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SENSORY, PHYSICO-CHEMICAL, AND FUNCTIONAL PROPERTIES OF LUPIN-BASED CHOCOLATE DESSERT FORMULATIONS PREPARED FROM EGYPTIAN-SWEET WHITE LUPIN SEEDS

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ABSTRACT: The aim of this study was to evaluate the sensory, physico-chemical, and functional properties of lupin-based chocolate dessert formulations. The lupin milk was prepared by grinding the peeled seeds after a debittering process which involved seed soaking for 6 hrs and the alkaline treatment with a hot solution of sodium bicarbonate (0.3%, 93°C) for 6 hrs. Five new formulations of lupin-based chocolate dessert were prepared by substituting the lupin milk with dark chocolate at different ratios ranging from 5 to 25% with an interval increase of 5 %. The obtained results showed the significant effect of the substitution ratio on the sensory, physicochemical and functional properties of lupin-based chocolate dessert. A significant gradual improvement in the sensory characteristics was observed with the increase in the substitution ratio. Similarly, a significant gradual increase in the proximate chemical composition, except the content of protein and Ca, was observed with the increase in the substitution ratio. Contrary, the water holding capacity showed also a decreasing tendency when the substitution ratio increased. Using lupin milk as alternative to cow milk and dark chocolate as a partial substitute to lupin milk in the preparation of chocolate dessert is a promising technique due to the increased level of protein, fat, fiber, ash and water holding capacity in lupin-based chocolate dessert compared to the commercial dessert (Danette).

Key words: Sweet white lupin seeds, chocolate dessert, sensory characteristics, chemical properties, color, water holding capacity.

INTRODUCTION

Lupin has recently been studied for its promoting-health effects because lupin seeds are rich in protein, dietary fibre, and minerals (Islam and Ma, 2016). Lupins are members of the family Leguminosae that include many species such as *Lupinus albus* (white lupin), *L. angustifolius* (blue or narrow-leafed lupin), and *L. luteus* (yellow lupin) and *L. mutabilis* (pearl lupin or Tarwi) (Hondelmann, 1984). White lupin (*Lupinus albus* L.) seeds contain considerable amount of protein ranging between 33 and 47%, which is higher than the other legumes and close to the soy protein content. Contrary to cereals, lupin proteins contain high amount of lysine and low amount of sulphur-

containing amino acids (Dervas *et al.*, 1999). The hull represents significant part of the lupin seeds (about 20%) with a high content of dietary fibre (generally insoluble and is higher than in soy bean), and antioxidants (Pisarikova and Zraly, 2010). Lupin has a considerable amount of oil/fat (5–20%) in the whole seed although it is not an oilseed crop (Mohamed and Rayas-Duarte, 1995). In addition to the high dietary fiber, fat and ash content, lupin seeds also have some functional properties such as emulsifying and solubility (Erbaş *et al.*, 2005; Yorgancilar and Bilgiçli, 2014).

Despite the importance of lupin seeds as a protein source, its uses in food applications is limited by the presence of toxic alkaloids and other anti-nutritional compounds (Muzquiz *et*

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al., 1998). However, these compounds could be eliminated by many processes (Jiménez-Martínez et al., 2001). Some studies highlight the possibility to improve the nutritional value of lupin seeds by applying methods such as dehulling (Erbaş, 2010), germination (Chilomer et al., 2012), roasting (Embaby, 2010), autoclaving (Brenes et al., 1993), fermentation (Omer et al., 2016), or extrusion (Masoero et al., 2005). Some of the studies show that the dehulling process can have a positive effect on the content of certain nutrients. The effect of dehulling and different debittering process on the chemical composition of lupin seeds was studied by many authors (Saez et al., 2015; Erbaş, 2010). The dehulling of white lupin seeds resulted in an increase in the crude protein content, while the crude fiber decreased (Struti et al., 2020). Similarly, Van Barneveld et al. (1999) showed that dehulled lupin seeds have a comparable protein level to that of soybean meal. On the other hand, debittering processes containing cooking and soaking caused significant decrease in chemical compositions (ash and minerals) and antinutrients such as phytic acid (Ertaş and Bilgiçli, 2014). Similarly, Mostafa et al. (2013) reported that the total alkaloid of lupin seeds decreased after the debittering process. Therefore, debittering process is necessary for removing toxic substances and antinutrients and also for improving eating quality of lupin. Functional foods are gaining more attention due to their health promoting benefits. Lupin seeds with 34.44–39.42 % dietary fiber content (3.64–5.21 % soluble and 30.80–34.22% insoluble) may also be a potential source for the production of dietetic food (Martínez-Villaluenga et al., 2006). Lupin-enriched foods boost satiety and reduce energy intake lowering the levels of blood pressure, blood glucose, and cholesterol levels. Lupin proteins are considered to be the key contributor to these claimed health benefits. In Egypt, lupin seeds are generally consumed as a snack food after removing of alkaloids and salting. Lupin seeds and flours are used in different cereal products as pasta, crisp bread, cookies, cakes, breakfast cereals, (Dervas et al., 1999; Erbaş et al., 2005). Market demands in different milk alternatives are in the rise in current years (Pontonio and Rizzello, 2021; Aydar et al., 2020). Due to increased change of lifestyle, special demand of diets and functional foods popularity, milk alternatives are recently

attracting attention. Substitutes of milk in formulated food products are derived either from plants such as cereal, legume or nut-based milks (Pontonio and Rizzello, 2021) or plant-based food byproducts (Gemechu, 2020). There are growing commercially available milk alternatives having significant physicochemical properties (Jeske et al., 2017). Similar to soy milk, lupine milk is among the alternative constituents of animal product source foods such as cheese (Al-Saedi et al., 2021), yogurt (Mohamed et al., 2019), tofu (Jayasena et al., 2010), and ice cream (Asres et al., 2022). The consumption of dark chocolate, which has a greater content of cocoa and lower content of sugar, is associated with a reduction in blood pressure, cardiovascular diseases, type 2 diabetes, fat deposition, and mortality rates (Allen et al., 2008; Almoosawi et al., 2012). Besides, Sørensen and Astrup (2011) reported the significant effect of dark chocolate on a greater satiety feeling, lower energy intake in prospective meal and reduced appetite for sugary snacks in comparison with milk chocolate consumption. A large body of evidence has shown the beneficial effects of functional foods on promoting health and reducing risk of certain diseases (Andersen and Fernandez, 2013). In this study, lupin milk was prepared from white lupin seeds after debittering process and then substituted with dark chocolate at different ratios ranging from 5 to 25% to produce functional chocolate dessert which is similar to the commercial chocolate dessert (Danette). Therefore, the objective of this study was to evaluate the effect of partial substitution for lupin milk with dark chocolate on the sensory and chemical properties, color characteristics, and water holding capacity of lupin-based chocolate dessert.

MATERIALS AND METHODS

Raw Materials

Egyptian sweet white lupin seeds (*Lupinus albus* L.), dark chocolate (Dreem dark cooking chocolate), and commercial chocolate dessert (Danette: manufactured by Danone Company, Egypt) were purchased from the local market (Zagazig City, Sharqia Governorate, Egypt). Sodium bicarbonate (NaHCO_3) was purchased from Hi-Lab for chemicals (Zagazig, Sharqia, Egypt).

Debittering Process

The lupin seeds were manually cleaned to remove the foreign materials and immature and damaged seeds. The seeds were then soaked in distilled water at ratio of 1: 6 (seed: distilled water) for 6 hrs, with water changing every two hrs. The lupin milk was prepared according to **Jiménez-Martínez *et al.* (2001)** with a slight modification. Briefly, 500 g of lupin seeds was immersed in a hot solution of sodium bicarbonate solution (0.3%) and maintained at $93\pm 2^{\circ}\text{C}$ for 6 hrs in order to eliminate the alkaloids such as lupanine and albine. The seeds were then washed several times with running clean tap water to completely eliminate the traces of the alkaline solution, and then manually peeled (Fig. 1).

Preparation of Lupin Milk

The peeled seeds were mixed with distilled water with a ratio of 1:4 to facilitate the subsequent grinding operation. The lupin milk was prepared by grinding the mixture using a food processor (Moulinex, Type: LM201, 700W, France) at high speed for 5 min. it is worth mentioning that the pH of the lupin milk was close to 7.3.

Preparation of Lupin-based Chocolate Dessert

The dark chocolate (Dreem cooking chocolate) was firstly grated by a kitchen grater and five formulations were prepared by substituting the lupin milk with the dark chocolate at different ratios ranging from 5 to 25% with an interval increase of 5 %. The lupin milk was heated in a hot water bath at $93\pm 2^{\circ}\text{C}$ and the respective ratio of dissolved chocolate was added. After substituting, the blend was maintained at this temperature for 20 min. After a complete dissolving, the blend was immediately cooled in a cold water bath to 4°C . The final product was then packed in plastic cups (Fig. 1) and stored under cooling conditions until uses. A commercial chocolate dessert (Danette: manufactured by Danone Company, Egypt) was used for comparison, it contains modified cow milk, sugar, coconut oil, cacao, starch, and chocolate flavoring. According to preliminary trials, the maximum level of substitution was limited to 25 % of dark chocolate (Dreem cooking chocolate) because the samples

prepared at a higher level than 25 % were refused by panelists due to the high firmness and solidified texture.

Assessments and Characterizations

All characterization and assessments were performed in triplicates for the lupin milk, lupin-based chocolate dessert formulations, and commercial chocolate dessert (Danette).

Sensory evaluation

The lupin-based chocolate dessert formulations and the commercial chocolate dessert (Danette) were subjected to sensory evaluation by a panel of 20 members including 5 males and 15 females aged from 25 to 65 years. A commercial chocolate dessert (Danette: manufactured by Danone Company, Egypt) was used as a reference. Samples were coded using random three-digit numbers and the panelists were provided with a glass of water for rinsing between samples, and with an instructed sheet as described by **Mounir *et al.* (2019)**. The different samples were evaluated for the overall acceptability based on their appearance, color, taste, odor, and texture, using a nine-point hedonic scale (1 = extremely dislike to 9 = extremely like).

Determination of chemical characteristics

The proximate chemical composition was determined for the lupin milk, the lupin-based chocolate dessert formulations, and the commercial chocolate dessert (Danette). The contents of protein, fat, fibers, ash, and water were determined according to the methods described in **AOAC (2019)**. Carbohydrates were calculated by difference using Eq. (1) as follows:

$$\text{Carbohydrates (\%)} = 100 - (\% \text{ Protein} + \% \text{ Fat} + \% \text{ Ash} + \% \text{ water}) \quad (1)$$

Major and trace elements such as calcium (Ca), phosphorus (P), magnesium (Mg), iron (Fe), and zinc (Zn), were determined according to **Gul and Safdar (2009)**, using an atomic absorption spectroscopy (Thermo Fisher Scientific, model iCAP 7400 analyzer, Germany).

The pH of different samples was measured in triplicate by digital pH meter (model 646 Digital, USA) according to **Abbasi and Azizpour (2016)**.

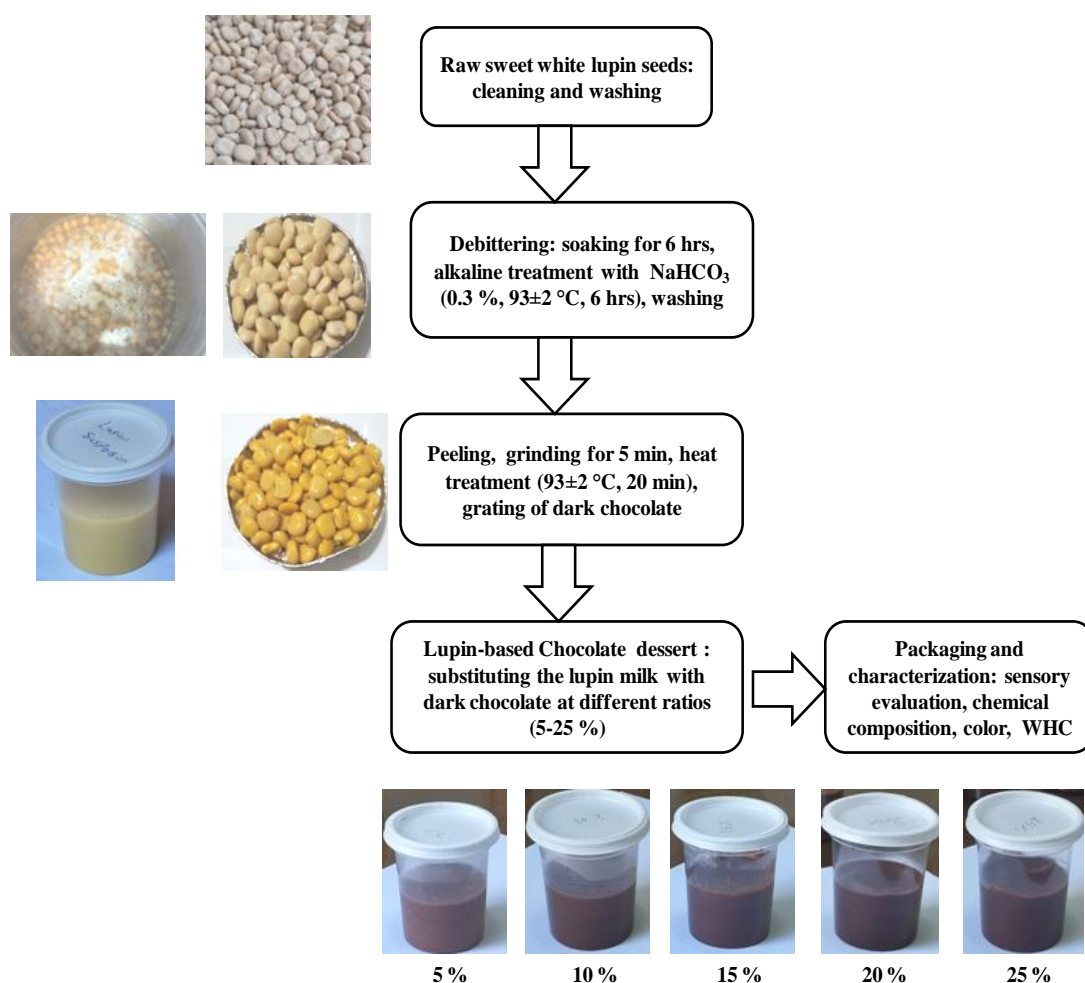


Fig. 1. Preparation protocol of lupin milk and lupin-based chocolate dessert formulations

Color quantification

The color of lupin-based chocolate dessert formulations and the commercial chocolate dessert (Danette) was determined according to **Mounir *et al.* (2021)**, using a Hunter's Lab color analyzer (Color Flex EZ Spectrophotometer, USA) which was calibrated with a black and white standard tile using the L^* a^* b^* scale before measurement. Samples were placed in the standard cup and the color values of sample were recorded as L^* , a^* , and b^* .

Chroma (C^*) and total color change (ΔE) were calculated according to **Vega-Gálvez *et al.* (2012)** using Eq. (2) and (3), respectively:

$$C^* = \sqrt{a^{*2} + b^{*2}} \quad (2)$$

$$\Delta E = \sqrt{\Delta L^{*2} + \Delta a^{*2} + \Delta b^{*2}} \quad (3)$$

Browning index (BI) was determined according to **Shittu *et al.* (2007)** using Eq. (4):

$$BI = \frac{100(x-0.31)}{0.17} \quad (4)$$

$$\text{Where: } x = \frac{a^* + 1.75 L^*}{5.645 L^* + a^* - 3.012 b}$$

Water holding capacity

The water holding capacity was determined according to **Mounir (2015)**, for lupin-based chocolate dessert formulations and commercial chocolate dessert (Danette), but with slight modifications. Briefly, a sample of 15 g was centrifuged at $960 \times g$ for 30 min (centrifuge Model 3-30K, Rotor (12156), SIGMA, Germany). The supernatant was carefully removed by decanting the sample and weighted. The WHC was calculated using Eq. (5):

$$\text{WHC (\%)} = \frac{\text{mass before centrifuge} - \text{mass after centrifuge}}{\text{mass before centrifuge}} \times 100 \quad (5)$$

Statistical Analysis

The obtained results were presented as mean value \pm standard deviation of triplicates. Analysis of variance (ANOVA) test was performed using CO-STAT program (Co-Stat version 6.311, USA) in order to identify the significant differences between the response parameters at ($p \leq 0.05$) confidence level. In addition, a correlation matrix was performed to define the relationship between the substitution ratio and the response variables, and each to other.

RESULTS AND DISCUSSION

Sensory Characteristics

Table 1 shows the sensory characteristics of lupin-based chocolate dessert formulations and the commercial chocolate dessert (Danette). The different formulations of lupin-based chocolate dessert and the commercial chocolate dessert (Danette) were evaluated for their overall acceptability based on the appearance, color, taste, odor, and texture. The overall acceptability of lupin-based dessert formulations varied from 7.14 to 8.93 and was significantly depended on the substitution ratio, where a significant gradual increase was observed with an increase in the substitution ratio. Lupin-based chocolate dessert formulations prepared at 20 and 25%, as substitution ratios, showed a higher acceptability compared to the other formulations and to the commercial chocolate dessert (Danette), with an increase in the acceptability by about 11 and 13%, respectively, compared to the commercial chocolate dessert (Danette). This may be due to 1) the strong chocolate flavor, where **Carvajal-Larenas (2019)** reported that lupin milk had good sensory characteristics but only when it was flavored, 2) the high content of fat and fiber which contributed to improving the quality of eating, palatability, and texture (creamy texture), as a result of the substitution of lupin milk with dark chocolate (**Drewnowski, 1997**). The consistency of lupin-based chocolate dessert formulations is another supporting explanation. This consistency and its stability are based on

the effect of total solids in the end product (**Carvajal-Larenas, 2019**); where the protein content was cited as the most important feature to reach a good consistency (**Akalin et al., 2012**). The total solids of these formulations (T4 and T5) were higher than that of the commercial chocolate dessert (Danette), being 24.80, 27.60, and 24.47%, respectively (Table 2).

Chemical Characteristics

Proximate chemical composition

The chemical composition of lupin milk, lupin-based chocolate dessert formulations, and the commercial chocolate dessert (Danette) is shown in Table 2. The lupin milk had 7.10% protein, 1.25% fat, 0.98% fiber, 0.87% ash, 3.99% carbohydrates, and 86.80% moisture. These findings were in the range of previous reported results (**Asres et al., 2022; Struti et al., 2020**). The chemical composition of lupin-based chocolate dessert formulations depended on the substitution ratio, where the content of individual components in analyzed lupin-based chocolate dessert formulations showed large differences in the chemical composition; 5.11-6.82% protein, 2.26-4.90 % fat, 1.53-3.67% fiber, 1.41- 4.75% ash, 5.25-12.52% carbohydrates, and 72.40-84.26% moisture. The lupin-based chocolate dessert formulations had higher contents of protein, fat, fiber, and ash than that of the commercial chocolate dessert (Danette), where the later had 3.13% protein, 1.27% fat, 2.26% fiber, and 1.43% ash (Table 2). A significant gradual increase in the contents of fat, fiber, ash, and carbohydrates was observed with the increase in the substitution ratio. While, the moisture content and protein showed a decreasing tendency with increasing the substitution ratio. It is worth mentioning that the carbohydrates content in lupin-based chocolate dessert was greatly less than that of the commercial chocolate dessert, although there was a gradual increase of the content in the formulations studied. The increase in the contents of fat, fiber, and ash was estimated by about 286, 62, and 232%, respectively for lupin-based chocolate dessert formulation prepared at 25%, as a substitution ratio, compared to the commercial chocolate dessert (Danone). The increase in the contents of carbohydrates, fat, fiber, and ash could be explained by the fact that

Table 1. Sensory characteristics of lupin-based chocolate dessert formulations

Sample	Appearance	Color	Taste	Odor	Texture	Acceptability
T ₁	7.13±0.27 ^f	7.11±0.25 ^f	6.95±0.16 ^f	7.12±0.24 ^e	7.10±0.16 ^f	7.14±0.18 ^f
T ₂	7.32±0.12 ^e	7.28±0.16 ^e	7.23±0.13 ^e	7.27±0.17 ^d	7.21±0.14 ^e	7.32±0.16 ^e
T ₃	7.60±0.11 ^d	7.54±0.11 ^d	7.60±0.12 ^d	7.36±0.12 ^{cd}	7.38±0.15 ^d	7.72±0.12 ^d
T ₄	8.41±0.10 ^b	8.45±0.11 ^b	8.57±0.10 ^b	8.60±0.11 ^b	8.65±0.14 ^b	8.71±0.10 ^b
T ₅	8.90±0.14 ^a	8.91±0.12 ^a	8.93±0.11 ^a	8.85±0.15 ^a	8.90±0.13 ^a	8.93±0.11 ^a
Danette	7.71±0.10 ^c	7.73±0.11 ^c	7.76±0.14 ^c	7.40±0.13 ^c	8.11±0.12 ^c	7.86±0.15 ^c

T₁: lupin-based chocolate dessert prepared from lupin milk substituting with 5 % Dreem dark cooking chocolate; T₂: lupin-based chocolate dessert prepared from lupin milk substituting with 10 % Dreem dark cooking chocolate; T₃: lupin-based chocolate dessert prepared from lupin milk substituting with 15 % Dreem dark cooking chocolate; T₄: lupin-based chocolate dessert prepared from lupin milk substituting with 20 % Dreem dark cooking chocolate; T₅: lupin-based chocolate dessert prepared from lupin milk substituting with 25 % Dreem dark cooking chocolate; Danette: commercial chocolate dessert manufactured by Danone Company, Egypt.

Superscript letters are statistically significantly different ($P \leq 0.05$).

Table 2. Chemical composition of lupin milk, lupin-based chocolate dessert formulations, and commercial chocolate dessert (Danette)

Sample	Crude Protein	Fat	Crude Fiber	Ash	Carb.	W	TS
L. milk	7.10±0.07 ^a	1.25±0.08 ^f	0.98±0.07 ^g	0.87±0.03 ^f	3.99±0.18 ^g	86.80±0.12 ^a	13.20±0.12 ^g
T ₁	6.82±0.08 ^b	2.26±0.10 ^e	1.53±0.08 ^f	1.41±0.06 ^e	5.25±0.13 ^f	84.26±0.10 ^b	15.74±0.10 ^f
T ₂	6.53±0.06 ^c	2.92±0.08 ^d	2.07±0.06 ^e	2.23±0.09 ^d	6.82±0.19 ^e	81.50±0.12 ^c	18.50±0.12 ^e
T ₃	6.04±0.07 ^d	3.86±0.09 ^c	2.65±0.09 ^c	3.21±0.07 ^c	8.65±0.18 ^d	78.23±0.13 ^d	21.77±0.13 ^d
T ₄	5.65±0.06 ^e	4.46±0.08 ^b	3.14±0.06 ^b	4.17±0.06 ^b	10.53±0.14 ^c	75.20±0.11 ^f	24.80±0.11 ^b
T ₅	5.11±0.08 ^f	4.90±0.07 ^a	3.67±0.08 ^a	4.75±0.09 ^a	12.52±0.13 ^b	72.40±0.10 ^g	27.60±0.10 ^a
Danette	3.13±0.09 ^g	1.27±0.08 ^f	2.26±0.07 ^d	1.43±0.05 ^e	18.52±0.09 ^a	75.33±0.13 ^e	24.47±0.13 ^c

L. milk: lupin milk; Carb: carbohydrates; T₁: lupin-based chocolate dessert prepared from lupin milk substituting with 5 % Dreem dark cooking chocolate; T₂: lupin-based chocolate dessert prepared from lupin milk substituting with 10 % Dreem dark cooking chocolate; T₃: lupin-based chocolate dessert prepared from lupin milk substituting with 15 % Dreem dark cooking chocolate; T₄: lupin-based chocolate dessert prepared from lupin milk substituting with 20 % Dreem dark cooking chocolate; T₅: lupin-based chocolate dessert with lupin milk substituting with 25 % Dreem dark cooking chocolate, Danette: commercial chocolate dessert manufactured by Danone Company, Egypt; W: moisture content (% wb); TS: total solids (%).

Superscript letters are statistically significantly different ($P \leq 0.05$).

the dark chocolate is rich in carbohydrates (> 50%), fat (> 34%), fiber (>13%), and minerals such as potassium, magnesium, and iron (0.9-1.5% ash) (Melo *et al.*, 2020; Cinquanta *et al.*, 2016), that in turn contributes to increasing the contents of fat, fiber, and ash. For lupin-based chocolate dessert formulations, since carbohydrates were calculated by difference (Eq.1), a low

protein and moisture content will give a high content of carbohydrates. Whereas, the decrease in protein content may be due to the lower protein content of dark chocolate than that of the lupin milk, where the dark chocolate contains about 6.4% protein (Torres-Moreno *et al.*, 2012). Despite the lower level of protein, the lupin-based chocolate dessert formulations still

have higher content than that of commercial dessert (Danette). Contrary, these formulations had lower carbohydrates than the commercial dessert, despite the increasing trend of carbohydrate with increasing the substitution ratio. The obtained results demonstrated that lupin-based chocolate dessert formulations have higher nutritional value in comparison with the commercial chocolate dessert (Danette), due to the increased content of protein, fat, fiber, and ash.

Major and trace elements

The major and trace elements are essential for the biological processes and the proper functioning of any living organism. Their supply depends on the bioavailability that is directly related to the composition of food (Jácimović *et al.*, 2022). Many studies have demonstrated that cocoa is a comprehensive source of many essential elements. Therefore, chocolate has the potential to provide a significant concentration of minerals in the human diet (Karas *et al.*, 2021; Oliveira *et al.*, 2021; Grassia *et al.*, 2019). The minerals content of lupinmilk, lupin-based chocolate dessert formulations, and commercial chocolate dessert (Danette) is shown in Table 3. The obtained results show that the lupin milk was rich in phosphorus (3325 mg/kg) followed by calcium (1875 mg/kg), potassium (1630 mg/kg), and magnesium (1161 mg/kg), while iron (17.14 mg/Kg) showed a lower content followed by zinc (13.86 mg/kg). The obtained results are in line with the literature (Ertaş and Bilgiçli, 2014). However, the lupin-based chocolate dessert formulations were rich in phosphorus and potassium followed by calcium and magnesium. The minerals content of lupin-based chocolate dessert formulations showed a wide range depending on the substitution ratio; P: 3463-4025 mg/kg, Ca: 1450-1781 mg/kg, K: 1719-1953 mg/kg, Mg: 1276-1536 mg/kg, Fe: 32.43-93.23 mg/kg and Zn: 18.51-26.32 mg/kg. The commercial dessert (Danette) was rich in calcium and potassium followed by phosphorus, magnesium, iron, and zinc, but with a lower content in comparison with the lupin milk and lupin-based chocolate dessert formulations; P: 856 mg/kg, Ca: 1238 mg/kg, K: 1146 mg/kg, Mg: 137mg/kg, Fe: 28.71 mg/kg, and Zn: 3.62 mg/kg. It was observed that the minerals content in the lupin-

based chocolate dessert formulations increased, except for Ca, with the increase in the substitution ratio. This behavior could be explained by the fact that the dark chocolate is rich in P (2324 mg/kg), K (3651 mg/kg), Mg (1564 mg/kg), Fe (479 mg/kg), and Zn (105 mg/kg) (Jácimović *et al.*, 2022; Karas *et al.*, 2021; Grassia *et al.*, 2019), and this content was higher than that of the lupin milk. Since the Ca content of the dark chocolate was about 908 mg/kg (Jácimović *et al.*, 2022) which was lower than that of the lupin milk, therefore the content of Ca in the lupin-based chocolate dessert formulations decreased with increasing the substitution ratio. It is worthwhile mentioning that the content of Ca in the lupin-based chocolate dessert formulations was higher than that of the commercial dessert (Danette), despite the decreased level of Ca with increasing the substitution ratio with dark chocolate. The lowest Ca content was observed for the formulation prepared at 25% as a substitution ratio, with an increase of about 17% compared to the commercial dessert (Danette).

pH value

Food is a complex matrix of biological and chemical ingredients where the unstable interactions between these ingredients may affect its quality and safety. Since pH influences two major attributes of food safety, quality, and consistency, it is important to measure the pH of food for consumer health and acceptability. Table 3 shows the pH value of lupin milk, lupin-based chocolate dessert formulations, and the commercial dessert (Danette). The pH of lupin milk was 7.30 while that of commercial dessert was found to be 6.36. The lower pH of commercial dessert, compared to the lupin milk, may be due to the presence of milk and other ingredients which have a low pH. On the other hand, some major differences are shown in the pH value of lupin-based chocolate dessert formulations, ranging from 7.54 to 8.44. Moreover, the pH of lupin-based chocolate dessert showed an increasing trend with increasing the substitution ratio with dark chocolate. The high pH of these formulations may be due to the combined effect of the high pH of both lupin milk (pH: 7.30) and dark chocolate (pH: 9.4).

Table 3. Minerals content (mg/Kg) and pH of lupin milk, lupin-based chocolate dessert formulations, and commercial chocolate dessert (Danette)

Sample	P	Ca	K	Mg	Fe	Zn	pH
Lupin milk	3325±20 ^f	1875±30 ^a	1630±21 ^f	1161±18 ^f	17.14±0.11 ^g	13.86±0.12 ^f	7.30±0.03 ^f
T ₁	3463±18 ^c	1781±28 ^b	1719±0.18 ^e	1276±16 ^e	32.43±0.15 ^e	18.51±0.13 ^e	7.54±0.03 ^e
T ₂	3502±20 ^d	1687±26 ^c	1753±16 ^d	1309±14 ^d	56.25±0.17 ^d	19.73±0.14 ^d	7.72±0.04 ^d
T ₃	3646±22 ^c	1570±23 ^d	1819±15 ^c	1435±16 ^c	74.71±0.13 ^c	23.35±0.13 ^c	8.08±0.03 ^c
T ₄	3764±21 ^b	1542±23 ^d	1921±17 ^b	1491±17 ^b	86.34±0.14 ^b	25.16±0.14 ^b	8.13±0.04 ^b
T ₅	4025±22 ^a	1450±21 ^e	1953±15 ^a	1536±15 ^a	93.23±0.15 ^a	26.32±0.16 ^a	8.44±0.03 ^a
Danette	856±6.24 ^g	1238±24 ^f	1146±16 ^g	137±3.61 ^g	28.71±0.11 ^f	3.62±0.07 ^g	6.36±0.04 ^g

T₁: lupin-based chocolate dessert prepared from lupin milk substituting with 5% Dreem dark cooking chocolate; T₂: lupin-based chocolate dessert prepared from lupin milk substituting with 10% Dreem dark cooking chocolate; T₃: lupin-based chocolate dessert prepared from lupin milk substituting with 15% Dreem dark cooking chocolate; T₄: lupin-based chocolate dessert prepared from lupin milk substituting with 20% Dreem dark cooking chocolate; T₅: lupin-based chocolate dessert with lupin milk substituting with 25% Dreem dark cooking chocolate, Danette: commercial chocolate dessert manufactured by Danone Company, Egypt.

Superscript letters are statistically significantly different ($P \leq 0.05$).

Color Characteristics

Color is an important visual quality criterion of foods which may affect sensory perception and consumer acceptance because the color is related to the texture and sensory and nutritive attributes of foods (Pieniazek and Messina, 2017). The human eye cannot identify the slight differences in color. Even if two colors seem very identical, there may be considerable variations when evaluated with an instrumental color analyzer. The color parameters of lupin milk, lupin-based chocolate dessert formulations, and the commercial chocolate dessert (Danette) are shown in Table 4. The values of L^* , a^* , and b^* of lupin milk were 71.3, -3.0, and 29.3, respectively, indicating the greenish-yellow color. Besides, the lupin milk had a high color intensity and low browning index; 29.5 and 0.97, respectively, which may be due to the high values of both L^* and b^* , and the low value of a^* . The obtained color parameters are in the line with the reported results in the literature (Ertaş and Bilgiçli, 2014). On the other hand, some major differences are shown in color parameters, L^* , a^* , and b^* of lupin-based chocolate dessert formulations, where the individual parameters were found in the range of 26.6-37.5, 5.5-6.9, and b^* : 5.1-9.3, respectively, indicating the significant effect of the substitution ratio on

these parameters. It was observed that the values of L^* and b^* decreased, while the value of a^* increased, with the increase in the substitution ratio, reflecting the increased color darkness as a result of the substitution with dark chocolate that have a direct effect on the color parameters (particularly the color lightness). The dark chocolate had low values of L^* (15.4) and b^* (4.6) (Table 4). Similarly, the color intensity, the total color change, and the browning index showed a wide range of difference depending on the substitution ratio; C^* : 8.6-10.8, ΔE : 40.3-51.7, and BI: 12.7-19.8. The decrease in the color intensity (C^*) may be due to the decreased value of b^* ; lower color yellowness (Mounir et al., 2019), while the increase in the browning index (BI) could be explained by the combined effect of adding dark chocolate with a dense brownish color (BI: 30.56), and the pH influence. The browning index depends on the pH value where the Millard reaction increases at higher values of pH (Karseno et al., 2018). In addition, the browning index is related to the color parameters; L^* , and a^* , because it was calculated according to the Eq. (4), therefore the lower the value of L^* and the higher the value of a^* , the higher the browning index. Photo (1) shows the color variation between the lupin milk, lupin-based formulations, and the commercial chocolate dessert (Danette).

Table 4. Color characteristics and water holding capacity of lupin-based chocolate dessert formulations and commercial chocolate dessert (Danette)

Sample	L*	a*	b*	C*	ΔE	BI	WHC
Chocolate	15.4±0.06	6.3±0.07	4.6±0.06	7.8±0.09	ND	30.6±0.21	ND
Lupin milk	71.3±0.11 ^a	-3.0±0.09 ^g	29.3±0.09 ^a	29.5±0.09 ^a	0.00 ^f	0.97±0.09 ^g	34.8±0.11 ^a
T1	37.5±0.09 ^b	5.5±0.06 ^f	9.3±0.07 ^b	10.8±0.04 ^b	40.3±0.12 ^e	12.7±0.07 ^f	28.4±0.13 ^b
T2	33.0±0.07 ^c	5.7±0.07 ^e	7.9±0.08 ^c	9.7±0.07 ^c	44.8±0.17 ^d	14.5±0.16 ^e	27.5±0.12 ^c
T3	29.6±0.08 ^e	6.1±0.06 ^d	7.4±0.04 ^d	9.5±0.07 ^d	48.0±0.13 ^c	16.8±0.12 ^c	20.8±0.12 ^d
T4	27.8±0.09 ^f	6.4±0.04 ^b	6.1±0.05 ^f	8.8±0.04 ^f	50.2±0.18 ^b	18.2±0.08 ^b	13.9±0.09 ^e
T5	26.6±0.09 ^g	6.9±0.06 ^a	5.1±0.07 ^g	8.6±0.08 ^g	51.7±0.18 ^a	19.8±0.10 ^a	8.5±0.09 ^f
Danette	30.6±0.06 ^d	6.3±0.06 ^c	6.7±0.04 ^e	9.2±0.07 ^e	ND	16.2±0.10 ^d	4.8±0.08 ^g

Chocolate: dark chocolate (Dreem dark cooking chocolate); T₁: lupin-based chocolate dessert prepared from lupin milk substituting with 5 % Dreem dark cooking chocolate; T₂: lupin-based chocolate dessert prepared from lupin milk substituting with 10 % Dreem dark cooking chocolate; T₃: lupin-based chocolate dessert prepared from lupin milk substituting with 15 % Dreem dark cooking chocolate; T₄: lupin-based chocolate dessert prepared from lupin milk substituting with 20 % Dreem dark cooking chocolate; T₅: lupin-based chocolate dessert with lupin milk substituting with 25 % Dreem dark cooking chocolate, Danette: commercial chocolate dessert manufactured by Danone Company, Egypt.

ND: None determined.

Superscript letters are statistically significantly different ($P \leq 0.05$)

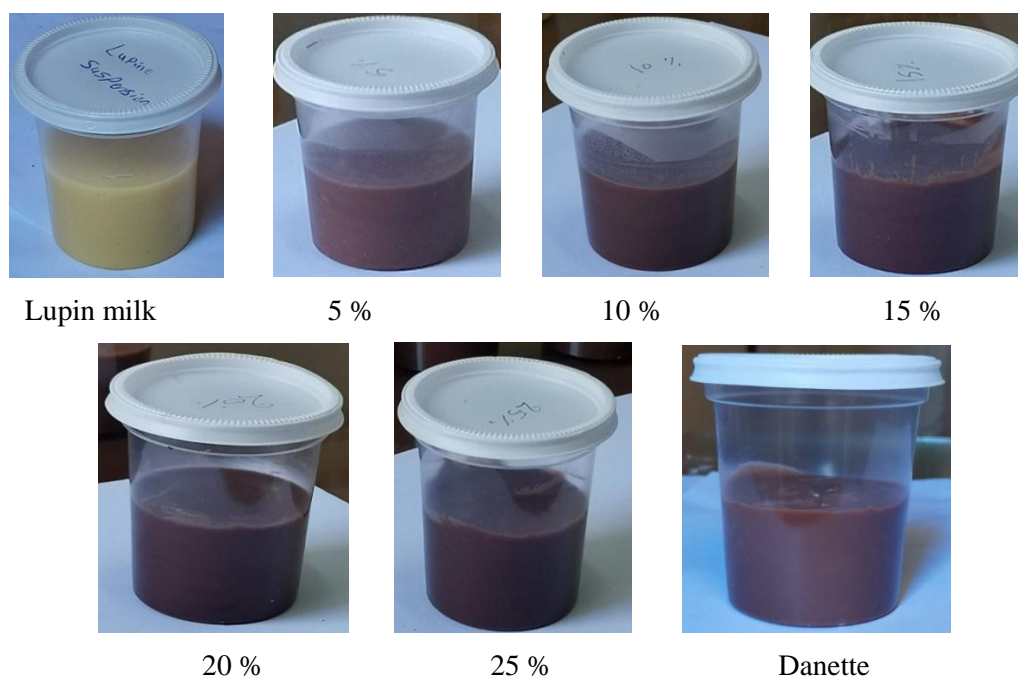


Photo 1. Photo of lupin milk, lupin-based chocolate dessert formulations, and commercial dessert (Danette), which illustrates the color variation

Water Holding Capacity

The water holding capacity (WHC) of foods can be defined as the ability to hold its own and added water against external forces such as pressing, centrifugation, or heating. Water retention is the water adsorbed or retained by a wet or dry mixture of components such as protein or starch (Zayas, 1997). The functionality of proteins is related to their interaction with water, where the protein-water interaction define the functional properties of proteins in foods such as water binding and retention, foaming, emulsifying, solubility, viscosity, and syneresis. Therefore, water holding or retention is a limiting factor in protein food applications and plays an important role in the formation of food structure. The water holding capacity of lupin milk, lupin-based chocolate dessert formulations, and the commercial dessert (Danette) is shown in Table 4. The lupin milk showed the highest WHC with a value of 34.8%, while the lowest WHC was recorded for the commercial dessert (Danette) with a value of 4.8%, indicating the poor content of protein in the commercial dessert. On the other hands, the lupin-based chocolate dessert exhibited a wide range of WHC (8.5-28.4%) depending on the substitution ratio, where a significant gradual increase in WHC was observed with an increase in the substitution ratio with dark chocolate. The lower WHC of lupin-based chocolate dessert formulations may be due to the decreased content of protein compared to the lupin milk (Table 2). Since the water binding is attributed to the protein content of the product, the water holding capacity increases as protein content increases (Zayas, 1997). Moreover, the competition of solutes and protein for the available water is another supporting explanation; in this study some solutes such as carbohydrates including sugars and minerals such as potassium, iron, and magnesium, that came from the dark chocolate, increased in the formulations studied which in turn increase the competition between these solutes and protein for the available water leading to decreasing of the water holding capacity (Zayas, 1997).

Correlation Matrix

A correlation matrix was performed to define the relationship between the substitution ratio

and the response variables. These correlations showed the significant effect of the substitution ratio on the response variables studied (Table 5). Some correlations were observed between the substitution ratio and the response variables, and between the response variables each to other as follows: 1) a strong positive correlation was observed between the substitution ratio and the fat content, indicating that the fat which came from the dark chocolate contributed to increasing the fat level in lupin-based chocolate dessert formulations which in turn improved their sensory characteristics, 2) a strong positive correlation was observed between the substitution ratio and the total solids as a result of increased levels of carbohydrates, fat and minerals (ash) in the formulations studied, 3) a strong positive correlation was observed between the substitution ratio and the browning index (BI) which has a direct effect on the color lightness of the formulations studied, 4) a strong negative correlation was obtained between BI and Ca, while it was positively with Mg. These correlations reflect the effect of these elements on the development of Maillard reaction where Ca and Mg promoted the formation of colored Millard reaction compounds, but a blocking effect was observed in the presence of abundant amount of Ca (Ramonaityte, 2008), 5) a strong positive correlation was observed between the WHC and protein, indicating that the water holding capacity greatly depends on the concentration of protein in foods. The higher the protein content, the higher the water holding capacity, and 6) a strong negative correlation was found between the water holding capacity and the ash content; this behavior may be due to the competition effect of protein and minerals (solutes) for the available water, decreasing the water holding capacity as the ash content increased.

Conclusion

In this study, lupin milk as an alternative to cow milk was used in the preparation of chocolate dessert similar to a commercial dessert (Danette) manufactured by Danone Company in Egypt. Lupin milk was prepared by grinding the peeled seeds after a debittering process including soaking and alkaline treatment using a hot solution of NaHCO₃. The lupin-based

Table 5. Correlation matrix between the substitution ratio and the response variables, and between the response variables each to other

r	R	Protein	Fat	Fiber	Ash	Carb	TS	Ca	P	K	Mg	Fe	Zn	pH	L*	a*	b*	BI	WHC
R	1	-0.99	0.99	0.99	0.99	0.99	0.99	-0.99	0.97	0.98	0.98	0.97	0.98	0.98	-0.97	0.99	-0.99	0.99	-0.98
Protein	-0.99	1	-0.98	-0.99	-0.99	-0.99	-0.99	0.98	-0.98	-0.98	-0.98	-0.95	-0.98	-0.99	0.95	-0.99	0.98	-0.99	0.99
Fat	0.99	-0.98	1	0.99	0.99	0.98	0.99	-0.99	0.94	0.98	0.99	0.99	0.99	0.98	-0.99	0.98	-0.98	0.99	-0.97
Fiber	0.99	-0.99	0.99	1	0.99	0.99	0.99	-0.99	0.96	0.98	0.98	0.98	0.99	0.99	-0.97	0.99	-0.99	0.99	-0.98
Ash	0.99	-0.99	0.99	0.99	1	0.99	0.99	-0.98	0.95	0.99	0.99	0.98	0.99	0.98	-0.98	0.99	-0.99	0.99	-0.98
Carb	0.99	-0.99	0.98	0.99	0.99	1	0.99	-0.98	0.98	0.98	0.97	0.96	0.98	0.98	-0.95	0.99	-0.99	0.99	-0.99
TS	0.99	-0.99	0.99	0.99	0.99	0.99	1	-0.99	0.96	0.99	0.99	0.98	0.99	0.98	-0.97	0.99	-0.99	0.99	-0.98
Ca	-0.99	0.98	-0.99	-0.99	-0.98	-0.98	-0.99	1	-0.94	-0.95	-0.98	-0.98	-0.98	-0.99	0.98	-0.97	0.97	-0.99	0.95
P	0.97	-0.98	0.94	0.96	0.95	0.98	0.96	-0.94	1	0.95	0.94	0.89	0.93	0.96	-0.88	0.99	-0.95	0.95	-0.98
K	0.98	-0.98	0.98	0.98	0.99	0.98	0.99	-0.95	0.95	1	0.98	0.95	0.98	0.95	-0.94	0.98	-0.98	0.98	-0.99
Mg	0.98	-0.98	0.99	0.98	0.99	0.97	0.99	-0.98	0.94	0.98	1	0.97	0.99	0.98	-0.96	0.97	-0.95	0.99	-0.98
Fe	0.97	-0.95	0.99	0.98	0.98	0.96	0.98	-0.98	0.89	0.95	0.97	1	0.98	0.96	-0.99	0.95	-0.97	0.98	-0.92
Zn	0.98	-0.98	0.99	0.99	0.99	0.98	0.99	-0.98	0.93	0.98	0.99	0.98	1	0.98	-0.97	0.97	-0.96	0.99	-0.97
pH	0.98	-0.99	0.98	0.99	0.98	0.98	0.98	-0.99	0.96	0.95	0.98	0.96	0.98	1	-0.96	0.98	-0.96	0.99	-0.96
L*	-0.97	0.95	-0.99	-0.97	-0.98	-0.95	-0.97	0.98	-0.88	-0.94	-0.96	-0.99	-0.97	-0.96	1	-0.94	0.96	-0.98	0.91
a*	0.99	-0.99	0.98	0.99	0.99	0.99	0.99	-0.97	0.99	0.98	0.97	0.95	0.97	0.98	-0.94	1	-0.98	0.99	-0.99
b*	-0.99	0.98	-0.98	-0.99	-0.99	-0.98	-0.99	0.97	-0.95	-0.98	-0.95	-0.97	-0.96	-0.96	0.96	-0.98	1	-0.98	0.96
BI	0.99	-0.99	0.99	0.99	0.99	0.99	0.99	-0.99	0.95	0.98	0.99	0.98	0.99	0.99	-0.98	0.99	-0.98	1	-0.97
WHC	-0.98	0.99	-0.97	-0.98	-0.98	-0.99	-0.98	0.95	-0.98	-0.99	-0.98	-0.92	-0.97	-0.96	0.91	-0.99	0.96	-0.97	1

r: correlation coefficient; R: substitution ratio, Carb: carbohydrates; L*: color lightness; a*: -a* (greenness), +a* (redness), b*: -b* (blueness), +b* (yellowness); BI: browning index; WHC: water holding capacity (%).

formulations were prepared by substituting the lupin milk with dark chocolate at different ratios (5-25%). The final formulations were evaluated in terms of sensory, physico-chemical characteristics and water holding capacity. The obtained results showed the significant effect of the substitution ratio on the studied characteristics. The sensory, physicochemical, and functional properties were greatly improved compared to the commercial dessert. Using lupin milk as an alternative milk in the production of chocolate dessert is a promising technique due to its important role in increasing the chemical characteristics of chocolate dessert, which could direct its extensive applications in the food industrial sectors. On the other hand, using dark chocolate as a partial substitute for the lupin milk contributed to increasing the minerals content and improving the sensory characteristics of lupin-based chocolate dessert.

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الخصائص الحسية والفيزيوكيميائية والوظيفية لتركيبات حلوى الشوكولاتة المحضرة من بذور الترمس الأبيض الحلو المصري

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كان الهدف من هذه الدراسة هو تقييم الخصائص الحسية والفيزيوكيميائية والوظيفية لتركيبات جديدة من حلوى الشوكولاتة القائمة على الترمس. تم تحضير حليب الترمس عن طريق طحن البذور المقشرة بعد عملية إزالة المرارة و التي تضمنت نقع البذور لمدة 6 ساعات والمعالجة القلوية بمحلول ساخن من بيكربونات الصوديوم (0.3% ، 93 °م) لمدة 6 ساعات. تم تحضير خمس تركيبات من حلوى الشوكولاتة القائمة على الترمس باستبدال حليب الترمس بالشوكولاتة الداكنة بنسب مختلفة تتراوح من 5 إلى 25% مع زيادة فاصلة قدرها 5%. أظهرت النتائج التي تم الحصول عليها التأثير المعنوي لنسبة الاستبدال على الخواص الحسية والفيزيوكيميائية والوظيفية لحلوى الشوكولاتة القائمة على الترمس. فقد لوحظ تحسن تدريجي كبير في الخصائص الحسية بزيادة نسبة الاستبدال. كما لوحظ أيضاً زيادة تدريجية كبيرة في التركيب الكيميائي بزيادة نسبة الاستبدال ، باستثناء البروتين والكالسيوم. على العكس من ذلك، أظهرت قدرة الاحتفاظ بالماء اتجاهاً متناقصاً بزيادة نسبة الاستبدال. يعد استخدام حليب الترمس كبديل لحليب البقر والشوكولاتة الداكنة كبديل جزئي لحليب الترمس في تحضير حلوى الشوكولاتة تقنية واعدة نظراً لزيادة مستوى البروتين والدهون والألياف والرماد في حلوى الشوكولاتة القائمة على الترمس مقارنةً بالحلوى التجارية (دانيت).

الكلمات الإسترشادية: بذور الترمس الأبيض الحلو، حلوى الشوكولاتة ، الخصائص الحسية، الخواص الكيميائية، اللون، القدرة على الاحتفاظ بالماء.

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