Response Surface Optimization of Cupcake Formula Fortified with Date Seed Powder

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ABSTRACT: *Recently the demand for dietary fiber-enriched foods has increased due to consumers' interest in the nutritional and health benefits of functional foods. Date Seed Powder (DSP) is rich in dietary fiber and other nutrients such as minerals, antioxidants, unsaturated fatty acids, amino acids, and tocopherols. It is applicable as a cheap and valuable source to produce functional foods. In this study, DSP was added to the cupcake formulation at 5 levels (0.0, 2.5, 5.0, 7.5, and 10.0%), and the effect of this substitution on the physical, chemical, microbial, and sensory properties of the final product was investigated during 28 days of storage at 4 °C. Based on the results, adding DSP to the cake formula significantly increased the amount of fat and fiber of the cake. Substituting wheat flour with DSP increased the total phenolic compounds and moisture content of enriched cakes compared to the control. Furthermore, the peroxide, para-anisidine, and stiffness of fortified cakes were more than the control at all storage times. Adding DSP to the cake reduced the specific volume, L* and b* of crust and crumb. The highest overall acceptance of fortified cakes with DSP was 2.5% DSP. According to the response surface models, the optimal quality of cakes was observed at the DSP ratio of 3.6 % and 0 days after preparation. Results demonstrated that DSP could be used as a dietary fiber supplement for cake enrichment.*

KEYWORDS: *Phoenix dactylifera L.; Date seed powder; Fiber; Quality characteristics.*

INTRODUCTION

The health-promoting role of foods containing bioactive compounds has been proven. In line with the increasing demand for these products, the food industry is trying to allocate a larger portion of products to this category of foods. Fruit seeds are by-products of fruit processing. Their flours contain various nutrients and have many beneficial health effects and a high potential for use as new functional components for fortified baked products [1]. The effects of substitution of wheat flour with apricot, cherry, pomegranate, and pumpkin seeds flour on the dough rheology, as well as the physicochemical properties, and shelf-life stability of cake products, were investigated by *Ağirbaş et al.* [1]. In previous research, the effect of adding pumpkin seed flour on the quality properties of cookies [2], muffins [3], and gluten-free cakes [4] have been studied. The pomegranate seed flour was also used to enrich the cake [5, 6].

Phoenix dactylifera L., also known as the date palm,

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is a native fruit from Southwest Asia and North Africa [7]. Date palm fruit has special nutritional and therapeutic values [8]. The date fruit consists of a seed and a fleshy pericarp [7]. Date seed is considered a waste of many date processing plants. Date seeds contain 3.1–7.1% moisture, 2.3–6.4% protein, 5.0–13.2 fat, 0.9–1.8% ash and 22.5–80.2% dietary fiber [9]. In addition, it contains plenty of bioactive compounds and encompasses therapeutic effects against various diseases. Flavonoids, phenolic acids, sterols, anthocyanins, procyanidins, and tocopherols are the phytochemicals of a date seed. Hesperidin is the major flavonoid (17.27 mg/100 g) of date seed. It has some health benefits such as reducing the risk of cancer and preventing atherosclerosis and osteoporosis. The phenolic acids of date seed include protocatechuic, hydroxybenzoic, coumaric, ferulic, and caffeic acids [7]. In a study, α-tocotrienol, γ-tocopherol, γ-tocotrienol, δ-tocopherol, β-tocopherol, and α-tocopherol were measured as the major tocols in date seed oil. Due to the low content of polyunsaturated fatty acids and high content of phenolic compounds in date seed oil, this oil has higher oxidative stability than many fruits and vegetable oils [10]. Sodium (Na), potassium (K), magnesium (Mg), calcium (Ca), iron (Fe), manganese (Mn), zinc (Zn), cuprum (Cu), nickel (Ni), cobalt (Co), and cadmium are some notorious minerals of date seed [11]. The date seed is a rich source of phytohormones and it has anti-aging properties, reducing skin wrinkles [12]. As noted, date seed is a good source of dietary fiber. Water-soluble polysaccharides, hemicellulose, cellulose and lignin are the major dietary fibers in date seeds [13]. In addition to physiological benefits, increasing viscosity, texture modification, inhibiting of crystallization, and increasing oil and water holding capacity and emulsion stability are the positive technological effects of dietary fiber that led to abundant applications of these components in food industries [14]. Some researchers have used date seeds to produce various products such as citric acid [15] and jam [16]. *Almana* and *Mahmaud* [17] evaluated date seed as a source of alternative dietary fibers compared to wheat bran. *Basuny* and *Al-Marzooq* [11] replaced conventional oils with date seed oil in the production of mayonnaise.

High fiber content and the presence of different valuable nutrients such as phenolic components and natural antioxidants, made the date seed a good source for flour production. *Ambigaipalan* and *Shahidi* [18] substituted wheat flour with date seed flour hydrolysate and date seed flour in the muffin formula. *Shokrollahi* and *Taghizadeh* [19] and *Platat et al.* [20] used date seed flour to produce bread. Different types of cakes have high consumer acceptance for people of all ages and therefore have particular importance for transferring bioactive compounds to the human diet [21]. Wheat flour properties such as protein content, gluten quality and strength, and ash contents are the most important factors in cake production and the quality of the final product, especially final texture of the crumb and crust, volume, flavor, and the other quality attributes are affected by flour characteristics [22]. Finding the best ratios of wheat and date flours has industrial importance in producing date seed fortified cake. Response surface is a statistical technique for finding the optimized conditions [23]. Therefore, the aim of this study was to investigate the effects of substituting different ratios of wheat flour by DSP on some quality attributes of the final product using the response surface technique.

EXPERIMENTAL SECTION

The Date Seed Powder (DSP) was produced by grinding date fruit seeds after washing and toasting seeds at 210 °C for 70 minutes and then sieving. The toasting was performed to improve the color, creating a coffee-like flavor without increasing caffeine level, and increasing the bioavailability of its complex beneficial compounds. Wheat flour was purchased from Kamran Fars flour mill factory (Shiraz, Fars province, Iran). Other ingredients, including sugar, oil, egg, and baking powder, were supplied from Kerman markets. Table 1 shows the percentage of ingredients used in cake batter formulations containing different levels of DSP.

For cupcake batter preparation, the whole egg and sugar were initially mixed. Then, liquid ingredients (water, oil, and invert syrup) were added to the mixture, and finally, dry ingredients (wheat flour, DSP, and baking powder) were added simultaneously to the cake batter. Cake batter after preparation was poured on wax papers in special moulds designed for cupcakes and transferred to an industrial rotary oven (MBICO, F 2100 ST SI, Iran) and cooked at 180 °C for 32 minutes. After cooking, cakes were cooled at room temperature for about one hour and then packaged in zip-kip polyethylene bags (20×25 cm) and stored at the refrigerator (4±1°C) until performing tests. DSP was substituted wheat flour in levels of 0.0, 2.5, 5.0, 7.5, and 10.0%. The cakes were evaluated in terms

Table 1: Weight percent of ingredients in cake batter formulations containing different levels of date seed powder (DSP).

Ingredient (%)	Percent of DSP in cake batter formulations				
	0.0% DSP	2.5% DSP	5.0% DSP	7.5% DSP	10.0% DSP
Flour	23.50	22.91	22.32	21.74	21.15
DSP	0.00	0.59	1.18	1.76	2.35
Sugar	18.00	18.00	18.00	18.00	18.00
Oil	18.00	18.00	18.00	18.00	18.00
Egg	18.00	18.00	18.00	18.00	18.00
Water	19.50	19.50	19.50	19.50	19.50
Baking powder	1.00	1.00	1.00	1.00	1.00
Invert syrup	2.00	2.00	2.00	2.00	2.00

of physicochemical, microbial, and sensorial properties at 0, 7, 14, 21, and 28 days of storage at chill temperature.

Chemical analysis

The DSP, wheat flour, and cakes were analyzed for moisture content, total lipid, protein, total fiber, and pH value. The moisture of samples was determined by oven drying at 105±1 °C until reaching constant weight. Total lipid, protein, and total fiber contents were measured according to AACC methods No. 30-25.01, 46-12.01, and 32-05.01, respectively [24]. The pH value of flour, DSP, and cakes was measured using a pH meter (3020, Jenway, UK) at 23±1 °C. The gluten content of wheat flour and total phenolic content of cakes were measured by AACC method 38-12 [25] and Folin-Ciocalteu method [26], respectively. The fat compounds of cake samples were extracted using n-hexane at room temperature. The stability of cakes lipids, was determined based on the measurement of peroxide value (PV) and p-anisidine value (p-AV) according to the AOCS standard methods Cd 8-53 and Cd 18-90, respectively [27].

Specific volume

Cake volume was measured by the rapeseed displacement method [28], and then the specific volume was calculated by dividing the volume by the mass.

Texture analysis of cake

The texture profile analysis of the cakes was performed using a texture analyzer (CT-3 10kg, Brookfield, USA). For stiffness measurement, the cake samples were cut into rectangular cubes (with dimensions of 40 × 40 × 25 mm^3), placed on the flat stage, and compressed using a plastic cylindrical probe (50 mm diameter) at the speed of 1.5 mm/s and a distance of 10 mm.

Color measurement

The crust and crumb color factors were measured using a HunterLab colorimeter (TES-135A, Taiwan) standardized with a black and white ceramic plate. The samples were scanned at three different locations, and mean values of L^* (lightness/darkness), a^* (redness/greenness), and b^* (yellowness/blueness) were recorded.

Microbial analysis

Total counts of cake samples were determined after 0, 7, 14, 21, and 28 days of storage at the refrigerator (4±1°C). For this purpose, 10 g of cake sample was diluted in 90 mL peptone water (pH = 7.2 ± 0.2), and a ten-fold serial dilution in the same diluent was performed till 10^{-4}. For standard plate counts, 1 mL of each dilution was mixed with plate count agar (PCA) as culture media (pour-plating method). The plates were incubated at 30 °C for 48 h [29]. For yeast and mold counts, surface-plating by spreading 0.1 mL of each dilution on culture media (YGC agar) and incubating at 25°C for up to 5 days was accomplished [30]. Plating carried out in duplicate.

Sensory analysis

Sensory evaluation of control and DSP enriched cakes was carried out by 36 trained panelists for 5 attributes, including crust color and appearance, crumb color, taste and flavor, texture and mouth-feel, and overall acceptability using a 5-point hedonic scale, where 1 was representing poor, and 5 was representing excellent [31].

Statistical Analysis

Experiment design was performed using the response surface methodology (RSM) and historical data design using Design Expert version 7.0 software (State-Ease, Minneapolis, USA). Five levels were considered for each factor. Coded and actual values for each factor are shown in Table 2.

The optimum levels of the experimental factors, including the DSP ratio and storage time, were found. The analysis of the chemical properties of DSP, wheat flour,

Table 2: Coded and actual values of independent variables used for historical data RSM design.

Symbol	Variables	Levels				
		-1.0	-0.5	0.0	+0.5	+1.0
A	DSP ratio (%)	0	2.50	5.00	7.50	10.00
B	Storage time (day)	0	7	14	21	28

Table 3: Chemical properties of wheat flour and date seed powder (DSP).

Property	Wheat flour	DSP
Protein (%)	11.06±0.47[a]	5.64±0.08[b]
Fat (%)	3.74±0.51[b]	12.72±0.47[a]
Fiber (%)	1.33±0.13[b]	30.29±1.11[a]
Moisture (%)	12.84±0.21[a]	5.85±0.16[b]
Wet gluten (%)	28.00±1.50[a]	-
pH	5.96±0.33[a]	5.05±0.52[b]

Means values in the same row followed by the same letters are not significantly different (P<0.05).

and cake samples were performed using MSTAT-C software (Michigan, USA). Duncan's multiple-range test was used to compare the means at the 5% significance level.

RESULTS AND DISCUSSION

Chemical composition of DSP and wheat flour

The chemical analysis of wheat flour and DSP is shown in Table 3.

The results showed that wheat flour protein was 1.96 times more than DSP protein. *Al Farsi* and *Lee* [7, 32] reported that date seed contains 2.3-6.4% protein. According to the results, the fat content of DSP was significantly higher than wheat flour ($p<0.05$). The fat content of date seed was reported between 5.0-13.2% [1]. Due to its essential fatty acids as healthy compositions of edible oils, date seed oil has a high potential for producing cosmetic and pharmaceutical products [8]. In addition, the results of fiber measurement also showed a significant difference between the fiber content of DSP and wheat flour. This finding indicates that DSP is an appropriate source to supply dietary fiber for use in food formulas. The results of the study by *Al Farsi* and *lee* [32] showed that date seed dietary fiber content was 22.5-80.02%. As shown in Table 3, the moisture content of wheat flour was higher than DSP. by *Al Farsi* and *lee* [7, 32] reported that the moisture content of date seed was 3.1-7.1%. Those results are in agreement with the results of this study. In addition, the pH value of wheat flour suspension is higher than that of the DSP suspension.

Chemical characteristics of cake

Protein content

According to the results of protein measurement, the protein content of DSP-fortified cakes was lower than the control (Table 4).

However, based on the analysis of variance, this difference was not significant. As the results of wheat flour and DSP protein measurements showed, the DSP protein content was lower than that of wheat flour, so replacing wheat flour with DSP reduced the protein content of the final product. A non-significant increase in the amount of protein content of pita bread with increasing the DSP level was reported by *Platat et al*. [20].

Fat content

By replacing a part of wheat flour with DSP the fat content of the final product was increased (Table 4). However, based on the mean comparison results, the difference between the fat content of control and fortified cakes with 2.5 and 5% DSP was not significant. Based on the results of fat measurement, the fat content of the DSP was more than that of wheat flour, and that increase in fat content in the DSP fortified cakes is not unexpected. A linear increase in the fat content of date seeds fortified

Table 4: Chemical properties and specific volume of cake samples containing date seed powder (DSP).

Property	Treatment				
	Control	2.5% DSP	5.0% DSP	7.5% DSP	10.0% DSP
Protein *	6.87±0.86^{a}	6.79±0.22^{a}	6.60±0.40^{a}	6.47±0.21^{a}	6.23±0.18^{a}
Fat *	47.71±0.24^{b}	48.31±0.83^{b}	48.81±1.24^{ab}	50.73±0.88^{a}	51.13±1.90^{a}
Fiber *	4.10±0.12^{c}	8.59±0.23^{b}	8.63±0.21^{b}	9.10±0.29^{ab}	9.26±0.27^{a}
pH	7.67±0.30^{a}	7.65±0.18^{a}	7.58±0.47^{a}	7.41±0.33^{a}	7.32±0.08^{a}
Specific volume (cm^3/g)	1.62±0.08^{a}	1.47±0.06^{b}	1.47±0.05^{b}	1.44±0.09^{b}	1.41±0.07^{b}

Means values in the same row followed by the same letters are not significantly different (P<0.05).
** Expressed as % on dry weight basis*

pita bread with increasing date seed levels has been reported [20]. The date seed contains a wide range of saturated fatty acids (lauric, capric, stearic, myristic, palmitic, margaric, arachidic, heneicosanoic, behenic, and tricosanoic) and unsaturated fatty acids (palmitoleic, oleic, linoleic, and linolenic acids) lauric acid is the major saturated fatty acid, and oleic acid is the major unsaturated fatty acid in date seed [10, 33]. The oleic acid content of date cultivars varied from 41.1 to 58.8% [34].

Fiber content

The effect of replacing a portion of wheat flour with DSP on the fiber content of cake was significant (Table 4). Replacing wheat flour with DSP increased the fiber content of the cake. Some previous research has indicated a significant increase in the amount of fiber in pita bread, by increasing the DSP levels [20]. Due to the high nutritional value of dietary fiber, the date seed is suitable for the production of fiber-rich foods [35]. The major dietary fibers in date kernel are insoluble dietary fibers, including lignin, cellulose, and hemicelluloses [36]. Protection against heart disease and cancer, control of blood lipids, regulation of glucose absorption and insulin secretion, and prevention of constipation and diverticular diseases is the health effects of dietary fibers [37].

pH value

The pH of the cake suspensions are presented in Table 4. With increasing the DSP ratio in the cake formula, the pH of the cake was decreased, but based on the analysis of variance, this decrease was not significant.

Specific volume

It was observed that using DSP in cake batter formulation, the specific volume of the cake was reduced. This finding is in agreement with the results obtained by *Shokrollahi* and *Taghizadeh* [19], who reported a decrease in the specific volume of date seed dietary fiber-supplemented bread. It seems that less carbon dioxide retention due to low gluten content has led to a decline in DSP-supplemented cake volume [38, 39].

Peroxide value (PV)

According to the results presented in Table 5, by enriching the cake with DSP, the PV of the final product was increased. Also, the PV of samples gradually increased during the storage period. After 4 weeks of storage, the highest and lowest peroxide values were related to the fat extracted from the cake samples with 10% DSP (1.45 mEq O_2/kg oil) and the control (1.14 mEq O_2/kg), respectively. As it's clear in Table 6, the results of ANOVA show that the first-order terms of DSP ratio and storage time had highly significant effects on the PV of the fortified cake ($p<0.001$). Fig. 1(a) shows that the PV of fortified cake is affected by the storage time and DSP ratio. The obtained model for PV prediction based into the significant terms ($p<0.05$), is brought in Table 7. Whereas the date seed contains large amounts of unsaturated fatty acids [34], higher levels of PV in cakes fortified with DSP are not unexpected. Also, according to the observations of *Ayoubi et al.* [5], cake fat peroxide has increased with increasing pomegranate seed powder (PSP) level and storage time.

Table 5: Historical data RSM design and observed responses of cake formulations.

Run	Actual variables		PV (mEq O_2/kg)	p-AV	TPC (mg GAE/100 g)	MC (%)	Stiffness (g)
	A	B					
1	0.0	0	0.66	2.07	45.32	18.82	736
2	2.5	0	0.72	2.09	51.83	23.52	1330
3	5.0	0	0.75	2.17	53.14	23.55	1392
4	7.5	0	0.78	2.25	63.08	23.65	1613
5	10.0	0	0.82	2.41	63.81	23.68	1984
6	0.0	7	0.73	2.35	45.27	18.13	1278
7	2.5	7	0.74	2.38	46.63	23.11	1413
8	5.0	7	0.85	2.43	46.73	23.39	1462
9	7.5	7	0.91	2.52	54.75	23.55	1642
10	10.0	7	0.95	2.66	55.06	23.69	1676
11	0.0	14	1.03	2.29	23.77	17.11	1667
12	2.5	14	1.06	2.40	25.31	21.78	2099
13	5.0	14	1.13	2.54	27.72	22.33	2121
14	5.0	14	1.18	2.35	26.07	23.37	2177
15	5.0	14	1.07	2.72	29.37	21.39	2064
16	7.5	14	1.18	2.59	35.74	22.40	2591
17	10.0	14	1.19	2.65	36.03	22.58	2668
18	0.0	21	1.06	2.41	11.97	16.95	2033
19	2.5	21	1.12	2.48	15.72	19.95	2263
20	5.0	21	1.24	2.51	17.54	20.02	2642
21	7.5	21	1.32	2.55	20.64	20.13	2787
22	10.0	21	1.36	2.59	21.48	20.14	2939
23	0.0	28	1.14	2.55	10.56	16.45	2367
24	2.5	28	1.32	2.62	13.60	19.74	2368
25	5.0	28	1.34	2.65	13.89	19.82	2666
26	7.5	28	1.42	2.68	14.73	19.88	2912
27	10.0	28	1.45	2.75	16.63	20.04	3057

A: DSP ratio, B: storage time, PV: Peroxide value, p-AV: p-anisidine value, TPC: Total phenolic compounds, MC: Moisture content.

p-Anisidine Value (p-AV)

Table 5 shows the changes in p-AV of the fat extracted from the cakes during storage. Based on these results, using DSP in the cake batter formulation was led to an increase in p-AV. In addition, the p-AV was increased during storage time. At the end of the storage, the lowest and highest p-AV was observed in the control (2.55) and the cake fortified with 10% DSP (2.75), respectively. The higher concentration of secondary lipid oxidation products in DSP-enriched cakes can also be attributed to the high unsaturated fatty acids in date seed [34]. *Ayoubi et al.* [5] also reported that by increasing the PSP levels in cake formulation and during storing time, the p-AV of the extracted lipids from the cake samples increased.

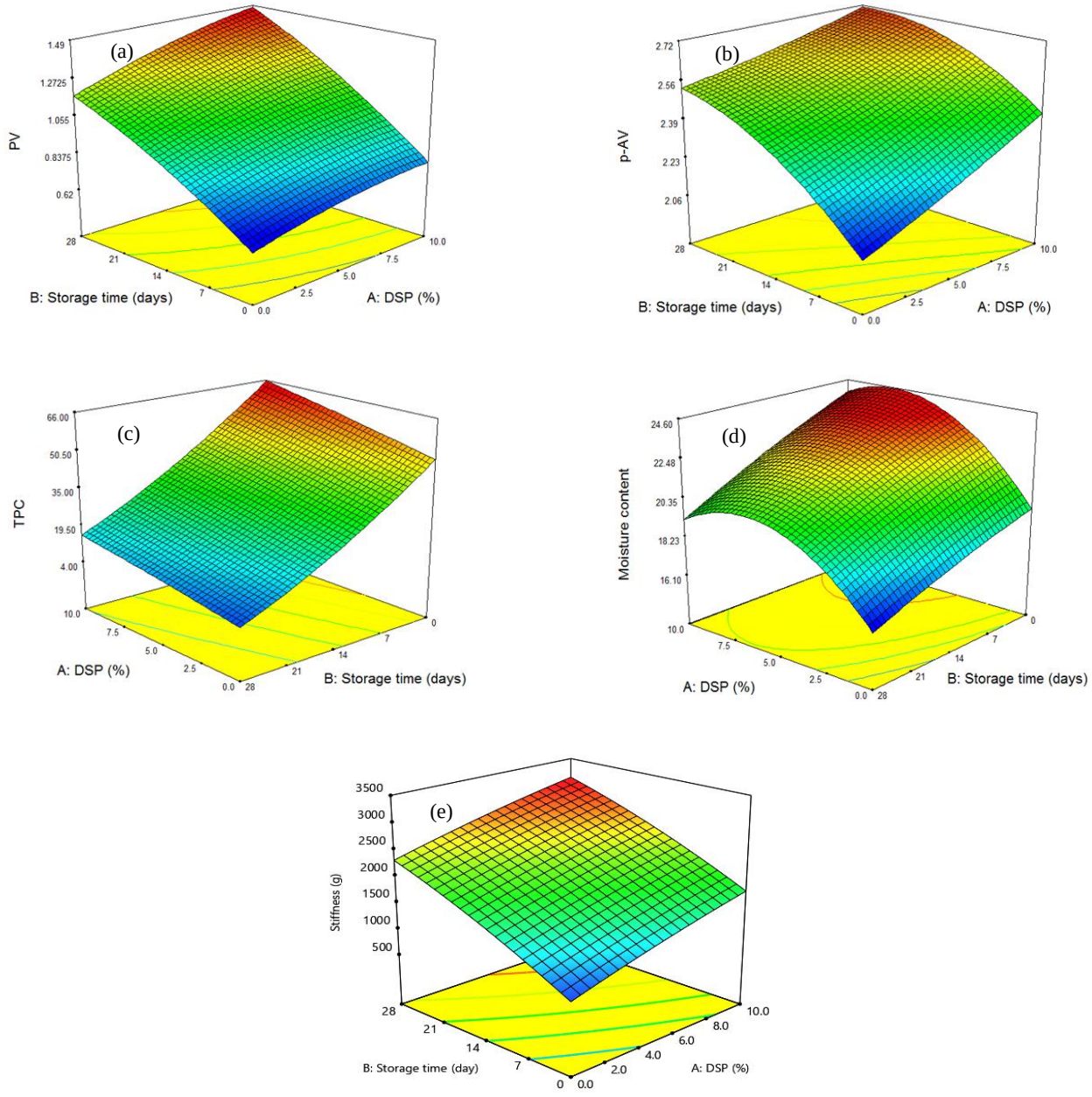

Fig. 1: The effects of date seed powder (DSP) ratio and storage time on PV (a), p-AV (b), TPC (c), MC (d), and Stiffness (e) of cake samples containing different levels of DSP.

The results of ANOVA (Table 6) show that the first-order terms of DSP ratio and storage time had highly significant effects ($p<0.001$) on the p-AV of the DSP fortified cake. Furthermore, the second-order term of storage time had a significant effect ($p<0.05$) on the p-AV of cakes. The effects of independent variables on p-AV are illustrated in Fig. 1(b). Taking into account the significant terms ($p<0.05$), the obtained model for the prediction of p-AV variations is written in Table 7.

Total Phenolic Content (TPC)

As shown in Table 5, by replacing wheat flour with DSP the TPC of the cake samples was increased. It has been found that procyanidins, anthocyanins, glycosides, and flavonoids are the major phenolic compounds in date seed [40]. The TPC of samples decreased gradually during the storage period, while the TPC of control was less than the DSP fortified cakes at all times. The first-order terms of DSP ratio and storage time had highly significant effects

($p< 0.001$) on the TPC of the fortified cakes (Table 6). Fig. 1(c) shows that fortified cake with 10% DSP had the highest level of TPC. The obtained model for TPC based on the significant terms ($p<0.05$), is given in Table 7. Similar results obtained by other researchers indicated that the addition of DSP led to an increase in TPC of bread [20].

Moisture Content (MC)

By replacing wheat flour with DSP, the MC of the cake was increased (Table 5). MC of control was lower than that of DSP enriched cakes at all storage times. Given the effect of fiber on water retention in baking products and the higher fiber content in DSP fortified cakes, this result is not unexpected. The results of ANOVA (Table 6) show that the first-order terms of DSP ratio and storage time and the second-order term of DSP ratio had highly significant effects ($p<0.001$) on the MC of the fortified cake. Fig. 1(d) shows that the MC of fortified cake decreased during the storage time, and the lowest MC was achieved at 0.0% DSP after 28 days of storage.

Platat et al. [20] showed that the moisture content of whole wheat pita bread was lower than DSP enriched samples. The significant effect of date seed flour hydrolysate on the increase of muffin moisture has also been reported *by Ambigaipalan* and *Shahidi* [18].

Stiffness

By adding DSP to the cake formula, the stiffness of the cake texture was increased (Table 5). It seems that as the DSP content was increased, the gluten content of the cake batter was decreased, so the holding gas capacity of the cake batter was decreased, and the product texture became more compact and firmer. Also, during the storage, the stiffness of the cake texture was increased. At all storage times, the control had the lowest stiffness, and the cake supplemented with 10% DSP had the highest stiffness. The results of ANOVA show that similar to other attributes, the first-order terms of DSP ratio and storage time had highly significant ($p<0.01$) effects on the stiffness of the fortified cake (Table 6). Fig. 1(e) shows the lowest stiffness was achieved at 0.0% DSP after 0 days of storage time. Taking into account the significant ($p<0.05$) terms, the obtained model for the prediction of hardness is written in Table 7.

Shokrollahi and *Taghizadeh* [19] also indicated an increase in firmness of bread fortified with date seed dietary fiber. The results of the study directed by Ambigaipalan and Shahidi [18] showed an increase in the stiffness of cake fortified by date kernel flour hydrolysate. *Olcay* and *Demir* [41] reported the firmness of cake had been increased by the substitution of wheat flour with kumquat powder due to the dilution of the gluten with dietary fiber.

According to RSM models, the optimal quality of cakes was observed at DSP ratio of 3.6 % and 0 days after baking. The experimental and predicted values of measured quality attributes under optimized conditions are given in Table 8.

Color indices of cakes

Replacing wheat flour with DSP had a significant effect on the crust and crumb colors of the cake. Using DSP in cake formulation significantly reduced the L^* of the crust (Table 9).

Shokrollahi and *Taghizadeh* [19] also indicated that the addition of date seed dietary fiber in bread formula caused a significant reduction in the lightness of the bread. The redness of the crust decreased with increasing DSP. The crust yellowness of the fortified cake with DSP was significantly lower than the control. The least amount of L^*, a^*, and b^* of the crumb was related to the cake containing 10% DSP. Increasing the amount of DSP led to a decrease in L^* of the crumb. In addition, the results of the analysis of variance indicated a significant increase in crumb redness with increasing the amount of DSP. Fig.2 shows the color changes in fortified cakes with different levels of DSP. Shokrollahi and Taghizadeh [19] obtained similar results, indicating that the addition of date seed dietary fiber led to an increase in a^* of fortified bread. The crumb yellowness in cakes fortified with DSP was significantly less than the control. Significant changes in b^* of fortified bread with date seed dietary fiber have also been reported by *Shokrollahi* and *Taghizadeh* [19].

Microbial Characteristics

According to the microbial results, no growth was observed in any cake samples. Although it is possible that all primary microbial populations have died during cooking, it is also possible that the bacteria have been transformed into a Viable But Non-Culturable (VBNC). So by common cultivation methods, it's not possible to count them. When a bacteria transites to VBNC state, its size

Table 6: Analysis of variance (ANOVA) of the PV, p-AV, TPC, MC, and Stiffness.

Source	Sum of squares	Degree of freedom	Mean square	F-value	p-value
PV					
Model	1.41	5	0.28	95.35	< 0.0001
A-DKP ratio	0.17	1	0.17	58.67	< 0.0001
B-Storage time	1.22	1	1.22	410.16	< 0.0001
AB	7.569×10^{-3}	1	7.569×10^{-3}	2.55	0.1251
A^2	4.621×10^{-3}	1	4.621×10^{-3}	1.56	0.2257
B^2	9.852×10^{-3}	1	9.852×10^{-3}	3.32	0.0827
Residual	0.062	21	2.967×10^{-3}		
Lack of Fit	0.056	19	2.960×10^{-3}	0.98	0.6222
Pure Error	6.067×10^{-3}	2	3.033×10^{-3}		
Cor Total	1.48	26			
p-AV					
Model	0.72	5	0.14	18.67	< 0.0001
A-DKP ratio	0.23	1	0.23	29.88	< 0.0001
B-Storage time	0.45	1	0.45	57.58	< 0.0001
AB	0.012	1	0.012	1.54	0.2290
A^2	4.662×10^{-4}	1	4.662×10^{-4}	0.060	0.8085
B^2	0.034	1	0.034	4.36	0.0493
Residual	0.16	21	7.738×10^{-3}		
Lack of Fit	0.094	19	4.949×10^{-3}	0.14	0.9945
Pure Error	0.068	2	0.034		
Cor Total	0.88	26			
TPC					
Model	7741.69	5	1548.34	78.90	< 0.0001
A-DKP ratio	565.08	1	565.08	28.80	< 0.0001
B-Storage time	7119.35	1	7119.35	362.79	< 0.0001
AB	11.34	1	11.34	0.58	0.4555
A^2	0.64	1	0.64	0.032	0.8589
B^2	45.90	1	45.90	2.34	0.1411
Residual	412.11	21	19.62		
Lack of Fit	406.66	19	21.40	7.86	0.1187
Pure Error	5.45	2	2.72		
Cor Total	8153.80	26			

Table 6: Analysis of variance (ANOVA) of the PV, p-AV, TPC, MC, and Stiffness

Source	Sum of squares	Degree of freedom	Mean square	F-value	p-value
MC					
Model	120.92	5	24.18	30.95	< 0.0001
A-DKP ratio	43.90	1	43.90	56.18	< 0.0001
B-Storage time	48.53	1	48.53	62.11	< 0.0001
AB	1.01	1	1.01	1.30	0.2679
A^2	26.58	1	26.58	34.01	< 0.0001
B^2	0.20	1	0.20	0.26	0.6142
Residual	16.41	21	0.78		
Lack of Fit	14.45	19	0.76	0.78	0.7017
Pure Error	1.96	2	0.98		
Cor Total	137.33	26			
Stiffness					
Model	8.633×10^{-6}	5	1.727×10^{-6}	45.68	< 0.0001
A-DKP ratio	2.229×10^{-6}	1	2.229×10^{-6}	58.98	< 0.0001
B-Storage time	6.353×10^{-6}	1	6.353×10^{-6}	168.08	< 0.0001
AB	1592.01	1	1592.01	0.042	0.8394
A^2	8305.27	1	8305.27	0.22	0.6441
B^2	36709.57	1	36709.57	0.97	0.3356
Residual	7.937×10^{-5}	21	37797.49		
Lack of Fit	7.874×10^{-5}	19	41440.14	12.98	0.0739
Pure Error	6384.67	2	3192.33		
Cor Total	9.427×10^{-6}	26			

P < 0.05 indicates statistical significance

PV: Peroxide value, p-AV: p-anisidine value, TPC: Total phenolic compounds, and MC: Moisture content

Taking into account the significant terms (p<0.05), the obtained model for MC prediction is written in Table 7.

Table 7: Response surface predicted models for physicochemical characteristics of cake samples containing date seed powder (DSP) in terms of coded factors.

Responses	Model	R^2
PV (mEq O_2/kg)	$Y = 1.09 + 0.12A + 0.31B + 0.035AB - 0.031A^2 + 0.045B^2$	0.96
p-AV	$Y = 2.50 + 0.14A + 0.19B - 0.044AB + 9.862\times10^{-3}A^2 - 0.084B^2$	0.82
TPC (mg GAE/100 g)	$Y = 31.19 + 6.72A - 23.87B - 1.35AB - 0.36A^2 + 3.09B^2$	0.95
MC (%)	$Y = 22.27 + 1.87A - 1.97B - 0.40AB - 2.35A^2 - 0.21B^2$	0.88
Stiffness (g)	$Y = 2131.90 + 422.32A + 712.92B - 15.96AB - 41.62A^2 - 87.51B^2$	0.92

A: DKP ratio, B: storage time, PV: Peroxide value, p-AV: p-anisidine value, TPC: Total phenolic compounds, and MC: Moisture content

Table 8: Experimental and predicted values of the responses in cake samples containing date seed powder (DSP) under optimized conditions.

Responses	Experimental	Predicted
PV (mEq O_2/kg)	0.69±0.05	0.71
p-AV	2.21±0.09	2.18
TPC (mg GAE/100 g)	54.93±1.13	55.79
MC (%)	23.49±0.83	23.17
Stiffness (g)	1182±57	1201

PV: Peroxide value, p-AV: para-anisidine value, TPC: Total phenolic compounds, and MC: Moisture content.

Table 9: The effect of different levels of Date Kernel Powder (DSP) on the color indices of cake samples.

Treatment	Color indices					
	*L**		*a**		*b**	
	Crust	Crumb	Crust	Crumb	Crust	Crumb
0.0% DSP	52.67±0.46^{a}	52.70±0.52^{a}	19.76±0.09^{a}	2.63±0.08^{e}	38.61±1.30^{a}	25.84±0.19^{a}
2.5% DSP	38.26±0.97^{b}	28.73±0.67^{b}	15.76±0.21^{b}	5.93±0.05^{d}	26.17±0.18^{b}	15.96±0.34^{b}
5.0% DSP	35.55±0.47^{c}	25.63±0.10^{c}	15.04±0.28^{c}	7.61±0.05^{c}	20.45±0.58^{c}	12.82±0.15^{c}
7.5% DSP	32.89±0.22^{d}	20.26±0.35^{d}	14.43±0.13^{d}	7.83±0.06^{b}	16.76±0.49^{d}	10.21±0.53^{d}
10.0% DSP	32.38±0.45^{d}	19.34±1.47^{d}	13.30±0.11^{e}	8.39±0.29^{a}	13.45±0.04^{e}	10.14±0.10^{d}

Means values in the same column followed by the same letters are not significantly different (P<0.05).

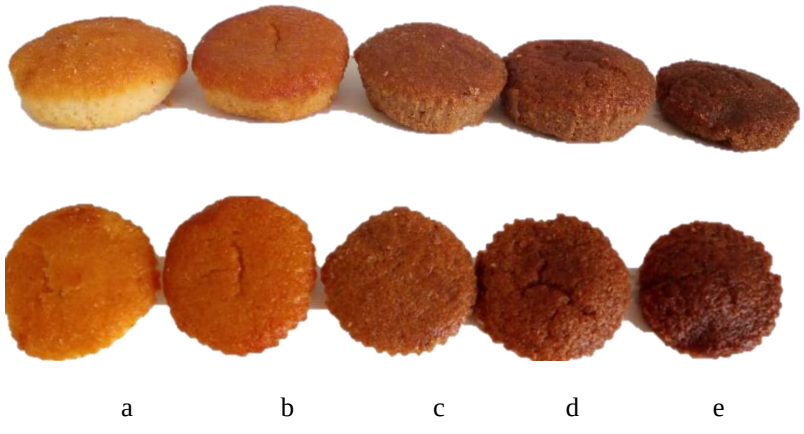

Fig. 2: The cupcakes produced using different date seed powder (DSP) levels (control (a), 2.5% DSP (b), 5% DSP (c), 7.5% DSP (d), and 10% DSP (e)).

decreases and some changes occur in its metabolic system. Since in this situation, microorganisms lose their ability to grow in the solid media, it is not possible to count them by conventional culture methods. Similar results were observed in wheat flour substitution with PSP in cake formulation [5].

Sensory evaluation

As it is evident in Table 10, based on the sensory evaluation results, the addition of DSP to the cake formula darkened the color and reduced the crust and crumb color and appearance scores. Also, as the DSP level increased, the taste and flavor score of the cake decreased. Increasing

Table 10: Sensorial properties of cake samples containing date seed powder (DSP).

Property	Treatment				
	Control	2.5% DSP	5.0% DSP	7.5% DSP	10.0% DSP
color	4.60±0.16^{a}	4.40±0.16^{a}	4.30±0.15^{a}	4.50±0.17^{a}	4.30±0.15^{a}
texture	4.80±0.13^{a}	4.60±0.16^{ab}	4.40±0.22^{b}	4.10±0.10^{c}	3.80±0.13^{d}
flavor	4.90±0.10^{a}	4.60±0.16^{b}	4.30±0.15^{bc}	4.00±0.15^{cd}	3.70±0.15^{d}
overall acceptance	4.80±0.13^{a}	4.60±0.16^{ab}	4.30±0.21^{bc}	4.20±0.20^{cd}	3.90±0.10^{d}

Means values in the same row followed by the same letters are not significantly different (P<0.05).

the amount of DSP resulted in reducing texture and mouthfeel score. The overall acceptability score results also showed that by increasing the replacement level, the score of overall acceptability decreased. Significant reduction of color, texture, and overall acceptance scores of fortified bread with date seed dietary fiber was reported by Shokrollahi and Taghizadeh [19]. Dark brown color, acceptable texture and taste, and low sensory acceptance in date seed flour enriched muffins reported by *Ambigaipalan* and *Shahidi* [18].

The acceptable taste and aroma of fortified bread with date seed dietary fiber have been reported by *Shokrollahi* and *Taghizadeh* [19].

CONCLUSIONS

The use of fiber supplements for enhancing the health benefits of food products improves the functional properties of many foods and could also provide environmental benefits for food producers. Date seeds as a cheap and high nutritional value source could be used as dietary fiber supplements in food product enrichment. The results showed by increasing the percentage of substitution of DSP in cake formula, the amount of fiber, fat, MC, and TPC increased, while the protein content decreased. Also, the addition of DSP to the cake was reduced the specific volume and increased the stiffness. The PV and p-AV were increased in cakes fortified with DSP, and color of the crust and crumb of DSP fortified cake was darkened. The highest overall acceptance of DSP fortified cakes was observed for cake fortified with 2.5% DSP. According to RSM models, the optimal quality of cakes was observed at DSP ratio of 3.6% immediately after baking. It can be concluded that DSP, as a rich source of bioactive compounds, can be used to produce functional cakes.

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