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Nutritional Enhancement and Caloric Reduction in Cupcakes via Chia Mucilage and Stevia Substitution

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ABSTRACT

This study investigated the substitution of fat with Egyptian chia mucilage and the incorporation of stevia as a natural sugar alternative in the formulation of low-calorie cupcakes, herein referred to as natural sweetener chia cupcakes. Fat was partially or fully replaced with chia mucilage at levels of 25%, 50%, 75%, and 100%, while stevia was incorporated at a fixed level. The impact of these substitutions on functional, physical, chemical, textural, sensory, microbial, and economic parameters was assessed. Chia seeds demonstrated favorable nutritional properties, with high contents of lipids (25.00%), protein (20.56%), crude fiber (30.4%), and total carbohydrates (49.34%), in addition to strong water-holding and absorption capacities. Sensory evaluation indicated high panelist acceptance for all formulations, with scores exceeding 5 or 7 and no statistically significant difference compared to the control. Specific volume ranged from 2.52 to 2.77cm³/g, showing no significant reduction with increasing levels of chia mucilage. Protein content reached a maximum of 11.03% in the 100% substitution group, while fat content and caloric values decreased significantly, with energy values declining from 453.86 to 390.5kcal/100g. Microbial counts were undetectable immediately post-baking, remained low after three days, and showed no detectable yeast, mold, or *E. coli* after seven days. Full fat substitution resulted in a 7.23% reduction in production cost, highlighting the potential of chia mucilage and stevia to produce nutritious, cost-effective, low-calorie baked goods without compromising product quality or consumer acceptance.

1. Introduction

The growing global population, coupled with the increasing prevalence of diet-related health issues, has driven the food industry to prioritize the development of healthier food products. Cardiovascular diseases, diabetes, and obesity are among the most prevalent diet-associated conditions, often exacerbated by sedentary lifestyles. Epidemiological studies have shown that diets rich in healthy fats (e.g., omega-3 fatty acids) and dietary fiber, and low in cholesterol, saturated fats, and trans fats, can significantly enhance human health and overall vitality (Gui et al., 2017; Shahidi, 2009). Increased consumption of whole grains, seeds, fruits, and vegetables natural sources of prebiotics has been associated with reduced low-density lipoprotein (LDL) levels and a decreased risk of cardiovascular disease. Nutrient-dense alternatives, particularly plant-based options, are increasingly valued for their potential to improve diet quality, especially among vegetarians. Chia seeds (*Salvia hispanica L.*), for example, have been identified as effective fat replacers in various food applications (Liz & Nicole, 2015). Their composition includes high levels of omega-3 and omega-6 fatty acids, dietary fiber, minerals, phytochemicals, and calcium, making them a valuable nutritional component.

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Due to their chemical, nutritional, and functional properties, chia seeds may offer protective effects against obesity, osteoporosis, cardiovascular disease, and diabetes. *Salvia hispanica L.* is an annual herb from the Labiatae (mint) family, characterized by large green leaves and purple or white flowers that produce nutrient-rich seeds (Arctos, 2018; Mohammed et al., 2019). Chia seeds are widely used in the development of functional foods due to their favorable biomolecular profile (Rabail et al., 2021). The European Food Safety Authority has confirmed that chia seeds are safe for human consumption, with no mycotoxins or harmful levels of heavy metals (Gabal, 2024). In the food industry, chia seeds are used in multiple forms including whole, crushed, as flour, or oil—and are incorporated into products such as meat substitutes, yogurts, juices, cookies, cakes, pasta, bread, desserts, ice cream, and breakfast cereals (Das, 2017). Their hydrophilic nature allows them to absorb up to 12 times their weight in water, forming a mucilaginous gel that can substitute for fats and eggs in various recipes (Gabal, 2024). Furthermore, ethanolic extracts of chia have shown antibacterial, antifungal, and antimycobacterial activity against a range of pathogens (Güzel et al., 2020). Chia oil contains approximately 65% α-linolenic acid (omega-3) and 22% linoleic acid (omega-6), with minimal saturated fatty acids (Gui et al., 2017). Its nutrient profile includes 31–34% fat, 35% fiber, 37–45% carbohydrates, and 4–5% ash (Silva et al., 2017; Fortino et al., 2017). Depending on cultivation conditions, chia protein content can reach up to 27%, which is significantly higher than in traditional grains such as rice (6%) and wheat (9.5%). Baked goods such as bread, cakes, crackers, and cookies are widely consumed globally. In these products, chia mucilage has been shown to slow starch gelatinization, reduce starch hydrolysis by amylases, and consequently lower the glycemic index (Schuchardt et al., 2016; Iglesias-Puig & Haros, 2013). Cake, in particular, remains a popular bakery item due to its sensory appeal and cultural relevance. Traditional cake formulations often contain 15–20g of fat per 100g, with fats and emulsifiers contributing to softness, volume, and texture. Several studies have explored chia mucilage as a fat substitute in baked goods. Lorenza et al. (2020) evaluated the sensory properties of chocolate cakes formulated with chia gel and found them acceptable after frozen storage. Felisberto et al. (2015) reported that replacing up to 25% of fat with chia mucilage maintained the nutritional and sensory characteristics of cakes. Fernandes and de las Mercedes (2017) investigated different drying methods for chia mucilage and assessed its effect on the technological quality of baked products when used to replace fat at 25%, 50%, 75%, and 100%. Mucilage's unique functional properties including its emulsifying, thickening, and texturizing capabilities have increased its relevance in food applications. Additionally, chia mucilage offers bioavailable macro- and micronutrients and may modulate satiety and reduce glycemic response (Soukoulis et al., 2018). The aim of the present study was to develop low-fat, naturally sweetened cupcakes by fully or partially substituting conventional shortening with chia mucilage, and to reduce caloric content through the use of stevia as a sugar alternative. The study evaluated the physicochemical, nutritional, textural, and sensory characteristics of the formulated cupcakes. An additional goal was to contribute to the improvement of ketogenic dietary patterns by replacing animal-based fats with plant-derived oils, thereby reducing the potential adverse effects associated with traditional ketogenic regimens.

Figure 1. (A): Chia plant with large leaves and purple flowers (B): Egyptian Chia seeds, (Mohammed et al., 2019)

2. Materials and Methods

Materials

Chia seeds (*Salvia hispanica L.*), Egyptian variety 1 from the 2024/2025 season, were supplied by the Crop Intensification Department, Agricultural Research Centre, Giza, Egypt. Wheat flour (72% extraction), commercial-grade sucrose (sugar), salt, fresh whole eggs, baking powder, shortening, traditional skimmed milk powder (SMP) containing 0.5% fat, 35.5% protein, 51% lactose, 8.5% ash, and 5% moisture, and vanilla essence were all purchased from a local supermarket. The natural sweetener "Sweet & Slim" (stevia-based), produced by AWA Food Solution Company, was used as a sugar substitute.

Methods

Determination of Physical and Functional Properties of Chia Seeds

Physical Characteristics

The physical properties of chia seeds were determined, including the weight of 1000 seeds, seed length, width, and thickness. Bulk density, defined as the ratio of the mass of the seed sample to its total volume, was measured according to the method described by Bande et al. (2012). Apparent (or true) density was determined following the procedure of Garnayak et al. (2008).

Functional Properties

Water-Holding Capacity (WHC) and Oil-Holding Capacity (OHC)

Water-holding and oil-holding capacities were evaluated following the method of Chau, Cheung, and Wong (1997). In brief, 1g (dry basis) of the sample was mixed with 10mL of distilled water or corn oil (density 0.92g/mL), respectively. The mixtures were centrifuged at 2200 × g for 30 minutes using a Rolco Refrigerated Centrifuge (Model CR-5850, 22cm radius, Buenos Aires, Argentina). WHC was expressed as grams of water retained per gram of sample, and OHC was expressed as grams of oil retained per gram of sample. The WHC and OHC were calculated using the following formula:

$$WHC=W_2-W_1/W_0$$

Where, W_o: weight of the dry sample (g), W_1: weight of the centrifuge tube plus sample before centrifugation (g) W_2: weight of the centrifuge tube plus sediment after centrifugation (g)

Water Absorption Capacity (WAbC)

Water absorption capacity was determined according to the AACC method (1984). A 2g sample (dry basis) was weighed and gradually mixed with water until saturation (approximately 35 mL). The mixture was then centrifuged at 2000×g for 10 minutes (Rolco Model CR-5850, 22cm radius, Buenos Aires, Argentina). Excess water was removed, and the residue was weighed. WAbC was calculated by dividing the weight gain by the volume of water added, and the results were expressed as grams of water absorbed per gram of sample.

Organic Molecule Absorption Capacity (OMAC)

Organic molecule absorption capacity (OMAC) was determined according to the method of Zambrano, Meléndez, and Gallardo (2001). A 3g sample was immersed in an excess of corn oil (approximately 10 mL) and kept at 25 °C for 24 hours. The mixture was then centrifuged at 2000×g at 25 °C for 15 minutes using a Beckman GS-15R centrifuge. OMAC was calculated based on the weight gain of the sample and expressed as grams of oil absorbed per gram of sample (g oil/g sample).

Chia Mucilage (CM) Extraction

Chia mucilage was extracted following the method of Boreno et al. (2010). Chia seeds (*Salvia hispanica L.*) were manually cleaned to remove impurities. The seeds were soaked in tap water at a ratio of 1:9 (seed:water, g/mL), stirred thoroughly, and left to rest for 30 minutes to allow the formation of a 10% chia gel (mucilage). The resulting mucilage was then used directly in the cupcake formulations.

Stereomicroscope Imaging of Chia Mucilage Formation

To observe chia mucilage formation during hydration, the method of Salgado-Cruz et al. (2013) was followed. A randomly selected chia seed was placed on a Petri dish and covered with a coverslip. Two drops of water were added to fill the space between the seed and the coverslip, ensuring contact with water. A sequence of micrographs was captured using a stereomicroscope equipped with a digital camera to document the mucilage formation over time.

Preparation of Cupcake Samples

Cupcakes were prepared according to the method described by Khalifa et al. (2015), with modifications to incorporate chia mucilage and stevia. The control cupcake was formulated using traditional shortening and commercial-grade sucrose. For the experimental formulations, shortening was replaced with chia mucilage at levels of 25%, 50%, 75%, and 100%. In these samples, stevia ("Sweet & Slim") was used as a sugar substitute. Based on the product's specifications, 1g of stevia is equivalent in sweetness to 10g of sugar; thus, 12.5g of stevia was used in place of 125g of sugar. The cupcake formulation (Table 1) involved mixing sugar and salt, followed by the addition of melted shortening (or chia mucilage in the substituted samples). Eggs were whipped with vanilla until a smooth, creamy texture was achieved. Separately, wheat flour was combined with baking powder and skim milk powder, then gradually incorporated into the egg mixture. The batter was blended thoroughly using a Moulinex 750 blender (France) to ensure homogeneity. The prepared batter was poured into baking cups and baked in a preheated electrical oven (Luxell – LX3570, Turkey) at 180 ± 10°C for 15 ± 2 minutes.

Table 1. Ingredients of cupcake samples

Ingredients (g)	Cake					
	Control 1	Control 2	CM-25%	CM-50%	CM_75%	CM-100%
Wheat flour	250	250	250	250	250	250
Sugar	125	—	—	—	—	—
*Stevia		12 .5	12 .5	12 .5	12 .5	12 .5
Fresh egg	110	110	110	110	110	110
Salt	3,5	3,5	3.5	3.5	3.5	3.5
Baking powder	12.5	12.5	12.5	12.5	12.5	12.5
Vanilla	2.0	2.0	2.0	2.0	2.0	2.0
Skim milk powder	25	25	25	25	25	25
Shortening	53	53	39.625	26.5	13.375	—
Chia mucilage	—	—	13.375	26.5	39.625	53

*Every 1 gram of stevia is equivalent to10 grams of sugar (mentioned with the product)
Control 1(00% Shortening), CM-25% (75% Shortening and 25% CM), CM-50% (50% Shortening-50% CM.), CM-75% (25% shortening -75% CM), CM100% (100% CM.)

Sensory Evaluation of low –calorie Cupcake samples

The cupcake samples were subjected to sensory evaluation two hours after baking. A total of 20 trained panelists from the staff of the Crops Technology Research Department, Food Technology Research Institute, Agricultural Research Center (Giza, Egypt) participated in the assessment. Each panelist was asked to rate the sensory quality attributes visual appearance, crust color, crumb color, texture, taste, odor, and overall acceptability using a 9-point hedonic scale, where 1 indicated "dislike extremely" and 9 indicated "like extremely," following the method of Pyler (1973). Samples, with and without chia seed mucilage (CSM), were presented in random order to prevent bias.

Texture Profile Analysis (TPA)

Texture profile analysis of the low-calorie cupcakes was performed using a TA-XT Plus texture analyzer (Stable Micro Systems Ltd., Surrey, UK). A cylindrical crumb sample ($2cm^3$) taken from the center of each cake was analyzed using a cylindrical probe with a diameter of 3.6 cm. The test parameters were set as follows: pre-test speed of 2.0mm/s, test speed of 1.0mm/s, post-test speed of 2.0mm/s, with 50% compression and a 5 g triggering force. A two-cycle compression test was carried out to simulate the action of chewing.The texture attributes measured included: Hardness (N), Springiness (mm), Cohesiveness (dimensionless ratio), Chewiness (N·mm). Data were analyzed using Texture Expert® software version 1.05. The method followed the guidelines established by the AACC (2000).

Determination of Cupcake Quality

Physical Characteristics

The physical properties of the cupcake samples were assessed by measuring their specific volume (cm^3/g). Specific volume was calculated as the ratio of the cupcake's volume (cm^3) to its weight (g), using the formula:

Specific Volume (cm^3/g)=Weight (g)/Volume (cm^3)

The average weight of each cupcake was measured using a precision electronic balance (Model CY204, USA). The volume was determined via the rapeseed displacement method. Specific volume values were calculated according to AACC Method 10-05.01 (AACC, 2010).

Color Measurement

Color analysis of the cupcake crust and crumb was performed using a portable Chroma Meter (Minolta, Model CR-200, Tokyo, Japan). The instrument provided L*, a*, and b* color values:

L* indicates lightness, with values ranging from 0 (black) to 100 (white)

a* represents the red-green axis, where positive values indicate redness and negative values indicate greenness

b* corresponds to the yellow-blue axis, with positive values indicating yellowness and negative values indicating blueness.

Measurements were taken at three different points on both the crust and crumb of each sample, and average values were reported.

Chemical composition of Chia and Cupcakes samples

Chemical Analysis of Cake

Moisture, protein, ash, crude fat and crude fiber content were determined according to the method described in AOAC (2012).

Available carbohydrate content was estimated by difference as mentioned by Fraser and Holmes (1959).

Available carbohydrate % (on dry basic)=100 – (protein %+fat %+ash% +fiber);

Total calories was determined according the following equation :

Total calories (kcal/100g)= 4 (% protein+ % carbohydrate)+ 9 (% fat), FAO/WHO (1991).

Determination of Total Phenolic Content and Radical Scavenging Activity of Chia Seeds

Preparation of Methanolic Extract

One gram of chia seeds was soaked in 10 mL of 80% methanol and subjected to overnight shaking at 100 rpm at ambient temperature (25–30°C) using an incubator shaker (New Brunswick, USA). The resulting mixture was filtered through Whatman No. 1 filter paper, and the filtrate was designated as the methanolic extract.

Total Phenolic Content (TPC)

Total phenolic content was measured using the Folin–Ciocalteu (FC) reagent method, as described by Singleton and Rossi (1965), with slight modifications. Briefly, 0.5 mL of the methanolic extract was mixed with 0.5 mL of FC reagent (diluted 1:1 with distilled water) and allowed to stand for 5 minutes at room temperature. Subsequently, 1 mL of 10% sodium carbonate (Na_2CO_3) solution was added. The mixture was incubated for 10 minutes at room temperature, and the absorbance was recorded at 730nm using a UV-Vis spectrophotometer. All determinations were performed in triplicate. Gallic acid monohydrate was used as a calibration standard, and the results were expressed as grams of gallic acid equivalents (GAE) per 100 grams of extract.

DPPH Radical Scavenging Activity

The antioxidant activity of the chia seed extract was assessed using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay, according to the method of Llorach et al. (2008), with slight modifications. A 0.08mM DPPH solution in methanol was freshly prepared. A 50µL aliquot of the methanolic extract was mixed with 950µL of DPPH solution and incubated in the dark for 5 minutes. The absorbance of the mixture was measured at 515nm using a UV-Vis spectrophotometer (Cary 50 Scan, Varian). Antioxidant activity (AA) was calculated as the percentage inhibition of DPPH radical using the following equation: $AA(\%)=100-[100\times(A_{control}/A_{sample})]$

Where, A_{sample} is the absorbance of the sample and $A_{control}$ is the absorbance of the DPPH solution without the extract.

Fatty Acid Profile

Crude fat was extracted from chia seeds and converted to fatty acid methyl esters (FAME) via esterification, following the procedure outlined by Spiric et al. (2010). The FAMEs were then analyzed using gas chromatography for profiling individual fatty acids.

Microbial Quality

Microbial analysis was conducted to determine the total bacterial count, yeast, mold, and coliform groups, following the standard microbiological procedures described by the American Public Health Association (APHA, 1992).

Statistical Analysis

All experiments were conducted in triplicate, and the results were expressed as mean ± standard deviation (SD). The data were subjected to one-way analysis of variance (ANOVA) to evaluate the significance of the effect of chia seed concentration on the various quality parameters of the cupcake samples. Differences among means were considered statistically significant at $p \leq 0.05$. When significant differences were found, Duncan's Multiple Range (DMR) test was employed to separate means using PASW® Statistics 18 (formerly SPSS).

3. Results and Discussion

Stereomicroscope images of chia mucilage formation during hydration time .

Stereomicroscopic analysis was performed to examine the hydration behavior and mucilage release from chia seeds (chia nutlets, CN). Micrographs were taken at different time intervals (5, 10, 20, and 30 minutes) using a stereomicroscope equipped with a digital camera. The results demonstrated a gradual formation of a transparent mucilage layer surrounding the seeds as hydration progressed. By 30 minutes, a well-defined mucilage envelope had formed, with an approximate thickness of 1 mm, which was consistent with observations from previous studies (Salgado-Cruz et al., 2013; García-Salcedo et al., 2018). The mucilage appeared firmly adhered to the seed surface, making it difficult to separate visually, confirming that water contact initiates mucilage release from the pericarp. This finding aligns with prior research indicating that mucilage excretion in *Salvia* species occurs via diffusion through the outer seed coat upon hydration (Hedge, 1970). The extent of mucilage release is highly dependent on several factors, including hydration time, temperature, pH, and the water-to-seed ratio. Extraction techniques such as soaking, stirring, or cooking can significantly influence the yield and physicochemical properties of the mucilage (Orifici et al., 2018). Some researchers have employed Response Surface Methodology (RSM) to optimize these parameters for enhanced mucilage yield and functionality. The stereomicroscopic observations in this study support the notion that chia mucilage is a highly functional hydrocolloid with potential applications in food systems requiring thickening, gelling, or moisture-retention capabilities

Figure 2. The stereomicroscope images of hydrated CM

(A) Chia seed before adding water (B) Chia after hydration (45 min)

Physical and functional properties % of *Chia (Salvia Hispanic L.)* seeds (on dry weight).

Data in Table 2 reveal that the physical characteristics of chia seeds (*Salvia hispanica L.*) included a 1000-seed weight of 1.53g, an average seed length of 1.95mm, 1.29mm a width, 0.79mm a thickness , and a bulk density of 0.78g/cm, respectively. The establishment of such seed biometric parameters is essential for the design and development of harvesting, sowing, processing, and storage equipment. The findings regarding the seed dimensions are generally in agreement with those reported by Mohammed et al. (2019), who found that the 1000-seed weight, length, width, thickness, and bulk density ranged from 1.1855 to 1.2043g, 1.93 to 1.82 mm, 1.19 to 1.29mm, 0.80 to 0.82mm, and 0.598 to 0.718g/cm respectively, for local and imported chia seeds. Similarly, Pizarro et al. (2014) reported that chia seeds are typically small, oval with dimensions ranging from 2.0–2.5mm in length, 1.2–1.5mm in width, and 0.8–1.0mm in thickness, with a 1000-seed mass of 1.11–1.227g and bulk density between 0.70 and 0.72g/cm^3.

Table 2. Physical and functional properties % of *Chia (Salvia Hispanic L.)* seeds (on dry weight)

Physical properties of Chia seeds	
Seed	
1000 seed weight (g)	1.53± 0.02
L (mm) of one seed	1.95±0.66
W(mm) of one seed	1.29 ± 0.09
Thickness (mm) of one seed	0.79 ±0.01
Volume (g/ml) of one seed	1.96 ±0.05
Bulk denticity (g/ml) of one seed	0.78±0.23
Color values	
*L**	54.65±0.45
*a**	3.73±0.12
*b**	6.24±0.08
Functional properties of chia seeds	
WHC (g/g)	10.72±0.09
WA_bC (g/g)	5.88±0.67
OHC (g/g)	2.03±0.08
OAC (g/g)	1.58±0.31

Water-Holding (WHC), Oil-Holding Capacity (OHC), Water Absorption Capacity (WA_bC), Organic Molecule Absorption Capacity (OMAC)

Color parameters of Chia seed

The color measurements presented in Table 2 showed that the Egyptian chia seeds exhibited values of 54.65 for lightness (*L**), 3.73 for redness (*a**), and 6.24 for yellowness (*b**), characterizing the seeds as deep gray or beige with dark specks. These values are relatively higher in lightness compared to those reported by Suleiman et al. (2015), who found that chia seeds typically ranged from 41.3 to 42.0 for *L**, 3.19 to 3.61 for *a**, and 7.43 to 8.61 for *b**. Variations may be attributed to differences in moisture content (10–20%), geographic origin, and post-harvest handling.

Water-holding capacity (WHC) of chia seeds

The water-holding capacity (WHC) and water absorption capacity (WAbC) of the Egyptian chia seeds were found to be 10.72g/g and 5.88 g/g, respectively. This indicates a strong ability of the seeds to retain water, likely due to the high dietary fiber content and the fibrous outer layer of the seed coat, which allows physical entrapment of water. These findings are in line with Segura-Campos et al. (2014), who recorded high WHC values (775–800g/100g) for whole chia seeds. The organic molecule absorption capacity (OMAC) and oil absorption capacity (OAC) were recorded as 2.03g/g and 1.58g/g, respectively. These values suggest a moderate potential for chia seeds to bind hydrophobic substances, such as oils and organic compounds. The mechanisms for such oil retention are largely due to physical entrapment within the seed matrix and interactions between fat and the polar side chains of proteins (Omran & Hussein, 2015). Overall, the high WHC and OAC of Egyptian chia seeds can be attributed to the presence of hydroxyl groups in fiber components, such as cellulose, which form hydrogen bonds with water molecules. These properties enhance the seeds functionality as natural thickeners or fat replacers in baked products (Capitani et al., 2012), making them suitable for use in low-calorie formulations such as cupcakes.

Table 3. Chemical composition % of Chia (*Salvia Hispanic L.*) seeds (On dry weight)

Moisture and dry matter content % of Chia	
Moisture content %	7.2 ± 0.06
Dry matter %	92.8 ±0.45
Chemical composition % and Caloric value of chia seeds (on dry weight %)	
*Protein %	20.56 ±0.02
Fat %	25.00 ±0.70
Ash %	5.20 ±0.10
Crude Fiber %	30.4 ±0.98
** Total carbohydrates %	18.84 ±0.09
Caloric Value (Kcal /100g)	382.6 ±0.08
Total phenols & Antioxidant activity of Egyptian chia (mg/g) on dry weight	
Total phenolics as GAE (mg/g)	(1.30)
***Antioxidant activity (%)	(1.78)

* N=6.25. **By difference
*** GAE gallic acid equivalent

Proximate Composition and Caloric Value of Egyptian Chia Seeds

The proximate composition data in Table 3 show that the moisture content of Egyptian chia seeds was 7.2%. This relatively low moisture level is beneficial for seed stability during storage, as it inhibits microbial growth and reduces the rate of biochemical degradation. Consequently, the dry matter content reached 92.8%, which forms the basis for evaluating other nutritional parameters. On a dry weight basis, the seeds contained 25.00% lipids, 20.56% protein, 5.2% ash, 30.4% crude fiber, and 18.84% total carbohydrates. These findings confirm that Egyptian chia seeds, particularly the “Egyptian 1” variety, are a rich source of oil and dietary fiber and can be classified among emerging oilseed crops. These results are consistent with the compositional ranges reported by Fernandes & Salas-Mellado (2017), who found chia seeds typically contain 30–33% oil, 15–25% protein, 26–41% carbohydrates (including 18–30% dietary fiber), 4–5% ash, and 90–93% dry matter. The high caloric value of chia seeds can be attributed primarily to their lipid content. As reported by Michele and Myriam (2014), chia lipids are a valuable source of energy, essential fatty acids, and fat-soluble vitamins (A, D, E, and K), which enhance the nutritional profile of the seeds and support their use in functional foods.

Phenolic Content and Antioxidant Activity of Egyptian Chia Seeds

The total phenolic content of Egyptian chia seeds was found to be 1.30mg GAE/g, indicating a moderate but significant presence of bioactive polyphenolic compounds. These compounds contribute substantially to the seed’s oxidative stability and health benefits by preventing lipid peroxidation and scavenging free radicals. The antioxidant activity, measured as DPPH radical scavenging capacity, was 1.78mg/g, which is consistent with previously reported values. Gomez-Favela et al. (2017) recorded phenolic contents around 1.41 mg GAE/g DW, while Osama et al. (2019) reported a broader range (0.757 to 1.908mg GAE/g), reflecting variability due to seed origin, extraction method, and growing conditions. Furthermore, Reyes-Caudillo et al. (2008) found antioxidant activity values of 1.68mg/g for imported chia and 1.85mg/g for Egyptian chia seeds. The antioxidant potency of Egyptian chia seeds may be attributed particularly to their content of caffeic acid, a phenolic acid with strong radical-scavenging capacity, as well as other flavonoid compounds. This highlights the potential of Egyptian chia seeds as a natural antioxidant source for application in food systems or functional ingredients. These results confirm the nutritional richness and functional potential of Egyptian chia seeds, aligning well with international standards and literature. The high content of fiber, oil, protein, and antioxidants makes them excellent candidates for functional food development, particularly in low-calorie baked goods and health-oriented formulations.

Fatty Acids Composition of Egyptian Chia Seeds

The fatty acid composition of Egyptian chia seeds is presented in Table 4. The results revealed that saturated fatty acids (SFAs) were primarily composed of palmitic acid (C16:0) and stearic acid (C18:0), which were present at 10.28% and 6.12%, respectively. In contrast, the lowest levels of SFAs were observed for behenic acid (C22:0) and arachidic acid (C20:0), measured at 0.20% and 0.46%, respectively. Among the monounsaturated fatty acids (MUFAs), oleic acid (C18:1) was the predominant component, accounting for 12.63% of the total fatty acids. This is consistent with previous reports indicating that oleic acid is typically the major MUFA in chia seeds. Regarding polyunsaturated fatty acids (PUFAs), chia seeds demonstrated a remarkable content. Alpha-linolenic acid (ALA, C18:3, ω-3) was the most abundant PUFA at 30.47%, followed by stearidonic acid (C18:4, ω-3) at 9.17%. These findings confirm chia seed's well-known status as one of the richest plant sources of omega-3 fatty acids. The n-6 to n-3 PUFA ratio (ω6:ω3) was calculated at approximately 1:1.9, and the saturated to unsaturated fatty acid ratio was 1:4.28, indicating a favorable lipid profile for cardiovascular health. Maintaining a low dietary ω6:ω3 ratio is associated with a reduced risk of chronic diseases, particularly coronary heart disease. These results are in agreement with earlier studies. Miranda-Ramos et al. (2020) reported linoleic acid contents ranging from 12.7 to 14.3g/100g of chia oil, and Nitrayová et al. (2014) confirmed the presence of palmitic, stearic, oleic, linoleic, α-linolenic, and arachidic acids in chia seeds, with linoleic acid reaching 18.89% in some cases. All studies consistently highlight the low ω6:ω3 ratio, positioning chia as a nutritionally valuable functional ingredient. In summary, the high levels of α-linolenic acid (ALA) and low ω6:ω3 ratio underline the potential of chia seeds in promoting heart health and maintaining a balanced serum lipid profile, making them a valuable addition to health-conscious diets.

Table 4. Identification of fatty acids composition of chia seeds

Fatty acids	Concentrations %	Fatty acids	Concentrations %
Palmitic acid C16:0	10.28	Oliec acid C18:1	12.63
Stearic acid C18:0	6.12	Linoleic acid C18:2 ω6	20.76
Arachidic acid C20:0	0.46	Linolenic acid C18:3ω3	30.47
Benhenic acid C22:0	0.20	Strodonic acid 18:4 ω3	9.17
Total saturated fatty acids	17.06	Polyunsaturated fatty acids	73.04
Saturated to unsaturated FA ratio	1:4.28	ω6:ω3 FA ratio	1:1.9

Table 5. Physical characteristics of different low calorie Cupcake samples

Samples	Volume (cm^3)	Weight (g)	Specific Volume (cm^3/g)
C1	115.00 ^{a}± 0.11	43.00^c ±0.22	2.6 7^a ± 0.03
C2	115.50^a± 0.51	44.50^a ± 0.50	2.59^a ± 0.08
CMS 25%	115.00^a ± 0.50	44.50^a ± 0.50	2.58^a ± 0.14
CMS 50%	115.00 ^{a} ± 0.17	44.00^b ±0.20	2.58 ^{a} ± 0.17
CMS 75%	115.00^a± 0.50	45.00^a ± 0.80	2.55^a ± 0.40
CMS 100%	112.50^a ± 0.25	44.50^a ± 0.50	2.52^a ± 0.15

C1 Control (100 % Shortening and sugar), C2 Control (100 % Shortening & stevia), CMS-25% (25% CM and 75% shortening), CMS-50% (50% shortening -50%CM.), CMS-75% (25% Shortenning-75% CM), CMS-100% (100% CM.)
a, b, c: any two means with the same letters in the same row mean that the results are not significantly different at ($P < 0.05$). Each

Physical Characteristics of Low-Calorie Cupcakes

Table 5 presents the effect of fat substitution with chia mucilage (CM) on the physical properties of low -calorie cupcake samples. The results indicate that the incorporation of chia mucilage did not result in statistically significant differences (p>0.05) in key physical parameters weight, volume, and specific volumewhen compared to the control samples prepared without chia mucilage. These physical attributes are essential indicators of product quality, as they significantly influence consumer acceptance. Volume and specific volume are particularly influenced by the quantity and functionality of fat in baked goods. Fat plays a crucial role in aeration and in stabilizing air cells formed during mixing, which affects the final texture and appearance of the product. The volume of the cupcakes across all treatments ranged from 112.5 to 115.0cm³, showing no significant variations between the control and chia mucilage-substituted samples. Similarly, the specific volume values were between 2.52 and 2.67cm³/g. Although a slight decrease in specific volume was observed with increasing levels of chia mucilage, the reduction was not statistically significant. This slight change may be attributed to the water-binding properties of chia mucilage, which can affect the aeration process during batter preparation, as previously reported by Felisberto et al. (2015).

In conclusion, these findings suggest that chia mucilage can be used as a fat replacer in low-calorie cupcakes without negatively impacting key physical properties, supporting its application in the development of healthier baked products.

Table 6. Effect of replacing Chia Muslage Seed (CM S) on Sensory evaluation of different low calorie cupcake samples

Samples	Crust color (9)	Crumb color (9)	Appearance (9)	Odor (9)	Texture (9)	Taste (9)	Overall Acceptability (9)
C1	8.7ᵃ±0.48	8.5ᵃ±0.85	8.35ᵃ±0.81	8.2ᵃ±0.23	8.6ᵃ ±0.52	8.6 ᵃ±0.53	8.9 ᵃ± 0.32
C2	8.6ᵃ±0.52	8.55ᵃ±0.50	8.00ᵃ±1.6	8.35ᵃ±0.75	8.55ᵃ±0.75	8.45ᵃ±0.64	8.72 ᵃ ± 0.96
CMS-25 %	8.5ᵃ±0.55	8.1ᵃ ±0.87	8.1ᵃ± 0.57	8.05ᵃ±0.28	7.8ᵃᵇ±0.48	8.12ᵃᵇ±0.85	8. 65ᵃ ± 0.78
CMS-50%	8.5ᵃ±0.55	8.1ᵃ ±0.87	8.15ᵃ±0.58	8.1ᵃ ±0.52	7.8ᵃᵇ±0.48	8.1ᵃᵇ±0.62	8.5 ᵃ ±0.72
CMS -75%	8.6ᵃ±0.46	8.1ᵃ ±0.87	8.1 2ᵃ±0.52	8.05ᵃ±0.76	7.9ᵃᵇ±0.52	8.02ᵇ±0.49	8.5 ᵃᵇ ±0.65
CMS-100%	8.4ᵃ ±0.52	8.1ᵃ± 0.87	8.17ᵃ±0.77	7.95ᵇ±0.58	0.75ᵇ±0.79	8.05ᵇ±0.49	8.25 ᵇ ±1.01

C1 Control (100% Shortening and sugar), C2 Control (100% Shortening & stevia), CMS-25% (25% CM and 75% shortening), CMS-50% (50% shortening -50%CM.), CMS-75% (25% Shortenning-75% CM), CMS-100% (100% CM.)
Hedonic a scale ranging from 1 to 9, 1= (dislike extremely) to ,9= (liked extremely) . Hedonic a scale ranging from 5 to 9=Desirable to purchase. n=20. a, b, c:any two means with the same letters in the same row mean that the results are not significantly different at ($P < 0.05$). Each means ± SD (n = 3).

Sensory Evaluation of Low-Calorie Cupcake Samples

The sensory evaluation results of the low-calorie cupcake samples are presented in Table 6 and Figure 3. No statistically significant differences ($p \leq 0.05$) were observed between the control sample and those prepared with up to 100% fat replacement using Chia Seed Mucilage (CSM 25%, CSM 50%, CSM 75%, and CSM100%) across all evaluated sensory attributes. The sensory attributes included crust and crumb color, visual appearance, texture, odor, taste, and overall acceptability. All cupcake samples, including those with full fat substitution, received overall acceptability scores ranging from 8.3 to 8.5 on the 9-point hedonic scale. These high scores reflect strong consumer acceptance and suggest a high likelihood of purchase by potential consumers. Furthermore, the data confirm that a complete fat substitution with chia mucilage did not adversely affect any of the sensory attributes. Panelists rated all treatments as acceptable, even with full replacement of traditional fat with chia mucilage. This outcome demonstrates the feasibility of developing low-calorie, health-oriented cupcakes without compromising sensory quality. The findings are in agreement with previous research by Ahmet and Özge Yeşildemir (2019), who highlighted the functional benefits of chia seeds in food applications, particularly in bakery products, due to their gel-forming and foam-enhancing capabilities. Additionally, Naumova et al. (2017) reported the high sensory acceptability of chia-based products, supporting the current findings. In conclusion, replacing shortening with chia seed mucilage and sugar with stevia can yield low-fat, low-calorie cupcakes with excellent sensory properties, making them a viable product for health-conscious consumers and the functional food market.

Color Parameters of Low-Calorie Cupcake Samples

Table 7 presents the color attributes of various low-calorie cupcake samples formulated with different levels of Chia Seed Mucilage (CSM). Color is a critical quality attribute in bakery products, as it significantly influences consumer perception and purchasing decisions.

Crust Color

Color measurements were conducted using the CIE L*a*b* color space, where $L*$ indicates lightness, $a*$ represents the red-green axis, and $b*$ the yellow-blue axis. A significant reduction in lightness ($L*$) of the cupcake crust was observed as the CSM level increased from 25% to 100% ($p < 0.05$). This darkening effect is likely due to natural pigments such as carotenoids and chlorophyll present in the chia seed coat (De Falco et al., 2017). Additionally, the increase in protein content with higher CSM levels may have enhanced the Maillard reaction, contributing to the browning of the crust.

All cupcake samples exhibited positive *a** values, indicating a reddish hue. However, no statistically significant differences in redness were detected among the CSM treatments and the control, except for the CSM 100% sample, which showed a notable decrease in redness (12.66). This reduction in red hue can be attributed to the total replacement of vegetable shortening with chia mucilage, which contains darker pigments. The *b** values, representing yellowness, varied significantly among treatments. Control samples showed higher *b** values, indicating a more yellow appearance. As the CSM concentration increased and shortening decreased, yellowness declined, likely due to the loss of pigment contribution from shortening and the increasing influence of chia's darker pigments.

Crumb Color

Regarding the crumb color, control samples exhibited the highest *L** values, signifying a lighter interior compared to samples containing CSM. As the level of CSM increased, crumb color became progressively darker, with CSM100% samples showing the lowest *L** values. This could again be linked to carotenoids and other naturally dark compounds present in chia.

Crumb a* values in CSM-containing samples shifted toward the green spectrum (negative *a** values), contrasting with the reddish tones of the control samples. This shift may reflect the greenish pigments inherent in chia seeds. A corresponding decline in crumb yellowness (*b**) was observed as CSM levels increased, while the control group maintained higher *b** values due to the presence of vegetable shortening. These findings align with Coelho and Salas-Mellado (2015), who observed similar color behavior in functional bread made with chia flour lighter crusts and darker crumbs. As reported by Agrahar-Murugkar et al. (2018), the crust color in baked products results primarily from Maillard reactions and sugar caramelization, whereas crumb color reflects the intrinsic hues of the formulation's ingredients.

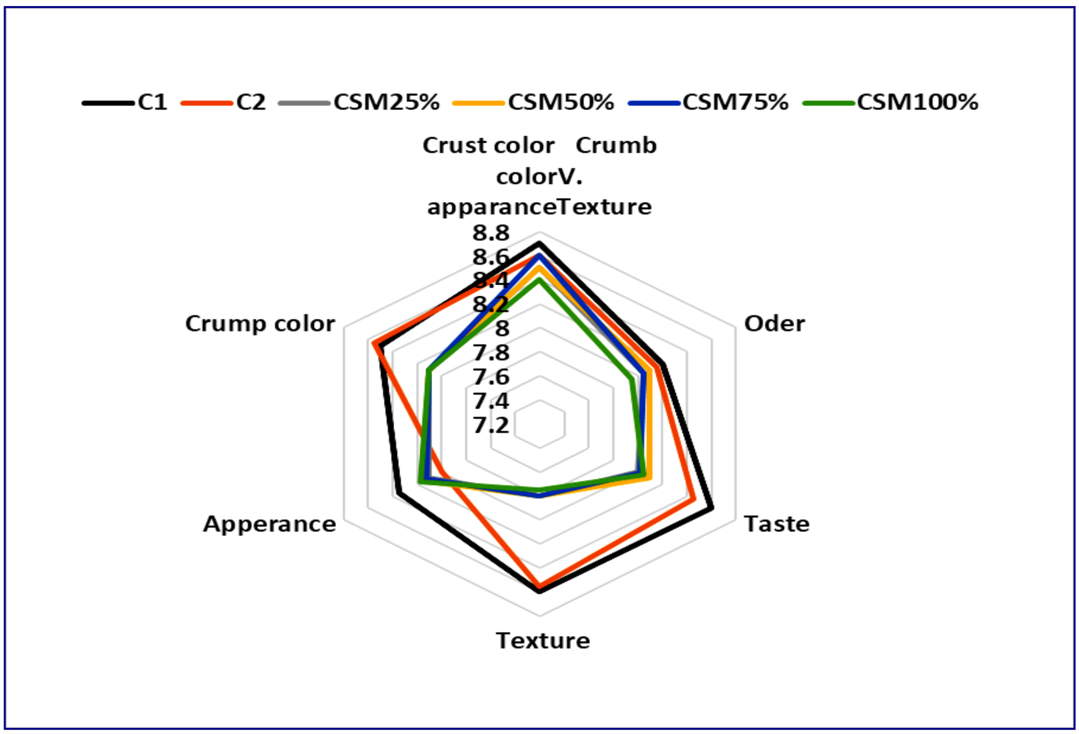

Figure 3. Sensory evaluation of low calorie cupcake samples

C1 Control (100% Shortening and sugar), C2 Control (100% Shortening & stevia), CMS-25% (25% CM and 75% shortening), CMS-50% (50% shortening -50%CM.), CMS-75% (25% Shortenning-75% CM), CMS-100% (100% CM.)

Table 7. Effect of replacing chia seed mucilage (CS-M) on color values of different low calorie Cupcake samples

Cake samples	Crust			Crumb		
	*L**	*a**	*b**	*L**	*a**	*b**
C1	63.59^{ab}±1.1	14.34^{a}± 0.08	$26.45^{\pm b}$1.66	87.63^{a}±1.45	1.02^{a}±0.95	22.21^{a}±0.2
C2	66.92^{a}± 2.5	13.75^{ab}±0.55	$36.29^{a\pm}$1.54	85.63 ^{a}± 0.7	1.59^{a}±0.072	23.3^{a}±1.77
CMS 25%	62.06^{bc}±1.28	13.98^{ab}±0.94	25.18^{b}±2.18	83.39^{b}±0.57	-3.31^{b}±0.073	21.36^{b}± 0.02
CMS 50%	58.66^{cd}±0.76	13.88^{ab}±0.71	18.9^{c}±1.00	82.46^{b}±1.35	-3.69^{b}±0.13	20.34^{b}±1.02
CMS 75%	58.21^{cd} ± 4.6	13.8^{ab} ±0.25	18.59^{c}±0.48	79.67^{c}±0.82	-3.99^{bc}±0.03	21.87 ^{b}±1.14
CMS 100%	56.94^{d} ±3.04	12.66^{b} ±1.36	17.87^{c}±0.12	72.07^{d}± 2.4	-4.24^{c}±0.13	17.54^{c}±0.50

C1 Control (100% Shortening and sugar), C2 Control (100% Shortening & stevia), CMS-25% (25% CM and 75% shortening), CMS-50% (50% shortening -50%CM.), CMS-75% (25% Shortenning-75% CM), CMS-100% (100% CM.)
a, b, c: any two means with the same letters in the same row mean that the results are not significantly different at (P < 0.05). Each means ± SD (n = 3).

Table 8. Impact of fat and stevia substitute on chemical composition (g/100g, on dry weight) and caloric value of different low calorie Cupcakes

Sample	Protein %	Lipid %	Ash%	Fiber %	Available Carbohydrate %	Caloric Value (Kcal /100g)	Fat source %/ Total calories %
C1	7.88^{f}±0.05	13.50^{b}±0.29	2.11^{c}±0.10	1.30^{cd}±0.10	75.21^{a}±0.05	453.86^{b}±0.12	26.77^{b}±0.78
C2	9.42^{e} ±0.11	15.62^{a}±1.11	2.17^{c}±0.10	1.50^{c} ±0.21	71.29 ^{b}±0.70	463.42^{a} ±0.34	30.33^{a}±0.66
CMS-25%	10.07^{d} ±0.05	9.26^{c} ±0.36	3.4^{bc}±0.53	2.50^{b}±0.33	74.77 ^{a}±0.29	422.70^{c}± 0.66	21.9^{c} ±0.78
CMS-50%	10.42^{c} ±0.04	8.10^{cd}±0.44	3.65^{b}±0.05	2.75^{b}±0.45	75.13^{a} ±0.25	415.10^{d}±0.47	17.56^{d}±0.44
CMS-75%	10.70^{b} ±0.06	6.06^{d}±0.20	4.03^{a}±0.15	3.30^{a} ±0.37	75.91^{a} ±0.78	400.98^{e}±0.55	13.60^{e} ±0.87
CMS-100%	11.03^{a}±0.01	4.70^{e} ±0.05	4.8^{a}±0.20	3.45^{a}±0.36	76.02^{a}±0.91	390.5 ^{f}± 0.09	10.83^{f}±0.45

C1 Control (100% Shortening and sugar), C2 Control (100% Shortening & stevia), CMS-25% (25% CM and 75% shortening), CMS-50% (50% shortening -50%CM.), CMS-75% (25% Shortenning-75% CM), CMS-100% (100% CM.)
a, b, c: any two means with the same letters in the same row mean that the results are not significantly different at (P < 0.05). Each means ± SD (n = 3)

Proximate Composition of Low-Calorie Cupcake Samples

Table 8 presents the proximate composition of control cupcakes and samples containing varying levels (25%, 50%, 75%, and 100%) of chia seed mucilage (CSM) as a fat replacer. The results show that increasing levels of CSM substitution led to a significant increase in protein, ash, and crude fiber content, accompanied by a notable decrease in fat content and caloric value. The protein content increased progressively with the level of chia mucilage. The highest protein value was observed in the 100% CSM sample (11.03%), followed by CMS 75% (10.70%), CMS 50% (10.42%), and CMS 25% (10.07%). In contrast, the control samples (C1 and C2) showed lower protein contents at 7.88% and 9.42%, respectively. This enhancement in protein content is attributed to the nutritional richness of chia seeds and is consistent with findings by Borneo et al. (2010), who reported increased protein content in cakes formulated with chia mucilage. The fat content decreased significantly with higher levels of chia mucilage substitution. The CMS100% sample had the lowest fat content at 4.75%, compared to CMS 75% (6.06%), CMS 50% (8.10%), and CMS 25% (9.26%). This trend reflects the successful replacement of shortening with mucilage, consistent with the findings of Borneo et al. (2010), who reported a 57% fat reduction in cakes with added chia mucilage. The ash and fiber contents also showed significant increases. Ash content rose from 2.11% in control samples to 4.8% in the CMS100% sample, while crude fiber content increased from 1.30% to 3.45% across the same range. This reflects the mineral and dietary fiber contribution of chia seeds, enhancing the nutritional profile of the cupcake formulations. In contrast, carbohydrate content showed a slight but significant increase, rising from 71.29% in control samples to 76.02% in the 100% CSM sample. This may be due to a relative increase in other macronutrients as fat content decreases.

Importantly, the caloric value of the cupcakes decreased significantly with higher levels of chia mucilage substitution. The CMS100% cupcake exhibited the lowest caloric value (390.5kcal/100g), followed by CMS75% (400.98kcal/100g), CMS50% (415.10kcal/100g), and CMS25% (422.70kcal/100g). Control samples had the highest values at 453.86 and 463.42kcal/100g. This reduction is primarily due to the lower fat content, given that fats contribute 9kcal/g more than twice the energy of proteins or carbohydrates. These results support the use of chia mucilage as an effective fat replacer in cake formulations, offering improved nutritional quality and lower caloric density, aligning with the findings of Felisberto et al. (2015), who highlighted the technological and nutritional benefits of chia mucilage in reduced fat bakery products.

Texture Profile Analysis of Different Cupcakes

Table 9 presents the effect of replacing fat with chia seed mucilage (CSM) on the texture profile of different cupcake samples. Texture measurements included hardness, cohesiveness, springiness, and chewiness, which are important indicators of cake quality and are directly related to the perceived softness of the cake. Firmness is defined as the force required to compress a food to a specific length at a specific rate. Based on the textural property results (Table 9), the effect of chia mucilage added at different concentrations as a fat replacer on the firmness of stale cupcake crumbs is clearly demonstrated. The hardness of all treated samples increased significantly during storage, which can be attributed to the staling process, dehydration, and starch retrogradation reactions.

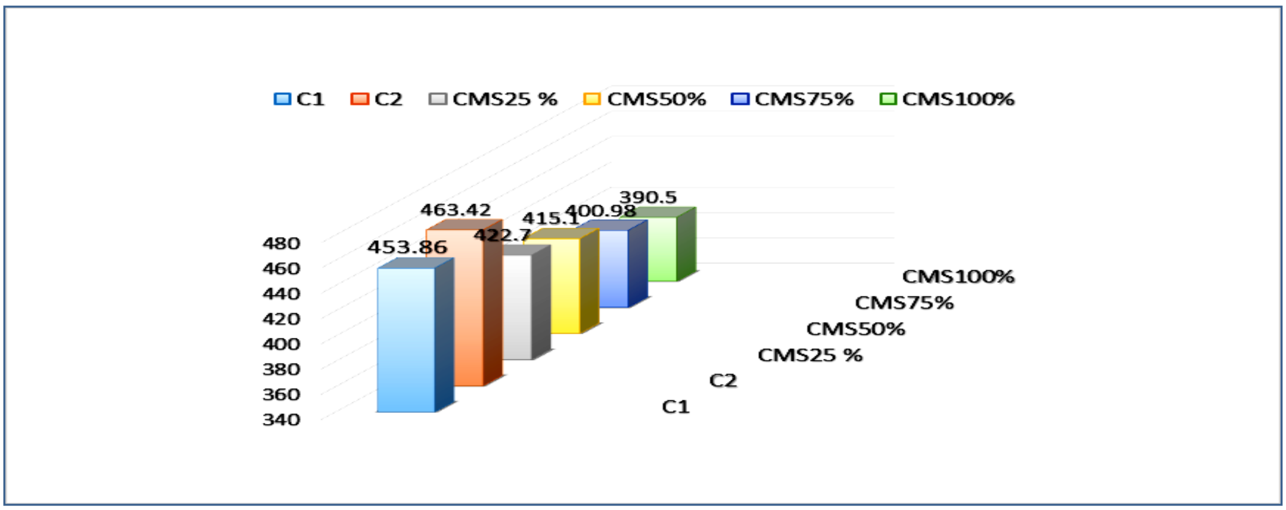

Figure 4. Caloric values (Kcal /100g) in different cupcake recipes

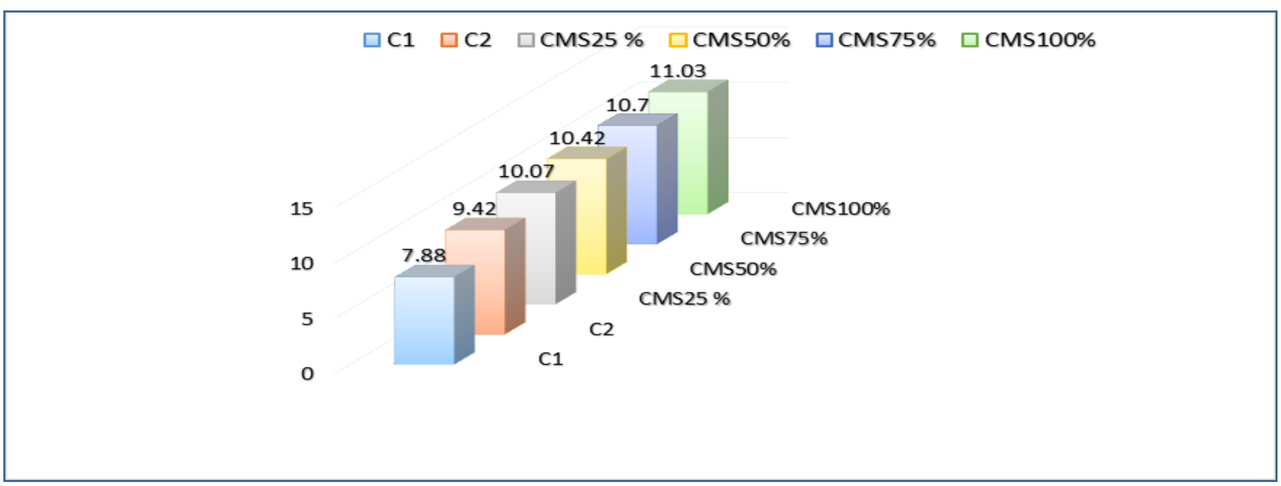

Figure 5. Protein % content in different cupcake recipes

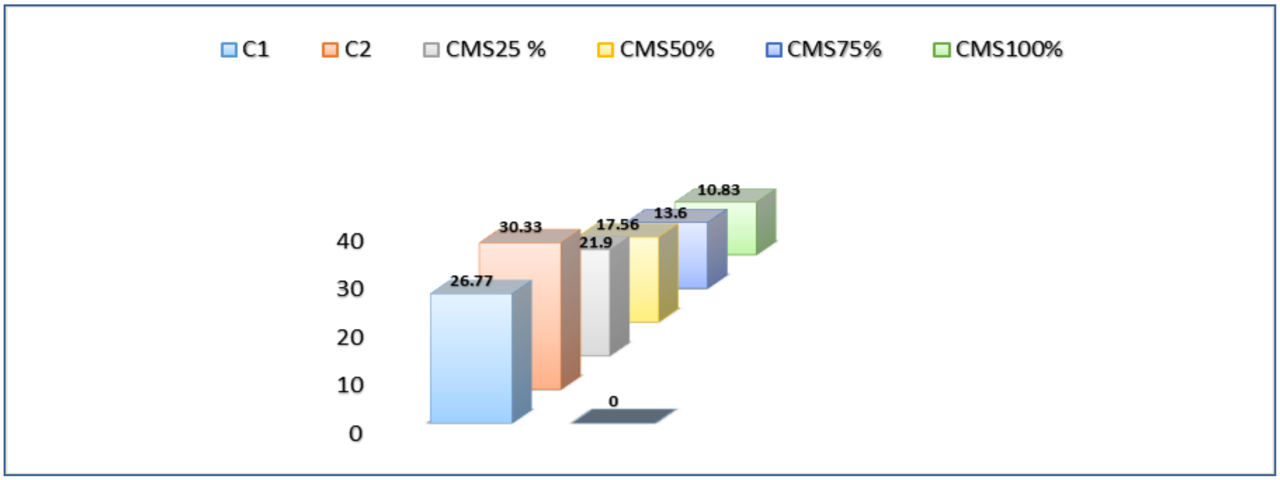

Figure 6. Fat source %/Total Calories

Table 9. Texture Profile Analysis of low calorie Cupcake samples during storage time (7 days) at room temperature (25±2˚C)

Samples	Hardness (N)	Chewiness (N)	Cohesiveness (N)	Springiness (mm)
At Zero Time				
C1	6.40 ^{e} ±0.34	0.051 ^{a} ± 0.01	0.52 ^{d} ± 0.01	8.60^{c} ± 0.20
C2	7.65 ^{cde} ± 0.25	0.06 ^{a} ± 0.01	0.51^{c} ±0.01	9.81^{b} ± 0.01
CMS25 %	7.88^{cde} ± 0.90	0.08 ^{a} ± 0.04	0.54^{c} ± 0.02	10.07 ^{ab}±c 0.26
CMS50%	8.85^{cd} ± 0.35	0.08 ^{a} ± 0.25	0.55 ^{c} ± 0.20	10.18^{ab} ± 0.18
CMS75%	9.60 ^{c} ± 2.60	0.11 ^{a} ± 0.23	0.56 ^{c} ± 0.03	10.27 ^{ab} ± 0.27
CMS100%	10.38^{bc} ± 0.26	0.12 ^{a} ± 0.22	0.63^{ab} ± 0.05	10.37 ^{ab} ±0.02
At the end of storage time 7 days				
C1	8.4^{cde}±0.37	0.05 ^{a}± 0.07	0.61^{ab} ± 0.05	10.2 ^{ab} ± 0.02
C2	18.17 ^{b} ±1.07	0.07 ^{a} ± 0.02	0.64^{ab} ±0.03	10.27 ^{ab} ± 0.12
CMS-25 %	19.5 ^{ab}±0.13	0.13^{a} ±0.05	0.66 ^{ab}± 0.05	10.38 ^{ab} ±0.13
CMS-50%	19.6 ^{ab} ±0.95	0.14 ^{a} 0.06	0.68 ^{ab}± 0.03	10.40 ^{ab} ±0.24
CMS-75%	20.5 ^{a} ± 0.90	0.15^{a} ± 0.03	0.70^{a} ± 0.02	10.46 ^{ab} ±0.25
CMS-100%	21.60 ^{a} ±1.15	0.18 ^{a}± 0.05	0.71 ^{a}± 0.06	10.49 ^{ab}± 0.23

C1 Control (100% Shortening and sugar), C2 Control (100% Shortening & stevia), CMS-25% (25% CM and 75% shortening), CMS-50% (50% shortening -50%CM.), CMS-75% (25% Shortenning-75% CM), CMS-100% (100% CM.)
a, b, c: any two means with the same letters in the same row mean that the results are not significantly different at (P < 0.05). Each

Texture Profile Analysis of Low-Calorie Cupcake Samples During Storage Time (7 Days) at Room Temperature (25 ± 2°C)

Several studies have examined the evolution of cake crumb texture during storage, particularly focusing on increased firmness due to the staling process, which is largely attributed to starch retrogradation (Paraskevopoulou et al., 2015). As shown in Table 9, the hardness of all cupcake samples increased significantly over the 7 day storage period. This increase was more pronounced in samples containing higher levels of chia seed mucilage (CSM), with the highest hardness observed in the CM100% sample (100% fat replacement). In contrast, the control samples (C1 and C2, containing no CSM) recorded significantly lower hardness values both at day 0 and at the end of the storage period. This firmness in high-CSM samples may be explained by several factors: chia mucilage's strong water retention capacity, its high protein and fiber content, and its interference with starch gel formation due to the presence of bran-like particles. These particles may disrupt the uniform structure of the cake crumb, leading to increased firmness and reduced specific volume. This negative correlation between hardness and specific volume aligns with observations in Table 5. Furthermore, the lack of a gluten network in these formulations might contribute to the increased crumb density and decreased gas retention, as reported by Aljobair (2022), who also found that chia seed flour addition increased cake hardness and reduced volume in a dose-dependent manner.

Similarly, Coelho and Salas-Mellado (2015) observed increased firmness and reduced loaf volume in wheat breads supplemented with 2–20% chia flour. Additionally, the combined reduction of sugar and fat content in cake formulations is generally associated with increased hardness (Milner et al., 2020). Chewiness, a secondary texture parameter, did not show statistically significant differences among the cupcake samples and control groups. The chewiness values ranged from 0.05 to 0.18, with minor variations across treatments, suggesting that CSM addition had minimal impact on this attribute. Cohesiveness reflects the internal bonding within the cake crumb and is influenced by batter structure. Cakes containing chia mucilage exhibited higher cohesiveness compared to control samples, likely due to the denser and more hydrated batter produced by CSM, which may have facilitated the formation of a more cohesive structure upon baking (Rosenthal and Thompson, 2021). At day 0, cohesiveness values ranged from 0.55 to 0.63 for CSM-containing samples, compared to 0.51 and 0.52 for the control samples (C1 and C2). By day 7, these values increased further, with CM25%, CM50%, CM75%, and CM100% samples recording 0.66, 0.68, 0.70, and 0.71, respectively, compared to 0.61 and 0.64 in control samples. These results suggest that storage time enhanced cohesiveness more prominently in the presence of chia mucilage. Springiness, which measures the ability of the cake to return to its original shape after compression, was also positively influenced by chia mucilage addition. The CM100% sample exhibited the highest springiness, potentially due to the formation of thicker air cell walls in the cake crumb, facilitated by the high water-binding and gel-forming properties of chia mucilage. At day 0, cupcakes with chia mucilage showed significantly higher springiness compared to controls. However, by the end of the storage period, the differences between low-calorie CSM cupcakes and control samples were not statistically significant. These findings align with those of Sung et al. (2020), who reported similar springiness behavior in rice flour cakes made with chia seed flour.
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Microbial quality (cfu/g) of different Chia cupcakes during storage time (7 days) at room temperature (25±2˚C)

Table 10 presents the effect of chia on the microbial quality of all cupcake formulations over a 7-day storage period at room temperature (25±2°C). The total bacterial count, *E. coli*, and yeast and mold levels were measured using the second dilution for total bacteria, yeast, mold, and coliform groups. At zero time, microbial counts for all tested organisms were undetectable, likely due to the baking temperature of 180 °C. However, low total bacterial counts were recorded for the different formulations: 20, 17, 30, 25, 20, and 9 cfu/g for C1, C2, CM25%, CM50%, CM75%, and CM100%, respectively. After 7 days of storage, there was a significant increase in total bacterial counts, with the same formulations reaching 60, 55, 32, 27, 21, and 12 cfu/g, respectively. Notably, *E. coli* and yeast/mold were not detected in any of the samples throughout the storage period. This absence may be attributed to the antimicrobial properties of chia seeds. In general, as the chia concentration increased, the total microbial count decreased. These findings align with previous studies demonstrating that chia seed protein fractions possess antibacterial activity against both Gram-positive and Gram-negative bacteria (Coelho et al., 2018). According to Güzel et al. (2020), ethanolic extracts of chia seeds exhibited antibacterial, antifungal, and antimycobacterial properties against various pathogens.

Moreover, Abdel-Aty et al. (2021) reported that methanolic extracts from germinated Egyptian chia seeds showed enhanced antibacterial activity compared to extracts from dried seeds. Germination increased the polyphenol content and improved antibacterial action against pathogenic bacteria such as *Salmonella typhi*, *Escherichia coli*, *Staphylococcus aureus*, and *Pseudomonas aeruginosa*. The antibacterial mechanism involved the generation of hydrogen peroxide (H_2O_2), which damaged bacterial proteins, leading to cell death.

Table 10. Microbial quality (cfu/g) of different Chia cupcakes during storage time (7 days) at room temperature (25±2˚C)

Samples	T.C 10^2 cfu/g	Y& M 10^2 cfu/g	Coliform group 10^2
At zero time			
C1	ND	ND	ND
C2	ND	ND	ND
CMS25 %	ND	ND	ND
CMS50%	ND	ND	ND
CMS75%	ND	ND	ND
CMS100%	ND	ND	ND
After 3 days storage time			
C1	20	ND	ND
C2	17	ND	ND
CMS25 %	30	ND	ND
CMS50%	25	ND	ND
CMS75%	20	ND	ND
CMS100%	9	ND	ND
After 7 days storage time			
C1	60	ND	ND
C2	55	ND	ND
CMS-25 %	32	ND	ND
CMS-50%	27	ND	ND
CMS-75%	21	ND	ND
CMS-100%	12	ND	ND

C1 Control (100% Shortening and sugar), C2 Control (100% Shortening & stevia), CMS-25% (25% CM and 75% shortening), CMS-50% (50% shortening -50%CM.), CMS-75% (25% Shortenning-75% CM), CMS-100% (100% CM.), ND .Not detected

Table 11. Total cost (L.E) & cost reduction (%) of low calorie Cupcake samples compered to control samples

Ingredients (g) & the cost (LE)	C1	C2	CMS-25%	CMS-50%	CMS-75%	CMS-100%
Wheat flour (1000g)	20.00	20.00	20.00	20.00	20.00	20.00
Eggs (8 eggs)	40.00	40.00	40.00	40.00	40.00	40.00
Sugar (500)	17.5	----	----	---	---	---
Stevia (50g)	----	20.00	20.00	20.00	20.00	20.00
Vanilla essence (8g)	4.00	4.00	4.00	4.00	4.00	4.00
Baking powder (50g)	10.00	10.00	10.00	10.00	10.00	10.00
Shorting (gm)	212.00	212.00	159.00	106.00	53.00	---
Price of Shortening (100g)	21.20	21.20	15.90	15.90	5.30	---
Chia seeds (g)	---	---	5.30	10.60	15.90	21.20
Price of Chia seeds	---	---	1.00	2.00	3.00	4.00
Skim milk (100g)	20.00	20.00	20.00	20.00	20.00	20.00
Cost/Kg of low caloric Cupcake	132.70	135.20	130.90	126.60	122.30	118.00
20% services (L.E)	26.54	27.04	26.18	25.32	24.46	23.40
Total cost/Kg low caloric cupcakes	159.24	162.24	157.08	151.92	146.76	141.6
N. of cupcakes	50.00	48.00	48.00	48.00	48.00	48.00
Cost of one cupcake (43-45g)	3.18	3.38	3.27	3.165	3.05	2.95
Cost reduction (%) of one cupcake	----	6.25	2.83	-0.63	-4.08	-7.23

C1 Control (100% Shortening and sugar), C2 Control (100% Shortening & stevia), CMS-25% (25% CM and 75% shortening), CMS-50% (50% shortening-50%CM.), CMS-75% (25% Shortenning-75% CM), CMS-100% (100% CM.)

Economic Analysis: Total Cost (L.E) and Cost Reduction (%) of Low-Calorie Cupcake Samples Compared to Control Samples

Table 11. presents the economic evaluation of low-calorie cupcakes sweetened with stevia and formulated with varying proportions of chia mucilage (CM) as a fat substitute. These are compared with two control samples: Control 1 (C1), which contains traditional shortening and sugar, and Control 2 (C2), which is sugar-free. The primary objective of this analysis was to develop a low-calorie cupcake by replacing both fat and sugar using chia mucilage and stevia, respectively. Among the samples, Control 2 had the highest cost at £3.38 per cupcake (weighing 43–45 grams), followed by Control 1 at £3.18. Among the treatments containing chia mucilage, the 25% CM substitution showed a slight cost increase compared to Control 1. However, from 50% up to 100% substitution, the cost progressively decreased below that of Control 1.

The cost reduction ranged from 0.64% to 7.23%, depending on the level of chia mucilage substitution. Notably, complete replacement of fat with chia mucilage (CM100%) resulted in the highest cost reduction of 7.23% compared to the control group. These findings suggest that chia mucilage not only contributes functional benefits but also reduces production costs. Therefore, developing sugar-free, low-fat cupcakes using chia mucilage and stevia can be a cost-effective approach for creating functional foods, particularly suitable for health-conscious consumers, including individuals with obesity or diabetes.

4. Conclusion

This study demonstrated that replacing fat with Egyptian chia mucilage and sugar with stevia in low-calorie cupcakes is an effective strategy for creating a healthier, functional product. Chia mucilage was used at substitution levels of 25%, 50%, 75%, and 100%, while stevia served as a constant, natural, calorie-free sweetener. The cupcakes were evaluated across various parameters, revealing that chia seeds possess favorable physicochemical properties such as high water-holding and oil absorption capacities. Sensory analysis showed high overall acceptability, particularly for the 25% substitution, with no significant differences from the control. Nutritionally, full substitution increased protein, fiber, ash, and carbohydrate content while reducing caloric value to 390.5 kcal/100g. Microbial assessments confirmed the safety of all samples during storage, with no detection of yeast, mold, or *E. coli*, likely due to chia's antimicrobial properties. Economically, chia mucilage reduced production costs, with a full replacement leading to a 7.23% cost reduction. Overall, chia mucilage and stevia present a promising combination for producing cost-effective, low-calorie cupcakes with enhanced nutritional value, microbial safety, and consumer appeal—ideal for individuals with obesity, diabetes, or other dietary restrictions populations.

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