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Research Article
Influence of Sweet Lupine Husk's Addition on Fat-Free Stirred Yogurt Properties

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Abstract

Background and Objective: Enrichment of free fat yogurt with functional by-product could be enhancing texture and sensory acceptability. The present study aimed to use sweet lupine husk powder as a functional by-product novel source of fiber that could be incorporated into stirred free fat yogurt for enhancement texture and sensory acceptability. **Materials and Methods:** Free lupine husk powder have been added with 0, 1, 3 and 5% to milk incubated with yogurt starter for preparation lupine husk yogurt and stored for 14 days and performed for variance analysis using ANOVA. The study was carried out at Quality Control Pilot Lab, Dairy Science Department and Alexandria University from June-August, 2019. **Results:** Organoleptic evaluation results revealed that yogurt (T_3) fortified with 1% sweet lupine husk scored the highest overall acceptability during storage. The pH decreased significantly ($p<0.05$) is correlated with increases in titratable acidity during storage at 4°C. Syneresis of T_1 yogurt was lower ($p<0.05$) than control during storage. Acetaldehyde contents, viscosity and hardness values of T_1 and T_3 were higher via a control on days 7 and 14, respectively. **Conclusion:** The addition of lupine husk (as a novel dietary fiber by-product) would complement its functional characteristics. The addition of 1% lupine husk scored highest in all parameters including sensory characteristics and texture.

Key words: Yogurt, sweet lupine, texture, fat replacer, dietary fiber, by-product, sensory evaluation

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

One of the recent trends for the creation of healthy food with enhanced functionality is the enrichment of yogurt with different sources of fiber[1]. Recently, the interest in dietary fiber-rich foods has increased and led to the creation of a huge market of fiber-rich ingredients and by products[2].

Enriching the yogurt by adding fiber-rich ingredients such as fruits, grains and nuts can enhance the textural and rheological functioning of yogurt[3]. Owing to the water-binding properties of yogurt, mostly water-soluble dietary fibers are used for its enrichment. Dietary fibers in yogurt act as a stabilizer to increases the viscosity, prevent syneresis and enhance the texture by increasing creaminess. There are two categories of dietary fiber, water-soluble and water-insoluble. Water-insoluble fibers minimize the transient time in the intestine, thereby increasing the volume of feces and making it soft. The water-soluble dietary fibers prolong the gastric emptying, reduce the inclusion of glucose and lessen the cholesterol levels[4].

The primary functional characteristic of yogurt is its texture. It can be enhanced through the utilization of gelling, thickening and stabilizing elements[5]. Pectin is widely used as a stabilizer due to its ability to produce aqueous gel in an acidic condition in fermented dairy products[6]. Stabilizers also prevent syneresis, improves the texture and consistency of yogurt[7]. Fibers are also used in yogurt as a substitute for the stabilizers due to similar textural properties[8].

Sweet Lupine Husk fibers are a new source of food that can be derived from the endosperm of sweet lupine (*Lupinus albus*). This has been approved by the legislation to be used as human food[9]. It is a significantly non-starch polysaccharide in the form of thickened cell walls of the lupine seed endosperm, with residual protein. It is described as a pale color powder with low odor and taste and can be utilized as a 'nonintrusive' fiber ingredient in foods[10].

The present study determines the amount of sweet lupine husk powder that is optimal incorporation in stirred free fat yogurt with the acceptability of yogurt qualification.

MATERIALS AND METHODS

Study area: The study was carried out at the Department of Food and Nutrition Sciences, College of Agricultural and Food Sciences, King Faisal University, Al Hofuf, Kingdom of Saudi Arabia from June-August, 2019.

Lupine husks powder: Local Egyptian breeds of sweet lupin (*Lupinus albus* L.) variety Giza) were obtained from the Agricultural Research Centre, Giza, Egypt. The husks were removed from the seeds in a laboratory hammer mill, grounded husk fibers then re-milled and sieved to pass through sieve 500 µm. The samples were packed and kept under refrigeration for analysis. Obtained lupine husk have moisture 8.76±0.06, Crude protein 4.8±0.26, Total ash 2.57±0.18, Crude fat, Starch 0.20±0.04, soluble dietary fiber 43.7±2.19, Insoluble dietary fiber 41.5±1.12 and Total dietary fiber 85.20±4.38 according to Ahmed[11].

Starter culture: Direct Vat Set (DVS) containing *Streptococcus thermophilus* and *Lactobacillus delbrueckii* sub sp. *bulgaricus* (YCX31) was obtained from Chr. Hansen's laboratories, Denmark.

Preparation of lupine husks free fat stirred yogurt: Yogurt is produced from fat-free cow milk and lupine husk powder was added. The milk is heated at 90°C for 10 min in a stainless container and then cooled off at 42°C. The starter is added at 3%. The samples were incubated at 42°C until coagulation occurred, then stirred, cooled and stored in a refrigerator at 4° C for 15 days as depicted in Fig. 1. Samples were taken for sensory, rheological, chemical and microbiological analysis at zero time, 7 and 15 days, respectively.

Chemical and physical analysis: Milk and yogurt samples were analyzed for total solid, protein content and total acidity were determined by following the standard procedures of the Association of Official Analytical Chemists[12]. The pH was determined by using a digital pH meter (Inolad model 720, Germany). Acetaldehyde contents were examined according to Lees and Jago[13].

The syneresis of yogurt was calculated immediately after coagulation and during the storage time of 15 days at 4°C±1 [14]. The viscosity of the samples was also measured using a rotational viscometer DV-II+Pro Brookfield Engineering

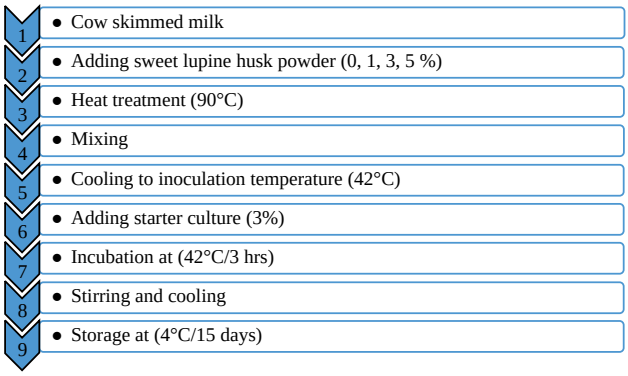

Fig. 1: Steps of lupine husks free fat stirred yogurt preparing

Laboratories, Inc. (USA). The analysis was carried out at 20°C by using spindle (L2) at a speed of 100 rpm and the reading was recorded as cP.

Texture profile analysis: Texture Profile Analysis (TPA) was carried out by a compression test that generated a plot of force (g) versus time (s) and was evaluated instrumentally using a texture analyzer (Texture Pro CT V1.2 Build 9, Brookfield Engineering Labs, Inc. USA). In the first stage, it was carried out at a pre-test speed of 1 mm sec^{-1} and a test speed of 1.7 mm sec^{-1}. The typical texture profile (force vs. time) curve was obtained with one complete run. Hardness, gumminess, cohesiveness, adhesiveness, and springiness of yogurt samples were calculated through the software program (Texture software, Brookfield Instruments).

Microbiological analysis: Regular microbiological methods (colony-forming unit [cfu]). *Streptococci* were counted on M-17 agar (Oxoid) and *Lactobacilli* were counted on Man Rogosa Sharpe agar (Oxoid). Both media were incubated under microaerophilic conditions at 37°C for 48 hrs.

Sensory evaluation: According to Peryam and Pilgrim[15] A 9-point hedonic scale (1 = dislike extremely and 9 = like extremely).

Statistical analysis: All data is presented as mean±SD for three replicates for each sample. Multiple comparisons were performed through the application of Univariate Analysis of Variance (ANOVA) using Stat graphics 16.1.11 (Stat Point Technologies, Inc. Virginia, USA). The differences were considered significant at $p<0.05$.

RESULTS AND DISCUSSION

The effects of lupine husk supplementations on total solids, protein and ash were significant ($p<0.05$). Total solid contents ranged from all treatments between 8.56 and 13.53% and total protein and ash values also increased in correlations with the addition of lupine husk powder (Table 1).

The pH values of all treatments were between 4.7 and 4.8 on day 1 and 4.36 and 4.46 on day 14, respectively. Lupine husk treatment pH values of the yogurt T_1 were non significant

Table 1: Chemical and physical characteristics of sweet Lupine husk fat-free stirred yogurt samples during storage (n = 3)

Days	T_0	Control	T_1	1%	T_2	3%	T_3	5%
Total solids (TS)								
1	A	8.9±0.1^{d}	A	10.06±0.05^{c}	A	11.66±0.05^{b}	A	13.53±0.07^{a}
7	B	8.72±0.005^{d}	B	9.71±0.028^{c}	B	10.90±0.105^{b}	B	12.92±0.005^{a}
14	C	8.56±0.005^{d}	C	9.36±0.115^{c}	C	10.63±0.057^{b}	C	12.27±0.005^{a}
Protein								
1	A	3.56±0.07^{c}	A	3.63±0.03^{bc}	A	3.66±0^{b}	A	3.77±0.01^{a}
7	B	3.33±0.011^{d}	B	3.46±0.005^{c}	B	3.41±0.005^{b}	B	3.63±0.045^{a}
14	C	3.17±0.015^{c}	C	3.33±0.023^{b}	C	3.13±0.005^{c}	C	3.45±0.041^{a}
Ash								
1	A	0.653±0.005^{d}	A	0.683±0.005^{c}	A	0.736±0.005^{b}	A	0.803±0.005^{a}
7	B	0.616±0.005^{d}	B	0.636±0.005^{c}	B	0.676±0.005^{b}	B	0.760±0^{a}
14	C	0.583±0.011^{c}	B	0.636±0.011^{b}	C	0.616±0.037^{bc}	C	0.726±0.011^{a}
pH								
1	A	4.7±0^{b}	A	4.7±0^{b}	A	4.8±0^{a}	A	4.8±0^{a}
7	B	4.6±0^{a}	B	4.6±0^{a}	B	4.5±0^{b}	B	4.6±0^{a}
14	C	4.46±0.05^{a}	C	4.36±0.05^{a}	C	4.36±0.05^{a}	C	4.46±0.05^{a}
Acidity (%)								
1	B	0.3±0^{c}	C	0.3±0^{c}	C	0.4±0^{b}	B	0.5±0^{a}
7	A	0.36±0.05^{c}	B	0.46±0.05^{b}	B	0.5±0^{b}	A	0.6±0^{a}
14	A	0.4±0^{b}	A	0.56±0.05^{a}	A	0.6±0^{a}	A	0.6±0^{a}
Syneresis (%)								
1	A	25±0^{c}	A	24±0^{d}	A	29±0^{b}	A	33.66±0.57^{a}
7	B	23±0^{c}	B	21±0^{d}	B	24±0^{b}	B	27±0^{a}
14	C	22±0^{c}	B	21±0^{d}	C	23±0^{b}	C	26±0^{a}
Acetaldehyde (mg kg^{-1})								
1	A	8.56±0.05^{a}	AB	8.06±0.05^{c}	A	8.3±0^{b}	A	8.06±0.05^{c}
7	B	8.1±0^{a}	A	8.16±0.05^{a}	B	8.16±0.05^{a}	B	7.83±0.05^{b}
14	C	7.46±0.05^{d}	B	7.96±0.05^{a}	C	7.83±0.05^{b}	C	7.66±0.05^{c}

A,B,C,D Letter indicate statistically variance between treatments

Table 2: Starter culture counts of sweet lupine husk fat-free stirred yogurt samples during storage (n = 3)

Days	T_0	Control	T_1	1%	T_2	3%	T_3	5%
S. thermophilus								
1	B	8 ± 0^{b}	B	8.66 ± 0.57^{a}	B	8 ± 0^{b}	B	8 ± 0^{b}
7	B	8.33 ± 0.57^{a}	AB	9 ± 0^{a}	A	9 ± 0^{a}	A	8.66 ± 0.57^{a}
14	A	9 ± 0^{a}	A	9.66 ± 0.57^{a}	A	9.33 ± 0.57^{a}	B	8 ± 0^{b}
L. bulgaricus								
1	C	7 ± 0^{b}	C	6 ± 0^{c}	B	7 ± 0^{b}	A	8.66 ± 0.57^{a}
7	B	9 ± 0^{a}	B	8 ± 0^{b}	A	9 ± 0^{a}	A	9 ± 0^{a}
14	A	10 ± 0^{a}	A	9 ± 0^{b}	A	9 ± 0^{b}	A	9 ± 0^{b}

A,B,C,D Letter indicate statistically variance between treatments

via control T_0 ($p>0.05$). T_2 and T_3 were found to be significantly ($p<0.05$) different from control (T_0). This increase in pH may be attributed to the water holding capacity of lupine husk thus diluting the concentrations of lactic acid and other organic acids produced by cultures. This might have resulted in an increase in pH of fiber -fortified yogurt[16]. The more the lupine husk was added, the more water might have been absorbed, thus increasing the pH values of all yogurt treatments. It was reported that the addition of various dietary fiber sources did not influence the pH of the fortified yogurts[17].

Titratable acidity increased in conjunction with the addition of lupine husk powder and storage. The highest titration acidity was detected on day 14 (0.6% of T_2 and T_3) and the lowest on day 1 (0.3% of T_0 and T_1). While the effects of titration acidity of the yogurts were significant at day 14, the lupine husk yogurt treatments were significant ($p<0.05$), with persimmon lupine supplemented yogurts exhibiting the highest titration acidity. Since the lupine husk content was increased from 1-5% (T_1-T_3), therefore, an increase in acidity is expected. But our results showed a low increase which may be attributed to the water holding capacity of the lupine husk, thus diluting the concentration of lactic acid and other organic acids produced by cultures in yogurt. However, the titratable acidity of yogurt samples was within an average of 0.9%, which recommended value for plain yogurts[18].

Syneresis or whey separation is an important defect in yogurt. Lupine husk powder addition to yogurt milk decreased significantly whey separation values during storage. Although the addition of lupine husk powder reduced the separation of whey at a concentration of 1%, it increased with the addition of lupine husk powder by 3 and 5%. The effects of different fiber sources addition on yogurt were varied. Güzeler *et al.*[19] described an increasing yogurt syneresis correlated with apricot fiber addition. While 1% orange fiber addition decreased syneresis and improved the textural properties of the yogurts as mentioned by Lario *et al.*[20]. Whey separation in yogurt reduced by adding wheat bran fibers[21], sweet potato[22], Rice bran[23] and orange[24]. The binding capacity of added dietary fibers to yogurt protein micelle makes the composition more capable to bind whey and avoid syneresis[25].

Acetaldehyde content as the main flavor compound in yogurt must be between 23 and 41 mg kg^{-1} of yogurt[26]. Yogurt free lupine husk had higher acetaldehyde values than lupine husk -added samples after both 7 and 14 days of storage ($p<0.05$). Acetaldehyde values of the yogurts in beginning in storage varied between 8.06 and 8.56 mg kg^{-1}, which were lower than the control treatment. That decrease is due to the alcohol dehydrogenase activity of yogurt starters on the relationship between acetaldehyde and ethanol[26,27].

The bacterial count of yogurt samples including *S. thermophilus* and *L. bulgaricus* is presented in Table 2. With an increase in the lupine husk, the total bacterial count for both strains increased. The mean values of the streptococci count of T_0 and T_1 differed significantly ($p<0.05$) on day 1. T_2 and T_3 were similar to the control one T_0. *Lactobacilli* count for $T_{0\ higher}$ than T_1, similar to T_2 and lower than T_0. During the storage period, increases in the total bacterial count with an increase in lupine husk level may be due to the increase in moisture content[28]. The increase in the *Lactobacilli* count with the increase in lupine husk content may also be due to the prebiotic effect of the lupine husk, which agreed with the addition of grape fiber to yogurt as mentioned by Dibazar *et al.*[29].

Table 3 reveals the textural characteristics (Viscosity, Hardness, Adhesiveness, Cohesiveness, Gumminess and Springiness) of the lupine husk yogurts and their control. The viscosity is defined as the area under the force versus time curve. The maximum viscosity indices were respectively recorded on days 14 for T_3, T_2 and T_1. The effects of lupine husk addition on the viscosity index of the yogurts were a significant increase on days 1, 7 and 14 ($p<0.05$), free lupine husk yogurts displaying lower viscosity index values than the lupine husk yogurt. Espirito *et al.*[30] reported that yogurts supplemented with fruit fiber exhibited reduced fermentation times but increased firmness, consistency and cohesiveness.

Table 3: Textural characteristics of sweet lupine husk fat-free stirred yogurt samples during storage (n = 3)

Days	T_0	Control	T_1	1%	T_2	3%	T_3	5%
Viscosity (cP)								
1	C	1120±0^{d}	C	1210±0^{c}	C	1300±0^{b}	C	1360±0^{a}
7	B	1180±0^{d}	B	1250±0^{c}	B	1312±0^{b}	B	1390±0^{a}
14	A	1210±0^{d}	A	1300±0^{c}	A	1330±0^{b}	A	1395±0^{a}
Hardness (g)								
1	C	140±0^{d}	C	160±0^{c}	C	180±0^{b}	C	230±0^{a}
7	B	155±0^{d}	B	181±0^{c}	B	210±0^{b}	B	256±0^{a}
14	A	173±0^{d}	A	201±0^{c}	A	240±0^{b}	A	301±0^{a}
Adhesiveness (g)								
1	A	-58.33±0.57^{d}	A	-55±0^{c}	A	-53±0^{b}	A	-50±0^{a}
7	B	-80±1^{d}	B	-71±0^{c}	C	-69±0^{b}	B	-65±0^{a}
14	C	-102.66±0.57^{d}	C	-89±0^{c}	B	-60±0^{a}	C	-69±0^{b}
Cohesiveness								
1	A	0.3±0^{d}	A	0.35±0^{c}	A	0.4±0^{b}	A	0.45±0^{a}
7	A	0.3±0^{d}	B	0.33±0^{c}	B	0.36±0^{b}	B	0.41±0^{a}
14	B	0.29±0^{d}	C	0.31±0^{c}	C	0.34±0^{b}	C	0.36±0^{a}
Gumminess (g)								
1	B	70±0^{d}	B	90±0^{c}	B	110±0^{b}	B	150±0^{a}
7	A	79±0^{d}	A	110±0^{c}	A	129±0^{b}	A	169±0^{a}
14	C	61±0^{d}	C	70±0^{c}	C	85±0^{b}	C	101±0^{a}
Springiness (mm)								
1	A	94±0^{c}	A	96±0^{b}	A	97±0^{a}	A	97±0^{a}
7	B	91±0^{c}	B	94±0^{b}	B	94±0^{b}	B	95±0^{a}
14	C	85±0^{d}	C	86±0^{c}	C	87±0^{b}	C	88±0^{a}

A, B, C, D letter indicate statistically variance between treatments

Table 4: Organoleptic characteristics of sweet Lupine husk fat-free stirred yogurt samples during storage (n = 3)

Days	T_0	Control	T_1	1%	T_2	3%	T_3	5%
Appearance								
1	A	8.41±0.58^{a}	A	8.41±0.80^{a}	A	8.08±0.66^{ab}	A	7.41±0.49^{b}
7	B	7.75±0.41^{a}	B	7.5±0.54^{a}	B	7.33±0.51^{ab}	B	6.91±0.20^{b}
14	B	7.75±0.27^{a}	AB	7.75±0.41^{a}	AB	7.58±0.37^{a}	B	6.91±0.37^{b}
Consistency								
1	A	9±0.31^{a}	A	8.83±0.25^{a}	A	8.66±0.40^{a}	A	8.25±0.27^{b}
7	A	8.83±0.25^{a}	A	8.75±0.27^{a}	A	8.5±0.31^{ab}	A	8.33±0.40^{b}
14	A	8.66±0.25^{a}	A	8.58±0.20^{a}	A	8.41±0.37^{a}	A	7.91±0.37^{b}
Flavor								
1	A	8.5±0.54^{a}	A	8.16±0.75^{a}	A	7.91±0.49^{a}	A	7.16±0.40^{b}
7	B	7.91±0.20^{a}	A	7.66±0.51^{a}	A	7.66±0.51^{a}	AB	6.83±0.25^{b}
14	B	7.91±0.20^{a}	A	7.66±0.51^{ab}	A	7.41±0.49^{b}	B	6.75±0.27^{c}
Overall acceptance								
1	A	9±0.31^{a}	A	8.83±0.40^{a}	A	8.25±0.41^{b}	AB	8.25±0.27^{b}
7	AB	8.83±0.25^{a}	A	8.66±0.25^{ab}	A	8.41±0.37^{b}	A	8.33±0.40^{b}
14	B	8.5±0.54^{a}	A	8.5±0.31^{a}	A	8.08±0.37^{ab}	B	7.75±0.52b

A, B, C, D Letter indicate statistically variance between treatments

Hardness is represented as the highest force sensed and recorded by the load cell of the texture analysis device. Day 14 yogurts exhibited the maximum (T_3: 301 g) and minimum firmness (T_0: 173 g). Present results matched with that obtained by Perina *et al.*[31] that the addition of passion fruit peel powder to yogurt significantly affected the hardness. Also, results are in agreement with those obtained by Crispin-Isidro *et al.*[32] which reported that yogurt hardness increases at a level of 2-4% inulin addition. This may be because fibers are acting as 'filler' between yogurt components.

The decrease in adhesiveness with an increase in lupine husk level may be due to the decreased binding capacity of yogurt thus reducing adhesiveness and increasing cohesiveness[33]. During the storage period T_1 lupine husk yogurt exhibited the lowest cohesiveness followed by T_2 and T_3. The effects of lupine husk supplementation on the cohesiveness of the yogurts were significant on days 1, 7 and 14 ($p<0.05$).

The organoleptic properties of experimental yogurt samples are shown in Table 4. Appearance scores were similar

in free lupine husk yogurt treatment and 1% addition during the storage period. Increasing the level of the lupine husk there was a little bit decrease in the consistency, flavor and overall acceptance scores. Free lupine husk yogurt treatment showed the significantly ($p<0.05$) highest score, followed by T_1, T_2 and T_3 yogurt samples containing 1, 2 and 3% of the lupine husk, respectively. The decrease in the flavor of yogurt with every increment of lupine husk may be due to the prevention of the release of the flavoring from the network formed by fiber addition. Fernandez-Garcia *et al.*[34] have also reported a decrease in flavor quality of yogurt by the addition of fiber.

Consistency scores for T_1 were nearest to control T_0 followed by T_2 and T_3. The mean values with an increase in lupine husk may be due to the stabilizing effect of fiber on the texture of yogurt. Lupine husk (polysaccharides) acts as a stabilizer in yogurt, thus improving its consistency[35]. However, a decrease in the texture of T_3 may be due to the undesirable grittiness produced by the addition of more lupine husk. Besides, more fiber content of lupine husk absorbs more moisture thereby making yogurt tougher[34] which may further support our results.

Hussein *et al.*[35] have reported that the addition of polysaccharides (fiber) improves body, texture, appearance and mouthfeel and retards whey separation of yogurt. The addition of polysaccharides (fiber) as a stabilizer in the manufacture of yogurt is a common practice. Texture enhancement and preventing syneresis is the main functions of hydrocolloids, which act as stabilizers[36] in yogurt. Tomic *et al.*[37] reported that the addition of different sources of fibers (soy, rice, oat, corn and sugar beet), at the level of 1.32%, led to lower overall flavor and texture scores a grainy flavor and a gritty texture were intense in all samples except in those made with oat fiber. However, fiber size is also an important factor in yogurt formulation because of its impact on general acceptance. Besides, Raju and Pal[38] reported that yogurt color changes with adding fiber, while no marked changes in yogurt are affected with bamboo or inulin fibers[17].

CONCLUSION

Fortifying yogurt or dairy products with fiber is of great interest to improve the functionality and create foods with health benefits. The addition of lupine husk (as dietary fiber) would complement its healthy characteristics. The results of the present study showed that fortifying yogurt (T_1) with 1% lupine husk scored highest in all parameters including sensory characteristics.

SIGNIFICANCE STATEMENT

This study discovers the possibility of adding sweet lupine husk powder in yogurt preparation that can be beneficial for fat replacement and texture enhancement. This study will help the researcher to uncover the novel by-products additive areas of fat replacers and texture enhancers in yogurt that many researchers were not able to explore. Thus a new additive in yogurt may be arrived at.

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