified cakes showed superior sensory acceptance. The findings indicate that guava fruit and leaf powders can serve as functional components in baked goods, with leaf powder providing superior functional advantages and fruit powder ensuring consumer acceptability.

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1. INTRODUCTION

The global rise in chronic diseases like diabetes, cardiovascular disorders, and obesity has fueled interest in functional foods that provide benefits beyond basic nutrition. These foods deliver bioactive compounds that support health and reduce disease risk. Incorporating plant-based ingredients into everyday products is a promising strategy, and guava (*Psidium guajava*), a nutrient-rich tropical fruit, has gained attention for its strong potential as a functional food component.

Guava is rich in vitamin C, dietary fiber, minerals, and bioactive compounds like polyphenols, flavonoids, carotenoids, and anthocyanins, which show antioxidant, anti-inflammatory, anti-diabetic, and anti-cancer effects (Gutiérrez et al., 2008; Kumar et al., 2021a). Guava leaves contain even higher concentrations of bioactive compounds than the fruit, making both valuable for functional food development (Naseer et al., 2018).

Research supports incorporating guava fruit and leaf powders into foods, especially baked goods, to boost nutrition and bioactivity. Guava fruit powder has been used in smoothies, juices, and cakes to enhance antioxidant capacity, phenolic and flavonoid content, fiber, and nutrient density (Angulo-López et al., 2021; Bano et al., 2025). Fortification of baked products with fruit powders is known to improve nutritional composition and provide functional benefits like antioxidant and anti-diabetic activity (Tolve et al., 2021). Similarly, guava leaf powder, rich in polyphenols such as tannins and flavonoids, contributes strong antioxidant and anti-inflammatory effects (Huynh et al., 2025).

In addition to its antioxidant properties, guava leaves have been shown to exhibit potent anti-diabetic effects. Guava leaf extracts have demonstrated the ability to inhibit α-amylase, an enzyme responsible for starch digestion, thus reducing postprandial blood glucose levels and improving glycemic control in diabetic individuals (Huang et al., 2021). These findings highlight the potential of guava leaf powder as an effective anti-diabetic agent, which could be integrated into functional food formulations to address the growing prevalence of type 2 diabetes worldwide.

Despite extensive reports on the nutritional and functional properties of guava fruit, the utilization of guava leaves in bakery applications remains limited. Guava leaves are rich in phenolic compounds and have demonstrated antioxidant and antidiabetic potential; however, their incorporation into commonly consumed foods poses sensory challenges. Therefore, this study aimed to evaluate the effects of guava fruit and leaf powder fortification on the nutritional quality, bioactive composition, antioxidant activity, antidiabetic potential, and sensory characteristics of cake formulations.

2. MATERIALS AND METHODS

2.1 Sample collection and preparation

Fresh guavas (*Psidium guajava*) and guava leaves were procured from the local market in Chattogram. The average weight of the fruits was 115 ± 3.2 g. Both fruits and leaves were stored in a refrigerator at 3-4 °C until further use. Prior to processing, the guavas and leaves were thoroughly washed with tap water and the leaves were destemmed. The guava fruit was cut into uniform pieces of approximately 2 × 2 cm, and the leaves were trimmed to a similar size for consistency. Both the fruit and leaves were subsequently utilized for all experimental treatments. To minimize variability, all guava fruits and leaves were collected from a single batch acquired simultaneously.

2.2 Study design

A single batch of cake was prepared for each treatment to maintain consistency. Guava fruits and leaves were washed, dried, ground into fine powder, and used to replace wheat flour at 3% and 5% levels. A control cake without substitution was also prepared. Cakes underwent proximate analysis, mineral profiling, and bioactive compound quantification. In vitro anti-diabetic activity was assessed. All measurements were performed in triplicate, averaged per batch, and analyzed using one-way ANOVA with Tukey's post hoc test at $p \leq 0.05$. For sensory evaluation, randomized portions from each batch were presented to panelists, who scored color, aroma, taste, texture, and overall acceptability.

2.3 Drying of fruit and leaves

After cutting the fruits and leaves, any excess liquid was removed by wiping, and they were dried at 50°C for 72 h using a cabinet dryer (E3 Drying Cabinet, Genlab, UK). The dehydrated peels were ground into a fine powder using a Panasonic MX-AC300 Mixer Grinder, with particle sizes ranging from 75 to 135 µm. The powder was packaged in airtight HDPE zipper bags and stored in a cool, dry place for future use. The dried peels were then finely ground and preserved at -20°C to prevent moisture absorption and degradation of bioactive compounds.

2.4 Preparation of cakes

Cake processing followed the method of de Oliveira et al. (2025) with minor modifications. Wheat flour was partially substituted with guava fruit flour (3% and 5%) or guava leaf flour (3% and 5%). Preliminary trials were conducted to optimize the formulation. Whole eggs were mixed with sugar and oil until a light cream was formed. The respective flour substitutions, blended with baking powder and corn flour, were gradually incorporated into the egg mixture and mixed with a hand mixer (MK-H4-W, Panasonic Co., Malaysia) until homogeneous. The batter was poured into molds and baked at 180 °C for 30-35 minutes. Baked cakes were cooled at room temperature, packaged in aluminum foil bags, and stored under ambient conditions. **Table 1** contains the codes for respective samples

2.5 Proximate composition analysis

The proximate composition of the formulated cakes was determined according to AOAC standard methods (AOAC, 2023). The moisture, ash, crude protein, crude fiber, and crude fat contents were measured using the dry ash technique, oven drying method, Kjeldahl's method, gravimetric method, and soxhlet method, respectively.

2.6 Mineral content analysis

The concentrations of Ca, K and P in formulated cakes were quantified using biochemical analyzer (Humalyzer 3000), AAnalyst 400 and UV-1800, Shimadzu, Japan respectively. All analyses adhered to the AOAC (2023) standard methods.

Table 1. Codes of samples

Parameter	Pretreatment description
C	Control cake (no substitution)
GF-3	Cake substituted with 3% guava fruit powder
GF-5	Cake substituted with 5% guava fruit powder
GL-3	Cake substituted with 3% guava leaf powder
GL-5	Cake substituted with 5% guava leaf powder

2.7 Bioactive compounds analysis

2.7.1 Extract preparation

The extract was prepared by combining 1 g of material in a Falcon tube with 10 ml of 100% ethanol + acetone, allowing it to sit for 72 h, filtering the solvent, and then collecting the filtrates.

2.7.2 Estimation of total phenolic content (TPC)

The determination of Total Phenolic Content (TPC) in the sample extracts was determined following the method outlined by Azizi et al. (2010) with minor adjustments. Stock solutions (1 mg/mL) of extracts and gallic acid standards (1-8 mg/mL) were prepared. For each assay, 0.3 mL of sample or standard was mixed with 1.5 mL of diluted FC reagent, left for 3 minutes, then combined with 1.5 mL sodium carbonate solution (75 g/L). After 60 minutes of incubation, absorbance was recorded at 765 nm using a UV spectrophotometer, with ethanol as the blank. Total phenolic content (TPC) was expressed as mg gallic acid equivalents (GAE) per gram of extract.

2.7.3 Determination of antioxidant activity by DPPH Assay

The DPPH assay was employed to assess the antioxidant activity of the extracts, following the method outlined by Azlim Almey et al. (2010) with slight modifications. In this procedure, 6 mg of DPPH was dissolved in 100 mL of absolute methanol to create a methane-rich DPPH solution. Subsequently, 1 ml of the methane extract was mixed with 2 ml of the DPPH solution. The resulting mixture was gently shaken and left in the dark for 30 minutes at room temperature. Absorbance readings were taken at 517 nm using a UV-VIS spectrophotometer (UV-2600, Shimadzu Corporation, and USA). For the control, 1 mL of methanol was combined with 2 mL of the DPPH solution, and methanol served as the blank.

2.7.4 Estimation of total flavonoids content (TFC)

The determination of the Total Flavonoids Content (TFC) in the samples was conducted using the aluminum chloride colorimetric method as described by Chang et al. (2020) with minor adjustments. Total flavonoid content (TFC) was determined using a colorimetric assay with aluminum chloride ($AlCl_3$). First, a diluted extract was combined with ethanol and a series of reagents in a cuvette, then left to incubate for 30 minutes. The absorbance of the mixture was then measured at 415 nm with a spectrophotometer. The TFC was calculated by comparing the sample's absorbance to a quercetin standard curve, with results expressed as milligrams of quercetin equivalents (QE) per gram of extract.

2.7.5 Estimation of total anthocyanin content (TAC)

Samples TAC would be calculated calorimetrically using the aforementioned technique, with a few minor modification (Akhtar et al., 2014). A UV-VIS spectrophotometer was used to detect the color intensity at 520 nm after adding 3 mL of ethanoic extract to a cuvette (UV-2600, Shimadzu Corporation, USA). As a control, ethanol was used. TAC was calculated and expressed as mg per 100 g (mg/100 g).

2.8 In-vitro antidiabetic activity by alpha amylase inhibition assay

2.8.1 Extract preparation

To conduct the aqueous extraction, 2 grams of finely ground plant material were combined with 40 mL of distilled water. The blend was then introduced into a rotary shaker and allowed to agitate for 24 hours. Subsequently, the mixture underwent centrifugation at 8000 rpm for 10 minutes. The resultant supernatant was filtered through Whatman No. 1 filter paper. To maintain the integrity of the crude extracts, they were stored in a freezer at -20°C until analysis, with a maximum storage duration of 1 week (Kim et al., 2013).

2.8.2 Methodology

To measure the inhibitory effect of the aqueous extract on α-amylase activity, we used a modified method from (Kazeem et al., 2013). To assess α-amylase inhibition, sample extracts were prepared at concentrations of 10, 25, 50, and 100 mg/mL in 20 mM PBS buffer (pH 6.9). The reaction began by mixing 1 mL of the sample with 1 mL of 0.5 mg/mL α-amylase and incubating for 30 minutes. Following this, 1 mL of 1% soluble starch was added and allowed to react for 10 minutes before the reaction was stopped with 1 mL of DNS reagent. The mixture was then heated, producing an orange-red color, and its absorbance was measured at 540 nm using a spectrophotometer. Finally, α-amylase activity was calculated by comparing the results to a positive control (acarbose) and various background controls.

$$\textbf{Inhibition of } \boldsymbol{\alpha}\textbf{-amylase activity (\%)} = \frac{\text{Absorbance of control} - \text{Absorbance of sample}}{\text{Absorbance of control}} \text{ x } 100$$

Where, the control possesses 100 % enzyme activity and the tested sample was plant extract or the standard (acarbose).

2.9 Sensory analysis

Sensory evaluation was conducted with 25 semi-trained panelists (staff, teachers, and students, 20-45 years) from the Food and Nutrition Laboratory, Chattogram Veterinary and Animal Sciences University. Panelists had prior experience in food sensory analysis. Uniform portions from each cake batch were served in white coded dishes to avoid bias. Randomization was applied to sample presentation, and each panelist evaluated all formulations (Kortei et al., 2020). Informed consent was obtained from all participants prior to the study. Participation was voluntary, and panelists were assured of confidentiality regarding their responses.

2.9.2. Statistical analysis

The data were systematically arranged and coded using Microsoft Excel 2019. A one-way ANOVA was conducted using SPSS software (version 19.0), and Tukey's Honestly Significant Difference (HSD) test was employed to identify significant differences among the means at a 95% confidence level ($p \leq 0.05$).

3. RESULTS AND DISCUSSION

3.1 Yield determination

Despite having a slightly lower overall yield percentage, guava leaves were more efficient in terms of weight retention after drying compared to guava fruit. For instance, 253 grams of fresh guava leaves produced 98 grams of dried leaves, yielding 38.73%, while a much larger initial amount-3 kg of fresh guava fruit-only yielded 388 grams, for a comparable 38.8% yield. This suggests that using guava leaves may be a more efficient way to produce dried powder.

3.2 Nutritional composition of cakes

The cake's proximate composition values, expressed as mean percentages with standard deviations (ME±SD), can be found in **Table 2.** Fortifying cakes with guava fruit and leaf powders significantly enhanced their nutritional profile, with changes mirroring those observed in other fiber-enriched bakery products. The most dramatic shift was a sharp rise in crude fiber, particularly in leaf-fortified formulations, which aligns with the high fiber content of guava leaves and addresses a critical dietary shortfall (Jiménez-Escrig et al., 2001; Mahajan et al., 2024).

The additions also increased protein content from 4.72% to over 7%, driven by the inherent protein and amino acids in guava tissues (Mahajan et al., 2024). Ash content, an indicator of mineral richness, also rose, reflecting the presence of potassium, calcium, and magnesium in guava (Khan et al., 2025). This aligns with similar findings in other fruit peel-fortified products (Gadallah et al., 2022).

While moisture content increased, which can impact shelf-life, this is a common effect of hydrophilic plant fibers (Jiménez-Escrig et al., 2001). Conversely, soluble carbohydrates decreased from 58.27% to about 48%, an important outcome that suggests a potential reduction in glycemic load, offering a key health benefit for consumers (Lin, 2022). Guava leaf fortification resulted in higher protein, ash, and fiber contents compared to fruit-fortified and control cakes, reflecting the naturally higher mineral and fiber density of guava leaves. These findings underscore guava powders' potential as a multi-functional ingredient for developing healthier, nutrient-dense bakery items.

3.3 Energy content of cakes

Figure 1 demonstrates the energy content of formulated cakes.

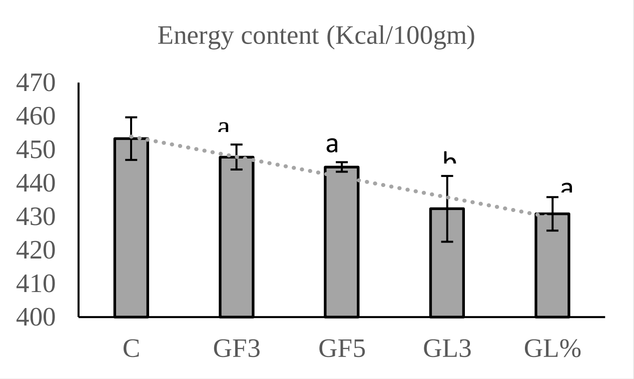

Fig. 1. Energy content of formulated cakes

The incorporation of guava fruit and leaf powders significantly reduced the energy content of cakes, with the control (C: 453.28 ± 6.36 kcal/100 g) showing the highest value and GL5 (430.81 ± 5.00 kcal/100 g) the lowest. A gradual decline was observed with increasing guava substitution, reflecting the replacement of energy-dense macronutrients with fiber, protein, and bioactive compounds abundant in guava derivatives (Kumar et al., 2021b; Pawar et al., 2024a). Guava fruit, with higher fiber and moisture but lower lipid content, also contributed to reduced metabolizable energy (Sahal et al., 2025). Similar reductions in energy density have been reported when cakes were enriched with date fruit powder Dana & Sonia (2024) or bakery products fortified with *Moringa oleifera* leaves (Govender & Siwela, 2020; Khaled et al., 2024). Although modest (≈20-25 kcal/100 g between C and GL5), this reduction may hold nutritional importance, as lowering the energy density of bakery products is a recognized strategy to curb calorie intake while increasing fiber and bioactive consumption (Alessandrini et al., 2019). Even at 3-5% substitution levels, guava fruit and leaf powders reduced caloric density without major formulation changes, with guava leaves exerting stronger effects due to compositional differences. Overall, guava derivatives demonstrated potential as functional ingredients to improve the nutritional quality of cakes.

3.4 Mineral composition of formulated cakes

Table 3 displays the mineral compositions of the cakes. Cake substituted with 5% GL exhibited higher quantities of Ca (75.00±1.00 mg/100 g), K (239.33±11.02 mg/100 g), and (196.67±5.77 mg/100 g). On the other hand, cake not substituted with GL AND GF contained lowest amounts of Ca (25.00±1.00 mg/100 g), K (38.66±0.58 mg/100 g), and P (150.00±10.00 mg/100g).

Table 2. Proximate composition of cakes

Sample ID	Moisture (%)	Crude fiber (%)	Ash (%)	Ether extract/fat (%)	Protein (%)	Soluble carbohydrate (%)
C	13.82±0.12^{d}	1.72±0.02^{d}	0.49±0.03^{b}	22.09±0.94^{c}	4.72±0.15^{b}	58.27±0.35^{a}
GF-3	16.80±0.08^{c}	2.59±0.12^{c}	0.79±0.03^{a}	24.17±0.24^{ab}	6.80±0.04^{a}	51.21±0.30^{b}
GF-5	18.31±0.05^{b}	3.64±0.13^{b}	0.81±0.03^{a}	24.95±0.15^{a}	7.13±0.12^{a}	47.94±0.89^{c}
GL-3	17.95±0.59^{b}	5.29±0.21^{a}	0.78±0.02^{a}	22.98±0.15^{c}	6.69±0.08^{a}	51.73±0.26^{b}
GL-5	19.54±0.31^{a}	5.51±0.18^{a}	0.80±0.03^{a}	24.34±0.54^{ab}	7.05±0.39^{a}	48.14±0.65^{c}

Values (means ± SD) of triplicate determination with different superscript letters are significantly different at $p \leq 0.05$. Where, C = Control cake; formulated without the addition of guava fruit/ leaf, GF-3 = cake prepared incorporating 3% guava fruit, GF-5 = cake prepared incorporating 5% guava fruit, GL-3 = cake prepared incorporating 3% guava leaves, GL-5 = cake prepared incorporating 5% guava leaves.

Table 3. Mineral composition of formulated cakes

Sample ID	Calcium (mg/100g)	Potassium (mg/100g)	Phosphorus (mg/100g)
C	25.00±1.00^{d}	38.66±0.58^{c}	150.00±10.00^{b}
GF-3	37.00±1.00^{c}	40.00±2.000^{c}	160.00±10.00^{b}
GF-5	39.33±0.58^{c}	39.67±2.080^{c}	166.67±15.28^{b}
GL-3	51.33±1.53^{b}	156.67±4.16^{b}	150.00±10.00^{b}
GL-5	75.00±1.00^{a}	239.33±11.02^{a}	196.67±5.77^{a}

Values (means ± SD) of triplicate determination with different sup different at $p \leq 0.05$. Where, C = Control cake; formulated without the addition of guava fruit/ leaf, GF-3 = cake prepared incorporating 3% guava fruit, GF-5 = cake prepared incorporating 5% guava fruit, GL-3 = cake prepared incorporating 3% guava leaves, GL-5 = cake prepared incorporating 5% guava leaves.

The calcium, potassium and phosphorus contents of the cakes were found to be significantly elevated by the incorporation of guava fruit and especially guava leaf powders. The control (C) exhibited the lowest values (Ca: 25.00 ± 1.00 mg; K: 38.66 ± 0.58 mg; P: 150.00 ± 10.00 mg per 100 g), while cakes with 5% guava leaf (GL-5) displayed the highest (Ca: 75.00 ± 1.00 mg; K: 239.33 ± 11.02 mg; P: 196.67 ± 5.77 mg per 100 g). The increase in mineral levels was proportionally greater with leaf inclusion than with fruit inclusion, and higher substitution levels (5%) led to more pronounced enhancement.

These results were consistent with previous analyses of *Psidium guajava* leaves, which were reported to possess high concentrations of calcium, potassium and phosphorus on a dry-weight basis (e.g. approx. 1660 mg Ca, 1602 mg K, and 360 mg P per 100 g DW) as compared to fruit (Kumar et al., 2021b; Pawar et al., 2024b). The mineral-rich nature of guava leaves was thus expected to drive up mineral content of the cakes when leaves were substituted. It was also observed in other studies that fortification of bakery products with leaf powders or vegetable powders led to marked increases in minerals. For example, cakes fortified with *Moringa oleifera* leaf powder showed significantly higher K and P contents compared to controls (Purkiewicz et al., 2024; Roni et al., 2021). Incorporating guava leaf powder at 3-5% significantly enriched the cake's mineral profile, especially with calcium and potassium. The greater increase in potassium in leaf-fortified cakes suggests that guava leaf tissue is a particularly rich source of this mineral. While all guava-enriched cakes showed mineral boosts, some less dramatic increases in phosphorus may have been limited by binding with phytates or losses during baking, which could also affect mineral bioavailability. This enhancement of key minerals holds significant nutritional value, particularly for populations with mineral deficiencies.

3.5 Bioactive compounds

The findings regarding bioactive compounds, including Antioxidant, TAC, TFC, and TPC, are detailed in **Table 4**.

The incorporation of guava fruit and leaf powders significantly enhanced the antioxidant and phytochemical profile of the cakes. The control sample showed the lowest antioxidant activity, flavonoid, phenolic, and tannin contents, whereas the highest values were observed in GL-5 (1.44 ± 0.002 mg TE/100 g antioxidant activity; 99.61 ± 0.05 mg QE/100 g TFC; 11.54 ± 0.02 mg GAE/100 mL TPC; 15.65 ± 0.56 mg TA/100 mL TAC). These results confirmed that the addition of guava derivatives, particularly leaves, markedly improved bioactive compound levels and antioxidant potential. The stronger effects of leaf powders compared to fruit powders were consistent with the compositional differences between the two. Guava leaves have been reported to contain exceptionally high concentrations of phenolic acids, flavonoids, and tannins, which contribute to their well-documented antioxidant and antimicrobial properties (Huynh et al., 2025; Kumar et al., 2021b). Fruit powders also enhanced bioactive content, but the relative increase was smaller, likely due to their lower phenolic density.

Similar trends have been reported in bakery fortification studies. Incorporation of *Moringa oleifera* leaf powder significantly increased total phenolic and flavonoid contents in cakes and breads, leading to higher antioxidant activity (Olusanya et al., 2020). The enrichment of muffins with grape pomace flour also enhanced phenolic content and radical scavenging activity, demonstrating the potential of plant-based powders to elevate functional properties of baked goods (Tseng & Zhao, 2013). Moreover, earlier work showed that polyphenolic stability during baking is partly retained, allowing final products to maintain enhanced antioxidant potential despite heat exposure (Abdel-Aal & Rabalski, 2022; Nicoli et al., 1999). The high tannin and anthocyanin contents in guava leaf-fortified cakes (GL-3 and GL-5) may also provide health benefits, since tannins are associated with antioxidant, anti-inflammatory, and cardioprotective properties (Chung et al., 1998). The higher tannin levels in leaf cakes compared to fruit cakes further support the contribution of secondary metabolites concentrated in leaves.

Leaf-fortified cakes exhibited markedly higher levels of phenolic-related compounds and antioxidant activity compared to fruit-fortified formulations. This enhancement may be attributed to the greater concentration and stability of phenolic compounds in guava leaves. However, the antioxidant response appeared to be influenced by the food matrix, suggesting interactions between phenolics and other cake components.

Table 4. Bioactives and antioxidant capacity of formulated cakes

Sample ID	Antioxidant activity (mg TE/100 g)	TFC (mg QE/100 g)	TPC (mg GAE/100mL)	TAC (mg TA/100 mL)
C	0.41±0.01^{d}	75.72±0.15^{e}	7.92±0.06^{e}	2.72±0.00^{e}
GF-3	1.14±0.003^{c}	102.75±0.05^{b}	14.33±0.05^{b}	4.63±0.27^{d}
GF-5	1.16±0.002^{c}	103.68±0.04^{a}	17.70±0.02^{a}	7.83±0.56^{c}
GL-3	1.37±0.002^{b}	95.28±0.07^{d}	9.53±0.06^{d}	14.16±0.32^{b}
GL-5	1.44±0.002^{a}	99.61±0.05^{c}	11.54±0.02^{c}	15.65±0.56^{a}

Values (means ± SD) of triplicate determination with different sup different at $p \leq 0.05$. Where, C = Control cake; formulated without the addition of guava fruit/ leaf, GF-3 = cake prepared incorporating 3% guava fruit, GF-5 = cake prepared incorporating 5% guava fruit, GL-3 = cake prepared incorporating 3% guava leaves, GL-5 = cake prepared incorporating 5% guava leaves.

3.6 Antidiabetic activity by α-amylase inhibition assay

Table 5 exhibit alpha-amylase inhibitory activity of cakes (3% GLP, 5%GLP) at the concentration of 100 μg/mL exhibited 61.23% and 62.671% of inhibition. 55.88% and 56.01% of inhibition in cake that were substituted with 3%, 5% GFP respectively. The cakes were investigated for their inhibitory effectiveness by determining their IC_{50} values. The positive control in this study, Acarbose, exhibited an IC50 value of 36.11μg/mL.

The α-amylase inhibition assay showed that guava fruit (GF) and especially guava leaf (GL) powder-fortified cakes exerted substantial inhibitory effects in a dose-dependent manner. At higher concentrations (100 μg/mL), GL-5 achieved ~62.67% inhibition, closely approaching the activity of acarbose (89.84%) albeit at higher concentrations. The IC_{50} values followed the order: GL-5 (30.05 μg/mL) < GL-3 (35.27) < GF-5 (53.12) < GF-3 (60.62) < Control (78.92 μg/mL), showing that leaf additions significantly enhanced α-amylase inhibitory potency.

These findings aligned with earlier work on *Psidium guajava* leaves, where strong α-amylase and α-glucosidase inhibitory effects were documented. For instance, Wang et al. (2010) found that guava leaf extracts inhibited α-amylase with IC_{50} values in a similar low µg/mL range. Guava leaf compounds such as quercetin, isoquercitrin and other flavonoids were shown to inhibit α-glucosidase (and to some extent α-amylase) with IC_{50} values not far off from acarbose, in comparable extract settings (Zheng et al., 2024).

Guava leaf powder proved to be a powerful inhibitor of α-amylase, with the GL-5 formulation (5% leaf powder) showing a significantly lower IC_{50} value. This suggests that the leaves contain a higher concentration of bioactive compounds, such as flavonoids, phenolics, and tannins - that effectively inhibit the enzyme responsible for starch digestion. While guava fruit powder also showed some activity, its compounds may have been less abundant or degraded during baking, leading to a weaker effect. From a nutritional perspective, this potent inhibitory activity means that cakes fortified with guava leaf powder could delay starch digestion and reduce post-meal glucose spikes. This offers a dietary approach to managing hyperglycemia that is similar to the effects of pharmaceutical α-amylase inhibitors. This finding aligns with other research on plant-based inhibitors from sources like cinnamon and mulberry, highlighting guava leaf powder's promise as a functional ingredient for managing blood sugar (Klomsakul & Chalopagorn, 2024; Rybak & Wojdyło, 2023). However, in vitro enzyme inhibition does not directly translate to efficacy in humans due to digestion, absorption, and metabolic breakdown. Also, the possible sensory changes (taste, texture) when using high levels of leaves might affect consumer acceptance.

The leaf-fortified cake (especially GL-5) was the most effective among aforementioned samples in inhibiting α-amylase, with IC_{50} approaching that of acarbose in function though not numerically matched. These results support using guava leaf powder as a functional ingredient to help modulate glycemic response from starchy foods.

Table 5. Antidiabetic activity of formulated cakes

	% inhibition					
Con. (µg/mL)	**Acarbose**	**C**	**GF-3**	**GF-5**	**GL-3**	**GL-5**
10	30.54	36.77	38.33	40.00	43.78	44.67
25	39.49	40.42	46.00	46.66	49.11	50.65
50	67.36	46.65	49.66	52.77	54.00	54.16
100	89.84	53.001	55.88	56.01	61.23	62.67
IC_{50} (µg/mL)	36.11	78.92	60.62	53.12	35.27	30.05

Values (means ± SD) of triplicate determination with different superscript letters are significantly different at $p \leq 0.05$. Where, C = Control cake; formulated without the addition of guava fruit/ leaf, GF-3 = cake prepared incorporating 3% guava fruit, GF-5 = cake prepared incorporating 5% guava fruit, GL-3 = cake prepared incorporating 3% guava leaves, GL-5 = cake prepared incorporating 5% guava leaves.

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3.7 Sensory properties of the formulated cakes

In Figure 2, the sensory evaluation showed a clear trend: as guava fruit (GF) and especially guava leaf (GL) powders were added, key sensory attributes-color, smell, appearance, texture, taste, and overall acceptability-declined somewhat relative to the control (C = 7 across most attributes). The GF-3 and GF-5 formulations maintained reasonably high scores (≈6.4-6.8) in many attributes, showing that modest fruit powder addition was tolerated well. However, the leaf-fortified cakes (GL-3, GL-5) incurred greater drops: taste dropped more sharply (GL-5 taste ≈4.5), and overall acceptability fell correspondingly (~5.5).

Color, smell, and appearance were less affected in GF than GL, implying that fruit powder induced less perceptible changes to visual or aromatic cues than leaf powder. Texture and taste were more sensitive: in GL-5, texture was rated lowest (≈5) and taste even lower (~4.5), suggesting that leaf inclusion at 5% altered mouthfeel and flavor in ways less preferred by panelists.

These patterns are consistent with findings from studies of leaf or vegetable powder fortification in bakery products. For instance, Roni et al. (2021) found that cakes fortified with *Moringa oleifera* leaf powder and banana flour saw acceptability drop as leaf concentration increased, largely due to darkening of color, and changes in texture and taste. In bread enriched with mango leaf powder, sensory panels reported lower color and flavor scores as leaf proportion increased, even though nutritional and antioxidant properties improved (Pirca-Palomino et al., 2024). Also, substitution with potent green leaf powders often led to bitterness, earthy or off-flavors, and denser crumb or coarser texture, which degrade acceptance at higher levels (Govender & Siwela, 2020).

Thus, in this study, the leaf powders (especially at 5%) appear to have crossed a threshold where flavor, aroma, and texture are noticeably compromised, whereas fruit powders up to 5% remained more acceptable. The overall acceptability score of 5.5 for GL-5 suggests that while still acceptable, the sensory penalty is non-trivial.

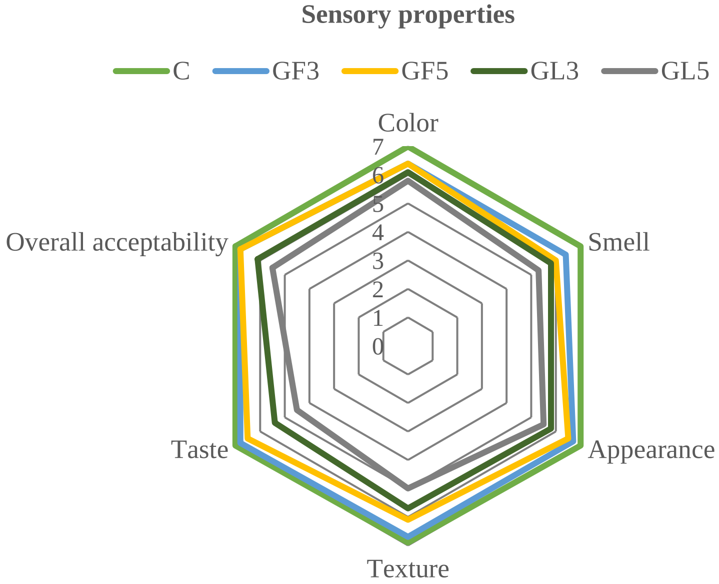

Fig. 2. Sensory properties of the formulated cakes. Where, C = Control cake; formulated without the addition of guava fruit/ leaf, GF-3 = cake prepared incorporating 3% guava fruit, GF-5 = cake prepared incorporating 5% guava fruit, GL-3 = cake prepared incorporating 3% guava leaves, GL-5 = cake prepared incorporating 5% guava leaves.

CONCLUSION

Guava fruit and leaf powders were successfully incorporated into cake formulations to enhance nutritional and functional properties. Guava fruit powder increased protein, fat, and ash content, while guava leaf powder significantly boosted dietary fiber and minerals, including calcium and potassium. Both additions reduced the cake's energy content. Bioactive profiling showed that guava fruit powder enriched cakes with phenolics, while guava leaf powder increased antioxidant activity, flavonoids, and tannins. Guava leaf cakes, particularly at a 5% concentration, showed potent α-amylase inhibition, comparable to acarbose, indicating significant anti-diabetic potential. From a sensory perspective, cakes with guava fruit powder (3-5%) were well-received, but those with guava leaf powder saw a decline in taste and aroma. This suggests that guava fruit powder is better for balancing nutrition and sensory quality, whereas guava leaf powder offers superior health benefits, particularly for glycemic control, despite its less favorable taste. Both ingredients hold promise as functional fortificants for bakery products, requiring a balance between nutritional gain and sensory acceptance.

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

DATA AVAILABILITY

The data used to support the findings of this study are available upon reasonable request from the corresponding author.

AUTHORS’ CONTRIBUTIONS

F.T.J.B. designed, conducted and analysed the study, A.C. analyzed and wrote the manuscript M.A. investigated and supervised the study.

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