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Effect of Roselle Calyces Concentrate with Other Ingredients on the Physiochemical and Sensory Properties of Cupcakes

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Abstract

Purpose: Roselle calyces are a major crop for export and used to make a common drink in Egypt. The objective of this research was to determine the physiochemical and the sensory properties of cupcakes formulated with roselle calyces concentrate incubated with 11 different food grade ingredients (FGI) prior to addition to the cupcake batters.

Methodology: Anthocyanins, fibre, moisture, colour and sensory evaluations were done along with batter and baking quality.

Findings: Roselle calyces cupcakes incubated with molasses and orange zest had the highest sensory scores (P<0.05). The parameter a* was significantly redder when roselle calyx concentrates were incubated with vinegar, lemon or orange juice. One hundred g of roselle cupcakes with lemon juice provided 420 mg/100 g anthocyanins and 10% of total dietary fibre.

Practical implications: The FGI is available and inexpensive. Roselle calyces cupcake with the FGI can be made at home and is less sour than the roselle calyces drink. These cupcakes would have a "clean" label.

Originality: This is one of the first studies to use FGI to treat roselle calyces concentrate. The FGI are sources of acids such as juices or vinegar, natural sweeteners such as honey and molasses that enhance the stability of anthocyanins and which may themselves have numerous phytochemicals.

Keywords: Roselle calyces; *Hibiscus sabdariffa* L.; Anthocyanins; Cupcakes; Fibre; Cost effective product; Needs assessment

Introduction

Roselle calyces (*Hibiscus sabdariffa* L.) is a tropical plant in the *Malvaceae* family and is known in Egypt as *Karkadah*. It is probably a native of West Africa and is now widely cultivated throughout the tropics and subtropics, e.g., Sudan, China, Thailand, Egypt, Mexico, and the West India [1]. Roselle calyces are one of the major Egyptian crops and are used in food, drinks, and cosmetics.

Roselle plant is cultivated mainly in Upper Egypt [2]. There are two types of roselle calyces: light and dark red colours. The dark red roselle calyx, (*H. sabdariffa* var. *sabdariffa*) is shorter and bushier, and planted for its edible calyces. The light red roselle (*H. sabdariffa* var. *altissimoe wester*) is an erect, sparsely branched annual growing to 4.8 m high, which is cultivated for its jute-like fibre.

Roselle calyx anthocyanins might be used as a natural food colourant [3], safer than most synthetic dyes that contain azo functional groups and aromatic rings, they may have negative effects on health including allergic and asthmatic reactions [4], DNA damage [5], and hyperactivity [6]. Some synthetic dyes are even considered to be potentially carcinogenic and mutagenic to humans [7]. However, using roselle calyces into food products would provide a "clean" label and would also add additional phytochemicals that might be beneficial to health while protecting the roselle calyces anthocyanins.

The pigments roselle calyces anthocyanin are inherently unstable, but when acylated becoming more stable. Stability is being improved in solutions and in baking [8,9], or by encapsulation [10] using relatively pure citric acid, acetic acid, citrus pectin, tartaric acid, and adipinic acid.

The colour stability of anthocyanins depends on a combination of factors including: structures of the anthocyanins, pH, temperature, oxygen, light, and water activity [11]. Enzymatic degradation and interactions with other food components such as ascorbic acid, sugars, metal ions, sulfur dioxide, co-pigments and food matrices are also important [9,11]. The bioactive compounds in roselle calyces, such as some flavonoids and anthocyanins, are unstable and may be degraded during beverage preparation to colourless or brown-coloured products, which represent a loss of their beneficial health properties [12]. Cupcakes are a convenient bakery product for consumers to buy or make at home. Therefore, the current study aimed to enhance roselle calyx anthocyanins pigment stability for both improved colour and health in cupcakes to evaluate 11 food grade ingredients (FGI): honey, molasses, vinegar, lemon rind, lemon juice, lemon zest, Nescafe, Arabica coffee, orange rind, orange juice, and orange zest.

Materials and Methods

Cupcake ingredients

Cupcake ingredients included all purpose wheat flour (72% extraction), sun-dried loose dark red roselle calyces, caster sugar, Nescafe, honey, unsalted butter, liquid skimmed milk, baking powder and iodized salt, eggs, pure vanilla powder, oranges, Arabica coffee, molasses, and sugar cane vinegar (5%) were all purchased from local markets in Cairo, Egypt. Vinegar, Arabica coffee, Nescafe, honey, and molasses were added to the batters as purchased. However, orange and lemon rinds, juices, and zests were obtained by peeling, squeezing or by grating as needed.

Chemicals

Cyanidin-3-glucoside was purchased from Sigma–Aldrich

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(Darmstadt, Germany). Heat stable α-amylase (*Bacillus licheniformis*, solution, A3306), protease (*Bacillus licheniformis*, lyophilized powder, P3910), and amyloglucosidase (*Aspergillus niger,* solution, A9913) were purchased from Sigma-Aldrich (St. Louis, MO, USA). All other chemicals and solvents were Analar grade.

Preparation of roselle calyces concentrate

A 100 g of dried loose roselle calyces were cleaned by removing visually observed non-calyces matter, then dried in a vacuum dryer at 28°C for 3 hr, cooled at room temperature (23°C), weighed and ground using a coffee machine. Ground roselle calyces were soaked overnight in 200 mL distilled water (DW). In the morning, the suspension was heated at 80°C for 1 hr after adding 450 mL (DW) in a 2 L Erlenmeyer flask. The suspension was strained, 10 g unprocessed sample of each FGI was incubated with 40 g of roselle calyces concentrate for 2 hours at room temperature, and each of these mixtures was added to the other cupcake ingredients to test their ability to keep the roselle calyces red.

Batter preparation

The basic formulation, modified from Gisslen [13], was used for the preparation of 250 g of each cupcake batter and is shown in Table 1. The dry ingredients were sieved together and then the melted butter, eggs, vanilla, and skimmed milk or roselle calyces concentrate (replacing the milk) or roselle calyces concentrate with a FGI were mixed together for 5 min. Cupcake papers were fitted into each of 12 wells cupcake tray (34 cm × 26 cm). Four cupcake papers were filled with each batter on a balance and 60 g batter added, and then baked in a preheated gas oven and baked at 175°C for ~20 min. After baking, cupcakes were left to cool and then packed in polyethylene bags and stored for 12 hr (overnight) in a dry place before sensory testing began. Both moisture determination and sensory testing were begun that morning.

Analytical methods

Moisture was determined using an Infrared Moisture Determination Balance (FD-610-Kett Electric Laboratory, Tokyo, Japan) by weighing 5 g of each cupcake crumb and measured at 80°C for 60 min.

The water loss was calculated according to the following equation:

$$\%\ \text{ML}\ (\text{Moisture loss}) = \frac{W_1 - W_2}{W_1} \times 100$$

Where,

W_1 is the weight of cupcake batter actually transferred into each cupcake paper (~60 g) and W_2 is the weight of the baked cupcake 12 hrs after baking [14]. Total dietary fibre of cupcake samples was determined according to AOAC method 960.52 [15]. Cupcakes samples were lyophilized (Snijders Scientific, Tilburg, Holland, capacity 3 kg ice). After lyophilization cupcakes were weighed again, ground, sieved through a 40-mesh sieve, and stored at -20°C for up to 15 days until analysis. Total dietary fibre was the weight of the residue less the weight of the protein and ash. Protein was determined by Kjeldahl using AOAC official method 991.20 [16]. Hydrolysis was done using a Tecator Digestion System 20, 1015 Digestor (Tecator, Höganäs, Sweden). Ash was determined gravimetrically according AOAC official method 930.30 [17] using a muffle furnace (Nabertherm, D2804, Lilenthial-Bremen, Germany) at 550°C for at least 6 hr. Calculation of total dietary fibre was done according to the following equation:

$$\%\ \text{TDF} = \left[\frac{(R_{Sample} - P_{Sample} - A_{Sample} - B)}{SW}\right] \times 100$$

Where,

Ingredient (g)	Cupcake batter formulation		
	Control	Roselle calyces control	Cupcake samples
Egg	40.0	40.0	40.0
Pure vanilla powder	1.0	1.0	1.0
Caster sugar	55.0	55.0	55.0
Wheat flour	80.0	80.0	80.0
Salt	0.25	0.25	0.25
Milk	50.0	0.0	0.0
Butter	21.0	21.0	21.0
Baking powder	2.75	2.75	2.75
Roselle calyces concentrate	0.0	50.0	0.0
*Treated roselle calyces	0.0	0.0	50.0
Total	250.0	250.0	250.0

*Eleven food grade ingredients were incubated with roselle calyces concentrate: honey, molasses, vinegar, lemon rind, lemon juice, lemon zest, Nescafe, Arabica coffee, orange rind, orange juice, and orange zest.

Table 1: The ingredients used in the cupcake batter formulations.

TDF = Total dietary fibre; R = Average residue weight (mg); P = Average protein weight (mg); A = Average ash weight (mg); SW = Average sample weight (mg); The Residue Weight = W_2-W_1; Ash Weight = W_3-W_1; B = R_{Blank} - P_{Blank} - A_{Blank}; W_1= Celite + crucible weight; W_2 = Residue + celite + crucible weight; W_3 = Ash + celite + crucible weight

Measurement of pH

The pH of roselle calyces concentrate and cupcake samples were measured according to the method of Von Elbe et al. [18] with slight modification. A 0.5 g sample of ground cupcake was mixed with 20 mL of DIW and vortexed for 3 min. The mixture was held at room temperature for 1 hr to separate the solids and liquid. After centrifugation of the liquid for 3 min at 3,050 *x g*, the pH of supernatants was measured.

Determination of anthocyanins

Anthocyanins were determined according to the method of Lee et al. [19]. The method is based on the monomeric anthocyanin pigments reversibly changing colour with a change in pH; the coloured oxonium form exists at pH 1.0, and the colourless hemiketal form predominates at pH 4.5. The difference in the absorbance at 520 nm at these two pH is proportional to the pigment concentration.

The absorption of anthocyanins was measured at both pH at 520 nm and 700 nm using an E-Chrom Tech Spectrophotometer (CT-2200, Taipei, Taiwan) with a DIW blank. The absorbances were measured within 20-50 min of preparation. Calibration curves were prepared using Cyd-3-Glu concentrations of 0, 150, 300, 600, and 1200 mg/L in DIW based on powder weight.

The concentration of anthocyanins were calculated in the samples and expressed as Cyd-3-Glu equivalents (mg/100 g), as follows:

$$\text{Concentration of anthocyanins} = \frac{A \times MW \times DF \times 10^3}{\varepsilon \times 1}$$

Where A = $(A_{520\,nm} - A_{700\,nm})$ pH 1.0 – $(A_{520\,nm} - A_{700\,nm})$ pH 4.5; MW (molecular weight) = 449.2 g/mol for Cyd-3-Glu; DF = dilution factor = 1.5; l = path length in cm; ϵ = 26,900 molar extinction coefficient of Cyd-3-Glu [19].

Known amounts of Cyd-3-Glu were also added to the samples before testing. The recovery ranged from 88-106% for the samples and 95% for the controls. This recovery was within the acceptable range.

Batter viscosity

A simple method to compare different batter viscosities was done using the method described by Ebeler et al. [20]. A funnel with a top inside diameter of 10 cm and a bottom inside diameter of 1.6 cm was used. The funnel was filled to the top with batter and then the batter was allowed to flow for 15 s, to stop the batter flow a palette knife was used to block the outlet. The amount of batter was weighed, divided by 15 to give flow rate in g/s, i.e., higher values indicate lower viscosities.

Batter specific gravity

The specific gravity (density) of the batters was estimated by dividing the weight of a certain volume of batter by the weight of an equal volume of water using the following equation:

$$\text{Specific gravity}\left(\text{g/cm}^3\right)=\frac{(\text{Weight of batter} - \text{filled container} - \text{Weight of container})}{(\text{Weight of water} - \text{filled container} - \text{Weight of container})}$$

A handmade plastic container having an internal volume of ~100 cm^3 was used. The batter was put in using a rubber spatula to carefully fill the dry clean container with minimal air pockets and carefully levelled off with the spatula.

Volume index

The volume index of baked cupcakes was measured one day after baking according to AACC methods 10-91 [21]. Cupcakes were cut vertically through their centre and the heights of the samples were measured at three points (B, C, D; B and D are 3/5 away from the centre (C) along the cross-sectioned cupcakes using the index template. These heights were used to calculate the volume index, contour, and symmetry as described in the official method:

$$\text{The volume index} = B + C + D$$

$$\text{Contour} = (2C - B - D)$$

$$\text{Symmetry} = |B - D|$$

Cupcake density

Since the baked cupcake density and specific volume are reciprocal only the cupcake density was measured using the seed displacement method (AACC, method 74-09) [22].

$$W_{seeds} = W_{total} - W_{cupcake} - W_{container}$$

$$V_{seeds} = \frac{W_{seeds}}{\rho_{seeds}}$$

$$V_{cupcake} = V_{container} - V_{seeds}$$

$$\text{Cupcake density} = \frac{W_{cupcake}}{V_{cupcake}}$$

Where,

W represents weight (g), V represents volume (cm^3), and ρ represents density (g/cm^3). Measurement of seed's density was done by measuring the internal dimensions of a cylindrical container in cm using a ruler to obtain the volume of the container (V). The container was weighed empty (W_1), the container was filled with rapeseeds (obtained from the Egyptian Ministry of Agriculture, Cairo, Egypt), levelled off and reweighed (W_2).

$$\text{The density of the seeds}\left(\text{g/cm}^3\right)=\frac{(\text{Weight of container full of seeds} - \text{Weight of container empty})}{\text{Volume of container}}$$

Colour measurement

Colour measurements (CIE L*a*b*) of baked cupcakes were done using an image analysis technique [23]. A colour image was obtained using a digital camera (Canon, Power Shot A470, 3.4X optical zoom, 7.1 megapixels, Shanghai, China) under controlled and defined illumination conditions using 2 Philips Natural Daylight 18 W fluorescent lamps with a colour temperature of 6500 K according to the manufacturer. The colour of the surface and the crumb of the cupcakes were measured. Sample photos were taken 30.5 cm above the cupcakes at an angle of 45° using a tripod. The pictures were downloaded to a personal computer using a USB digital film reader. Once the colour images of the cupcake samples were captured, the colour was analyzed quantitatively using Photoshop [24]. The captured photos were viewed in the Adobe Photoshop window and from the Info Palette and Histogram Window (Figure 1). The L*, a*, b*, values were calculated (Figure 1), using the following equations according to Yam and Papadakis [23] and Chakraborty et al. [25].

$$L^* = \frac{Lightness}{255} \times 100$$

$$a^* = \frac{240a}{255} - 120$$

$$b^* = \frac{240b}{255} - 120$$

Sensory evaluation

Sensory evaluation was used to assess the sensory acceptability of the cupcakes using an acceptance test. Cupcakes were subjected to sensory evaluation by untrained panellists. Prospective panellists were screened using the following criteria: 1) female >20 yr (n = 50), 2) employees who have eaten cake or cupcakes at least once a week for the last three months, and (3) have no food allergies (n = 15). They were neither trained nor given prior information about any of the ingredients

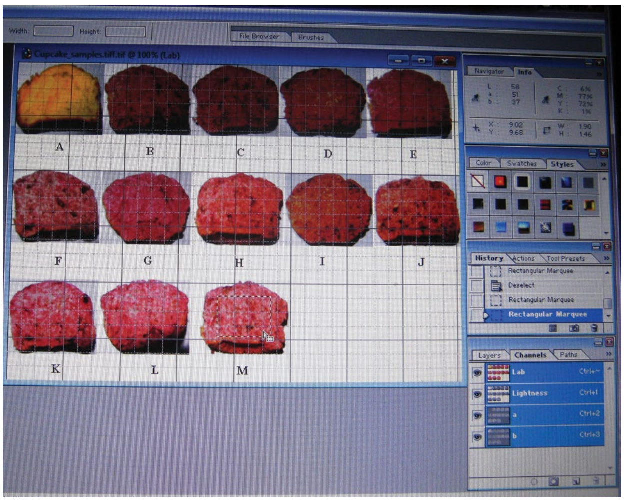

Figure 1: The Photoshop Window Screen of cupcake crumb samples appearing the determined area in M sample that will be measured, show the transversal cut of cupcakes, A= Control; B = Control roselle calyces concentrate; C = Honey; D = Molasses; E = Vinegar; F = Lemon rind; G = Lemon juice; H = Lemon zest; I = Nescafe; J = Arabica coffee; K = Orange rind; L = Orange Juice; M = Orange zest.

use. The room temperature cupcakes were presented to the panellists in random order [26,27]. The panellists were instructed to score their liking for each of the attributes being studied. A 5-point hedonic scale with 1 = dislike very much, 2 = dislike moderately, 3 = neutral, 4 = like moderately to 5 = like very much was used to evaluate the colour, appearance, texture (visual and eating), taste, volume, lightness, aroma and overall liking of the cupcakes.

Statistical analysis

Moisture content, pH, anthocyanins, batter viscosity, batter specific gravity, volume index, colour measurements, cupcake density and sensory evaluations were done in triplicate, and mean values and standard deviations were calculated using Excel for Microsoft Windows Operating System. One-way ANOVA was done and the differences of the means were evaluated using Tukey's HSD test (P<0.05) using SPSS 16.0 for Windows (SPSS Inc., Chicago, IL, USA).

Results and Discussion

The results of anthocyanin, fibre, moisture, water loss and pH of baked cupcake samples are summarized in Table 2.

Anthocyanin content

Monomeric anthocyanins in the roselle calyces concentrate were 520 mg/100 g Cyd-3-Glu. Anthocyanins increased statistically (P<0.05) for all cupcakes compared to the control; the most effective additives in retaining anthocyanins were vinegar, lemon and orange juice (Table 2). Incubating roselle calyces concentrate with honey, molasses, Arabica coffee and Nescafe significantly reduced anthocyanins (P< 0.05) and cupcakes became dark brown. The red flavylium cation may be degraded due to the high temperatures used during baking and converted into colourless carbinol pseudo base, which subsequently undergoes an opening of the pyrilum ring, leading to the formation of brown-coloured chalcone [8].

Žilić et al. [9] recently showed that adding citric acid to pigmented corn dough increased the phenolic compounds in corn cookies stabilizing the anthocyanins. The citric acid helped retain anthocyanins by lowering pH and by acylation of their sugar residues or flavylium cation.

Sample	Anthocyanins mg/100g	Fibre %	Moisture %	Water loss	pH
Roselle calyces C.	520 ± 0.7	15 ± 0.8	63 ± 2	NA	5.5 ± 1.3
Control	0	10 ± 1^{e}	25 ± 2^{d}	8 ± 0.2^{b}	7.9 ± 2.1^{a}
Roselle calyces	400 ± 0.1^{a}	14 ± 1^{a}	23 ± 2^{f}	5.6 ± 0.3^{c}	7.5 ± 1.5^{a}
Honey	250 ± 0.3^{c}	9 ± 0.3^{f}	27 ± 2^{b}	5 ± 0.4^{d}	5.7 ± 0.6^{d}
Molasses	220 ± 0.1^{c}	9 ± 0.6^{f}	26 ± 2^{c}	5 ± 0.2^{d}	7.1 ± 0.5^{b}
Vinegar	410 ± 0.1^{a}	8 ± 0.3^{g}	27 ± 3^{b}	4 ± 0.5^{e}	4.5 ± 0.3^{e}
Lemon r.	270 ± 0.3^{c}	11 ± 0.2^{d}	27 ± 1.7^{b}	3 ± 0.4^{f}	7.7 ± 0.4^{a}
Orange r.	300 ± 0.2^{b}	12 ± 1^{c}	27 ± 1^{b}	2.8 ± 1.2^{f}	6.4 ± 0.7^{c}
Lemon j.	420 ± 0.3^{a}	10 ± 1^{e}	26 ± 3^{c}	5 ± 1^{d}	4.6 ± 0.9^{e}
Orange j.	380 ± 0.2^{b}	9 ± 0.4^{f}	26 ± 3^{c}	4.4 ± 1.5^{e}	5.2 ± 0.6^{e}
Lemon z.	390 ± 0.1^{b}	11 ± 0.1^{d}	27 ± 2^{b}	4.7 ± 0.9^{d}	5.5 ± 0.1^{d}
Orange z.	260 ± 0.2^{c}	13 ± 0.5^{b}	28 ± 3^{a}	2.7 ± 0.1^{f}	5 ± 0.5^{e}
Arabica c.	230 ± 0.2^{d}	11 ± 1^{d}	24 ± 2^{e}	10 ± 3^{a}	5.8 ± 0.8^{d}
Nescafe	250 ± 0.2^{c}	9 ± 0.2^{f}	27 ± 3^{b}	4.7 ± 0.3^{d}	7.1 ± 0.4^{b}

Means that do not share the same letter in a column are significantly different according to Tukey HSD test (P□0.05). (n = 3 cupcakes/sample). C = control, r = rind, j = juice, z = zest, c = coffee.

Table 2: Average anthocyanin, fibre, moisture, water loss, and pH of baked cupcake samples.

Selim et al. [10] showed that low pH values (1.5 to 3) optimized anthocyanin stability. They showed that roselle calyces extracts heated for 30 min at temperatures of 60, 70, 80, 90, and 100°C, retained 99.9, 99.2, 94, 86, and 79% of their anthocyanins contents, respectively. In this study the heating above 50°C occurred at 2 stages: heating ground roselle calyces concentrate with water to extract the pigment at 80°C for 1 hr, and baking cupcakes at 175°C for ~20 min.

No studies were found that evaluated the effect of honey and molasses on anthocyanin stability during baking. Honey and molasses significantly reduced (P<0.05) anthocyanins in roselle calyces cupcakes. The reduction of anthocyanins in baked cupcakes ranged between 19-58%. Fructose in honey can interact with amino acids to give brown Maillard reaction products [28]. The brown colour may also be due to brown pigment in coffee and molasses along with some caramelization during heating.

Fibre content

Roselle calyces concentrate had 15% dietary fibres while the dietary fibre in cupcakes ranged from 8 to 14%. Duke and Atchely [29] found 2.3% fibre in roselle calyces while Gabb [30] found 12% fibre. These differences may be due to different cultivars or to differences in the methodologies for fibre determinations. The orange zest contained 13% dietary fibre and the lemon and orange rinds increased the dietary fibre in cupcakes to 11-12%, which may also reflect the high pectin (~30%) in orange rind [31]. Although the addition of fibre to baked products is associated with a decrease in volume due to partial dilution of gluten [32] yet the volume of the roselle calyces control samples gave a similar volume as the control. In another study, incorporation of 10% red raspberry juice decreased the volume of muffins [33]. They suggested that the rapid release of carbon dioxide (CO_2) due to the reduction in pH led to the decreased volume. The acidification also led to air pockets (pores) inside the muffins, which was also observed in the cupcakes treated with acids.

Moisture content and pH

Moisture content ranged between ~23-28%. Incorporation of 4% FGI significantly (P<0.05) increased the moisture content of cupcakes with a maximum of 28% with lemon and orange rind and orange zest. This could be attributed to the hygroscopic nature of lemon and orange. Incorporation of roselle calyces concentrate at 20% decreased the moisture (23.3%) compared to the control cupcakes. The roselle calyces concentrate's pH was 5.5 while it ranged in cupcakes from 4.5 to 7.7 in cupcakes with vinegar and lemon rind, respectively.

Batter viscosity

Batter viscosity (Table 3) ranged from 1.1 g/s to 1.9 g/s for vinegar (highest viscosity) and orange rind, respectively. The higher batter viscosity for molasses and honey may reflect their capability to absorb water and slow down the rate of gas diffusion during the early stages of baking [34]. The increase in the water absorption capacity of ingredients such as honey and molasses reduces the amount of free water available to facilitate the movement of particles in batters and consequently gives high viscosity to cupcake batters [35].

During baking, the wheat gluten forms a network that traps gas in the batter and provides a smooth texture. However, increased amount of CO_2 could produce peaks and tunnels in the crumb, which is not desired [36]. These were observed in the honey, lemon juice and Arabica coffee cupcakes. Wong [37] also reported that decreasing the pH would dissociate the micro fibrils within the gliadin protein fraction into its monomers, thereby increasing the formation of air pockets.

Batter density

Low batter density is desired in cupcake batter because it indicates that more air is incorporated into the batters, which distinguish the consistency of cakes [38]. Batter density ranged from 1.03 g/cm^3 to 1.2 g/cm^3 for orange juice, Nescafe and lemon juice, and Arabica coffee, respectively (Table 3). A low viscosity batter can not hold the air bubbles sufficiently. Batter density decreases during cake baking because of the loss of water and the increase of cupcake volume because of gas expansion. However, the final cupcake volume is not only dependent on initial air incorporated into the batter but also its capacity to retain air during baking [39].

Cupcake density

The cupcake density ranged from 0.391 to 0.5 g/cm^3 for molasses and Arabica coffee, respectively (Table 3). Excessive batter viscosity may cause disruption of air bubble production during baking. Thus, there is an optimum cupcake batter viscosity to achieve cupcakes with high volume; if the viscosity of the batter is too low or too high, the batter can not hold the air bubbles sufficiently and the cupcakes collapse in the oven [35].

The cupcake volume index ranged from 105 mm to 115 mm for lemon juice and orange rind, respectively (Table 3). The volume index indicates the amount of air entrapped in the cupcake crumb. Although a high volume index does not always indicate to a desirable cupcake, low volumes generally indicate a heavy and less desirable crumb [40]. The lemon and orange rind and Arabica coffee gave the cupcakes a small structure. It was also noticed that the crust of these cupcakes were cracked in the middle of the crust suggesting that these ingredients increased the gas production during baking that may have led to structure collapse.

Cupcake contour and symmetry

Cupcake contour ranged from 6 mm to 8.5 mm for Nescafe and orange rind, respectively (Table 3). The cupcakes with higher volumes showed higher central height. Generally, a peaked cupcake would have a higher contour value and a flat cake would have a lower value. All treatments resulted in cupcakes with intermediate contour values, reflecting cupcakes with appropriate rounded surfaces. Cupcake symmetry ranged from 1.2 mm to 2.5 mm for vinegar and orange rind, respectively (Table 3). A low symmetry value is more desirable.

Crust and crumb colour of cupcakes

Results in Table 4 showed that the crust colour was dependent on the FGI. It seems that FGI and baking temperature were the main factors that influenced the colour of the crust. The crumb of the control cupcake was lighter than the crust as the Maillard reactions occur mainly on the surface. The a* value increased significantly (P<0.05) with the acids (lemon and orange juice, and vinegar). The b* was significantly (P<0.05) reduced by addition of roselle calyces and all FGI. Some of these additives turned the cupcakes purple, red and brown (Figure 1). The pinkish colour increased significantly (P<0.05) when vinegar, and lemon or orange juice were used. However, when baking with honey or molasses a dark brown colour was obtained (Table 4, Figure 1).

A recent study evaluated the effect of various organic acids on colour retention during fruit juice storage, and found that acetic acid improved the colour stability in both elderberry and black currant juices, whilst the citric and tartaric acids only improved colour stability in elderberry juice [8]. It could be concluded that adding acids to the roselle calyces concentrate help to stabilize the red colour of roselle calyces probably by lowering the pH. Bronnum-Hansen et al. [41] noted that the efficiency of extracting solvents for anthocyanins increased with increasing concentration of citric acid. The lemon and orange juices currently used also improved the colour [8,9].

Sensory evaluation

The results are shown in Table 5. Cupcakes with FGI had high acceptance scores for texture (2.5-4.5) and aroma (2-5). However, cupcakes with molasses, Arabica coffee and Nescafe showed a significant (P<0.05) decrease in redness due to colour (dark brown). There was a significant difference (P<0.05) with respect to the acceptance of colour of the treated cupcakes compared to control. Cupcakes with honey and Arabic coffee had a lower acceptance for all attributes (3-3.3) although this was still above the value of 2.8 that was set as the acceptable point for overall liking. Smith and Johnson [42] noted when honey replaced sucrose in different types of cakes the quality was poor due to dense structure, low volume, dark crumb, and an undesirable flavour. However the volume of the honey cupcakes was not affected with low amount (4%) used. The panellists gave lower scores for taste and aroma of the honey samples. Most panellists liked the appearance and

Sample	Batter viscosity (g/s)	Batter density (g/cm^3)	Cupcake density (g/cm^3)	Volume index (mm)	Contour (mm)	Symmetry (mm)
Control	1.6 ± 0.1^b	1.0 ± 0.03^c	0.4 ± 0.04^b	106 ± 3^d	5.0 ± 0.3^f	1.5 ± 0.3^d
Roselle calyces	1.5 ± 0.03^c	1.0 ± 0.02^c	0.4 ± 0.03^b	107 ± 6^c	5.6 ± 0.4^e	1.6 ± 0.1^d
Honey	1.3 ± 0.05^c	1.0 ± 0.05^c	0.5 ± 0.01^a	110 ± 2^b	6.2 ± 0.5^e	2.2 ± 0.2^b
Molasses	1.4 ± 0.01^c	1.0 ± 0.03^c	0.4 ± 0.01^b	108 ± 3^c	5.8 ± 0.1^e	2.0 ± 0.2^c
Vinegar	1.9 ± 0.05^a	1.1 ± 0.1^b	0.4 ± 0.03^b	107 ± 4^c	6.3 ± 1.5^e	1.6 ± 0.3^d
Lemon r.	1.4 ± 0.02^c	1.1 ± 0.1^b	0.5 ± 0.02^a	114 ± 5^a	7.0 ± 0.2^c	2.4 ± 0.1^a
Orange r.	1.4 ± 0.04^c	1.1 ± 0.07^b	0.5 ± 0.01^a	115 ± 2^a	8.5 ± 1^a	2.5 ± 0.4^a
Lemon j.	1.6 ± 0.08^b	1.0 ± 0.02^c	0.5 ± 0.04^a	105 ± 5^d	7.5 ± 0.4^b	2.0 ± 0.3^c
Orange j.	1.6 ± 0.03^b	1.0 ± 0.01^c	0.4 ± 0.03^b	109 ± 2^c	6.7 ± 0.4^d	1.8 ± 0.1^c
Lemon z.	1.4 ± 0.05^c	1.1 ± 0.05^b	0.4 ± 0.02^b	109 ± 4^c	5.7 ± 0.2^e	2.0 ± 0.2^c
Orange z.	1.4 ± 0.04^c	1.1 ± 0.1^b	0.5 ± 0.01^a	110 ± 5^b	6.5 ± 0.1^d	2.3 ± 0.1^b
Arabica c.	1.0 ± 0.06^d	1.2 ± 0.06^a	0.5 ± 0.01^a	112 ± 2^b	6.5 ± 0.5^d	2.3 ± 0.4^b
Nescafe	1.5 ± 0.05^c	1.0 ± 0.1^c	0.4 ± 0.01^b	109 ± 2^c	6.0 ± 0.4^e	1.9 ± 0.2^c

Means that do not share the same letter in a column are significantly different according to Tukey HSD test (P□0.05). (n = 3 cupcakes/sample). r = rind, j = juice, z = zest, c = coffee.

Table 3: The effect of different roselle calyces concentrates treatments on batter and cupcake quality parameters.

texture of cupcakes with Nescafe, molasses and orange zest compared to the control cupcake. The Arabica coffee cupcakes were hard and dry. Overall the FGI treated samples had significantly (P<0.05) improved sensory attributes for various reasons. Panellist comments suggested that the molasses had a distinguished brown chocolate colour and spongy crumbs while both the Nescafe and molasses had an aroma and crumb texture was highly liked.

Molasses, Nescafe, lemon and orange zest cupcakes were the most liked (liking scores of 4.5-4.6). Low density, high volume, desirable lightness and, appearance and pleasant aroma, and appropriate colour seem to be the important parameters that resulted in the highest liking score. Least like were the honey, lemon and orange rind, and vinegar (Table 5). Cupcakes containing orange zest and molasses showed the highest desirability for almost all sensory parameters.

The FGI lowered the acceptability of the volume probably because these additives may have inhibited the swelling of starch granules and also by forming a film around the granules [43,44] thereby increasing the gelatinization temperature [45].

Sample	Crust			Crumb		
	L*	a*	b*	L*	a*	b*
Control	71 ± 3[a]	10 ± 0.2[f]	61 ± 3[a]	89 ± 3[a]	-0.5 ± 0.1[e]	65 ± 3[a]
Roselle calyces	16 ± 1[g]	15 ± 1[d]	8 ± 0.3[c]	54 ± 1[c]	40 ± 3[c]	25 ± 1[f]
Honey	24 ± 1[e]	19 ± 1[c]	10 ± 0.2[b]	51 ± 1[d]	42 ± 2[c]	25 ± 0.4[f]
Molasses	16 ± 1[g]	9 ± 0.4[f]	8 ± 0.4[c]	56 ± 2[c]	41 ± 1[c]	40 ± 2[c]
Vinegar	27 ± 1[d]	25 ± 1[b]	8 ± 0.2[c]	56 ± 3[c]	48 ± 3[a]	28 ± 2[e]
Lemon r.	22 ± 1[e]	11 ± 1[f]	3 ± 0.2[d]	61 ± 2[b]	29 ± 1[d]	25 ± 2[f]
Orange r.	23 ± 1[e]	17 ± 0.4[e]	5 ± 0.4[d]	54 ± 3[c]	45 ± 3[b]	30 ± 5[e]
Lemon j.	39 ± 1[b]	40 ± 2[a]	9 ± 0.4[c]	53 ± 2[c]	48 ± 2[a]	21 ± 3[g]
Orange j.	36 ± 1[c]	26 ± 1[b]	11 ± 3[b]	60 ± 4[b]	44 ± 2[b]	28 ± 8[e]
Lemon z.	27 ± 1[d]	20 ± 1[c]	7 ± 4[c]	60 ± 2[b]	45 ± 2[b]	34 ± 5[d]
Orange z.	29 ± 6[d]	19 ± 1[c]	7 ± 5[c]	61 ± 3[b]	41 ± 2[c]	32 ± 6[d]
Arabica c.	20 ± 7[f]	15 ± 1[d]	7 ± 1[c]	55 ± 2[c]	43 ± 2[c]	41 ± 3[c]
Nescafe	29 ± 2[d]	20 ± 1[c]	11 ± 1[b]	53 ± 3[c]	44 ± 2[b]	49 ± 2[b]

Means that do not share the same letter in a column are significantly different according to Tukey HSD test (P□0.05). (n =3 cupcakes/sample). The surface and crumb colour of cupcakes are reported as average L* (lightness), a* (redness), b* (yellowness) values. r = rind, j = juice, z = zest, c = coffee.

Table 4: Effect of roselle calyces cupcake on the colour of the crust and crumb of cupcakes.

Needs assessment

Cupcake is popular in Egypt and hundreds of bakery and specialised shops produce cupcakes. The market of cupcakes in Egypt is large, the market is targeting people through cupcake shops, TV cooking programmes and specialised cooking channels. However, our target in this study is children as they like coloured cupcakes and the most sensitive group for over consumption of synthetic food colourants as these shops use them apparently without official control.

Our cupcakes are red, purple and brown with clean label. Also our target is older people who looking for healthy and soft products, that can be made commercially to be labelled as functional cupcakes. Roselle calyces cupcake can provide food safety for children and protective agents for older people. Beneficial health effects associated with anthocyanin consumption include lowering the risk of cardiovascular disease (CVD) was reported [46]. The Egyptian National Hypertension Project found an adjusted overall prevalence of coronary heart disease of 8.3% and a high prevalence of hypertension (26%) that was an important driver of adverse cardiovascular outcomes [47].

Cost-effective product

The current study focused on using roselle calyces and FGI to produce a new cupcake-like product (CLP); as a cost effective (Roselle calyces and FGI are available and cheap) and functional cupcake food with high nutritional value (specifically bioactive compounds). The cost effective in the new product is reasonably cheap compared to the control cupcake costs. One of the most important factors that influence the cost effectiveness of producing cupcake is the choice and the prices of ingredients such as flour. The price of flour based on a retailer price in Egypt is between L.E 7-8 (1 kg), while the commercial cupcakes are L.E 10. The real calculation of each cupcake in this study as only ingredients for roeslle cupcakes is L.E 2.60 compared to control (using milk) L.E 3.20. With the assumption of reducing the cost to ~ 19% as (~ L.E 0.6) when roselle calyces and FGI added, in addition to its higher nutritional value. As we've found in the literature, total phenols, anthocyanin, and fibre content exert their beneficial effects in reducing risks of coronary heart disease [26,48-51]. Additionally, its use could contribute to reducing the cost of national heath services in the treatment of heart diseases [52].

Sample	Colour	Appearance	Texture	Taste	Volume	Lightness	Aroma	Overall liking
Control	5.0 ± 0.1[a]	4.8 ± 0.2[a]	4.9 ± 0.3[a]	4.8 ± 0.3[a]	5.0 ± 0.1[a]	5.0 ± 0.2[a]	4.8 ± 0.3[a]	4.9 ± 0.3[a]
Roselle calyces	3.9 ± 0.2[b]	4.2 ± 0.2[b]	4.3 ± 0.4[b]	3.7 ± 0.2[c]	4.5 ± 0.2[a]	4.4 ± 0.2[b]	4.3 ± 0.4[b]	4.2 ± 0.2[b]
Honey	3.4 ± 0.3[b]	3.6 ± 0.3[c]	3.7 ± 0.3[c]	2.2 ± 0.1[e]	3.3 ± 0.3[c]	3.5 ± 0.3[c]	2.0 ± 0.3[c]	3.0 ± 0.1[d]
Molasses	4.5 ± 0.1[a]	4.8 ± 0.1[a]	4.5 ± 0.3[a]	4.6 ± 0.2[a]	4.6 ± 0.1[a]	4.8 ± 0.1[a]	4.5 ± 0.3[a]	4.6 ± 0.2[a]
Vinegar	3.8 ± 0.1[b]	3.7 ± 0.1[c]	3.4 ± 0.3[c]	3.5 ± 0.2[c]	3.4 ± 0.1[b]	3.8 ± 0.1[c]	3.4 ± 0.3[c]	3.5 ± 0.2[c]
Lemon r.	3.7 ± 0.3[b]	4.2 ± 0.4[b]	3.0 ± 0.3[d]	3.2 ± 0.3[c]	3.4 ± 0.3[b]	3.3 ± 0.4[d]	3.5 ± 0.3[c]	3.0 ± 0.3[d]
Orange r.	3.0 ± 0.2[c]	2.8 ± 0.2[d]	2.3 ± 0.2[e]	3.0 ± 0.3[d]	2.7 ± 0.2[d]	2.8 ± 0.2[a]	3.8 ± 0.2[c]	2.9 ± 0.3[d]
Lemon j.	5.0 ± 0.3[a]	4.5 ± 0.6[a]	4.0 ± 0.4[b]	4.0 ± 0.1[b]	4.5 ± 0.3[a]	3.9 ± 0.6[c]	4.0 ± 0.4[b]	4.0 ± 0.1[b]
Orange j.	4.4 ± 0.2[b]	3.8 ± 0.2[c]	3.5 ± 0.2[c]	3.7 ± 0.3[c]	3.4 ± 0.2[b]	3.8 ± 0.2[c]	3.8 ± 0.2[c]	3.8 ± 0.3[c]
Lemon z.	4.5 ± 0.1[a]	4.2 ± 0.2[b]	4.3 ± 0.2[b]	4.1 ± 0.1[b]	3.9 ± 0.1[b]	4.2 ± 0.2[b]	4.5 ± 0.2[a]	4.5 ± 0.1[a]
Orange z.	4.6 ± 0.2[a]	4.4 ± 0.2[a]	4.8 ± 0.2[a]	5.0 ± 0.3[a]	5.0 ± 0.2[a]	4.8 ± 0.2[a]	5.0 ± 0.2[a]	4.6 ± 0.3[a]
Arabica c.	2.3 ± 0.2[d]	3.5 ± 0.2[c]	2.5 ± 0.2[d]	2.0 ± 0.3[e]	2.2 ± 0.2[d]	2.1 ± 0.2[d]	3.5 ± 0.2[c]	3.3 ± 0.3[c]
Nescafe	4.6 ± 0.2[a]	4.8 ± 0.2[a]	4.7 ± 0.2[a]	4.9 ± 0.3[a]	4.3 ± 0.2[b]	4.5 ± 0.2[a]	4.9 ± 0.2[a]	4.5 ± 0.3[a]

Means that do not share the same letter in a row are significantly different according to Tukey HSD test (P□0.05). A 5-point hedonic scale ranging from 1 = Dislike very much; 3 = Neither like or dislike; 5 = Like very much was used to evaluate the attributes in the table. r = rind, j = juice, z = zest, c = coffee. (n = 13 samples, n = 15 panellists ± S.D.; P□0.05), one cupcake each. The total n for all values is 102 (50 panellists and 13 samples x 4 tables).

Table 5: Sensory evaluation scores for cupcakes.

Conclusion

Roselle calyx concentrates were incubated with 11 food grade ingredients (FGI), and added to cupcake batters. The sensory panellists gave the best score (statistically significant at $P<0.05$) to the roselle calyx cupcakes with molasses and orange zest. The a* value was increased significantly while the b* value was significantly reduced. The consumption of 100 g of the roselle calyx cupcakes with lemon juice provided 420 mg/100 g anthocyanins and 10% total dietary fibre.

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