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Effect of Almond Oil Supplementation in Milk on Serum Lipid Profile in Hyperlipidemic Rats

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[NY]Conception and design, Collection and assembly of data, [MK]Analysis and interpretation of the data, [HJ]Statistical expertise, [MB, SK, SA]Final approval and guarantor of the article

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A B S T R A C T

Background: Flavored milk, derived from buffalo milk renowned for its calcium and vitamin richness, is increasingly sought after due to global health consciousness and concerns about cardiovascular health. Animal fat, integral to elevating serum lipoprotein levels, undergoes reduction in milk products, complemented by almond oil supplementation, resulting in a healthier product.

Objective: The aim of this study was to investigate the effects of almond oil milk on hyperlipidemia.

Methodology: This study was carried out to decrease fat levels in flavored milk through almond oil addition, analyzing its physicochemical and sensory attributes alongside its impact on serum lipid profiles.

Results: Proximate composition analysis revealed a decline in moisture and pH content with increasing almond oil concentration, while crude protein, fat, and ash percentages rose. Sensory evaluation highlighted significant treatment effects on taste, flavor, and overall acceptability, with higher ratings for 4% and 5% almond oil treatments. Subsequent rat efficacy studies, initiated with a cholesterol-rich diet to induce hypercholesterolemia, demonstrated a notable impact of almond oil treatments on serum lipid profiles. Cholesterol and LDL levels significantly decreased, while HDL levels increased, and triglycerides decreased, particularly in the T3 group with the highest almond oil concentration.

Conclusions: This study underscores almonds' efficacy in managing lipid profiles and cardiovascular diseases, suggesting the potential of almond-based products in mitigating metabolic ailments.

Keywords: Almond oil, milk supplementation, flavored milk, lipid profile, hyperlipidemia

Introduction

Almonds are considered a pleasant nut tree consumed by the people worldwide having broad applications in the numerous fields of industry like cosmetics, food and drug supplementation and pharmaceutical companies. [1] Amusing macro and micronutrient profile make it choose able for the extraction of the oil from this nut to a high demand in the recent time. Its production rate is being tried to elevate along with the high nutrient qualities it possesses. The aroma and taste make the almond oil interesting and likable for the flavoring attribute in the industrial products. [2] Food and Agriculture Organization (FAO) noticed the recorded rise in worldwide field production of almond to 3,182,902 amounts in tons dated 2018 from the last ten years. [3] Properties of the field almond like nutrient profiling and chemical compositional variations are significantly related to the harvesting conditions, time of cultivation, technical knowledge and handling of the cultivator and physical characters are part of the genotype of the type of plant. [4]

This tree nut fruit species has worldwide variations in the genetic makeup which make it the most occurred polymorphic in nature. One of the multiple traits on which it can be diversified depends on the extent of amygdalin present in the kernel of the nut. Amygdalin is the cyanogenic glucoside compound made up of first one sweet glucose, second one responsible for the bitterness in taste benzaldehyde and third a toxic compound hydrogen cyanide (HCN) resulting from the physical change like crushing or chemical change like enzymatic hydrolysis. Upon all these ratios the sweet or bitter almond is classified. [5] Contemporary study commenced to estimate the therapeutic power of almond nut oil.

Discussing in detail about the nutrient profile it has macro nutrients enlisting carbohydrates, proteins and fats, micronutrients include many water-soluble vitamins like riboflavin, niacin, Vitamin E, many minerals like iron, calcium, potassium, copper, magnesium, phosphorus, manganese, zinc and selenium, fatty acids like saturated fatty acids (SFA), polyunsaturated fatty acid (PUFA) and monounsaturated fatty acid (MUFA). It also has phytosterol and tocopherol content endorsing its industrial consumption. [6]

Almond oil is scientifically proven to possess the health promoting benefits like the alleviating risk of cardiovascular disorder, maintaining the glucose homeostasis, reducing the oxidative stress, protection in the neurological functions and dermatologic promoting role making it more common in the cosmetic aspects. [2] Showing significantly promising results in the biomedical science and pharmacological industrial research, there are many more benefits to be explored in the medical science of oncology and gynecology of this very oil. Natural extracts isolated from the plant origin scientifically validate to exhibit the therapeutic and antioxidant capabilities. [7] A debate on the health promotion by almond oil came up with enormous outcomes like reduction in the risk of heart disease, a set dose may control postprandial glycemia, weight management role through satiety, anti-inflammatory and antioxidant roles and improvement in sports performance. [8–12]

Assistance in improving the lipid profile in dyslipidemia patients in moderation and declining the inflammatory and oxidative stress process make them susceptible and statistically associated for the maintaining body adiposity in overweight women. Nut oil is experimentally clinical trial proven to possess a novel cardio protective role by supplementing the nut oil at two different administered dosage of 7.2 mL/kg and other 14.4 mL/kg for time of ten days. This dose was reported to have a reduction in the platelet aggregation, decreased production in reactive oxygen species (ROS) and increased efficiency in the vascular function. [10,11]

The hemodialysis (HD) treatment taking patients of chronic kidney disorder (CKD) are reported to have disrupted dyslipidemias due to the variations in the standard concentrations like lesson high-density lipoprotein cholesterol concentrations (HDL-c) and raised up low-density lipoprotein cholesterol concentrations (LDL-c) and triglycerides concentrations (TG). [12] A similar study proposed a therapeutic effect of this Baru nut oil weighing 500 mg in comparison to same mineral oil in alleviating the gastrointestinal discomforts including constipation from 45.0% to 15.0% in these patients due to strict less fluid consumption, fewer ingestion of dietary fibers, iron supplementation orally, phosphate binders and most importantly compromised physical activity.

The purpose of this study is to produce technically Baru oil-supplemented drinks and food beverages like almond milk products with a special intent to promising a claim of a healthy heart future. Consumers on vegetarian diet or aiming to lessen the animal fat content like this plant-based milk diurnal. The contemporary study has been deliberated to inspect the influence of altered levels of Baru almond oil on blood serum lipoprotein standards.

Methodology

The present study was aimed at developing almond oil flavored milk. The control treatment was prepared from standardized milk at 3% fat. Other treatments were made at 3% fat with different concentrations of almond oil supplementation. The supplemented milk was analyzed for physicochemical and sensorial analysis. All trials were performed at Dairy Technology Laboratory of NIFSAT, University of Agriculture, Faisalabad, Pakistan.

Raw buffalo milk was purchased from the Dairy Farm of University of Agriculture Faisalabad. Other ingredients for the flavored milk were procured from a local supplier in Faisalabad.

Physicochemical analysis of milk: The moisture content of raw and flavored milk was measured by oven drying method. [13] The change in weight before and after drying was calculated as percentage.

$$\%\,Moisture = \frac{Sample\ weight\ before\ drying - Sample\ weight\ after\ drying}{Sample\ weight\ before\ drying} \times 100$$

pH: The milk pH was measured by using a portable pH meter (Hanna HI 991001). [14] The pH meter was calibrated before measurement using buffer solutions (pH 4 and 7). All the sample readings were noted in triplicate.

Acidity: The percent acidity of raw and supplemented milk was calculated by titration method (947.05) mentioned in Nagraik *et al.* [15]

$$\%\,Acidity\ (lactic\ acid) = \frac{mL\ of\ 0.1N\ NaOH\ \times 0.009}{Weight\ of\ sample} \times 100$$

Lipid and Fat: The fat content of raw and supplemented milk was estimated by Gerber method mentioned by McCarthy et al. [13]

Crude protein (CP): The crude protein of raw and supplemented milk was estimated by Kjeldhal method mentioned by Nagraik et al. [15]

$$\% \, Nitrogen = \frac{Vol. of \; 0.1N \; H_2SO_4 \; \times dilution \; factor \; \times 0.0014}{Weight \; of \; sample \; \times Vol. of \; distillation} \times 100$$

$$\% \, Protein = \% Nitrogen \times 6.38$$

Preparation of flavored milk: Flavored milk was manufactured by following the procedure as described by Loeffler et. al. 16 with some modifications to incorporate almond oil.

Extraction of oil: By using screw pressing extraction, almond oil would be extracted and added in milk.

Oil extraction: Almonds seeds were removed and conditioned to 10% moisture content. The moistened seeds were milled and passed through sieve to ensure equal particle size (3.2 mm). The moistened particles were mixed with fresh water to attain 12% moisture content. These samples were stored in an airtight container for 48 hours. The container was set to agitation at regular intervals for uniform distribution of moisture. The samples were placed in drying oven at 25 °C to set moisture content to 4%. Oil was extracted with screw press method at 20 and 40 °C with Komet Screw Press (IBG Monforts, Mönchengladbach, Germany).

Fat standardization: Buffalo milk was standardized at 2% and 3% by Pearson's square method to make flavored milk. Milk was standardized at 2% for control treatment (T0), whereas 3% for other treatments (T1, T2, and T3). [14]

Product Development: Buffalo milk was pasteurized at 65 °C for 30 minutes. After pasteurization almond oil is added in milk at different levels along with lecithin than milk is centrifuged to make its appearance better then milk is cooled at 4° C before use.

Table I: Experimental Plan

Treatments	**Almond oil %**
T_0	0
T_1	3
T_2	4
T_3	5

T_0 = Control (Standardized milk with 2% fat)

T_1 = Standardized flavored milk (2% fat) with 3% almond oil

T_2 = Standardized flavored milk (2% fat) with 4% almond oil

T_3 = Standardized flavored milk (2% fat) with 5% almond oil

Sensory Evaluation: The flavored milk was evaluated by a panel of judges for its sensorial characteristics (color, appearance, taste, flavor, overall acceptability) on nine-point hedonic scale. 17

Physicochemical analysis of flavored milk

Moisture content

The percent moisture content of raw and flavored milk was determined by oven drying method. 15

$$\% \, Moisture = \frac{W2 - W3}{W2 - W1} \times 100$$

Titratable acidity: The percent acidity of raw and supplemented milk was calculated by titration method (947.05) mentioned by McCarthy et. al. [13]

$$\% \, Acidity \; (lactic \; acid) = \frac{mL \; of \; 0.1N \; NaOH \; \times 0.009}{Weight \; of \; sample} \times 100$$

Lipid and fat content: The fat content of raw and supplemented milk was estimated by Gerber method mentioned by Jalilzadeh-Afshari and Fadaei. [18]

Crude protein (CP): The crude protein of raw and supplemented milk was estimated by Kjeldhal method mentioned by Jalilzadeh-Afshari & Fadaei. [18]

$$\% \, Nitrogen = \frac{Vol. of \; 0.1N \; H_2SO_4 \; \times dilution \; factor \; \times 0.0014}{Weight \; of \; sample \; \times Vol. of \; distillation} \times 100$$

$$\% \, Protein = \% Nitrogen \times 6.38$$

Blood test: Sample collection: The study was carried out on 20 albino rats. The experimental rats were housed in the animal room of Agriculture University, Faisalabad, Pakistan. The room temperature and relative humidity were set to 23+2 0 C and 55 + 5% respectively. The light and dark period was set to 12 hours alternatively.

Efficacy plan

Hypercholesterolemic Rats	
Group 1	Control (Standardized milk with 2% fat)
Group 2	Standardized flavored milk (2% fat) with 3% almond oil
Group 3	Standardized flavored milk (2% fat) with 4% almond oil
Group 4	Standardized flavored milk (2% fat) with 5% almond oil

High cholesterol diet: High cholesterol diet (1.5% cholesterol) for a specific period was fed to rats to raise their lipid profile i.e., cholesterol, low density lipoprotein (LDL), triglycerides and lower high-density lipoprotein (HDL). Afterwards, almond oil in milk was provided to rats to check the therapeutic effect.

Lipid profile: The lipid profile (cholesterol, low density lipoprotein, high density lipoprotein, triglycerides) of serum drawn from rats were evaluated as described by Mohammed Basheeruddin Asdaq *et. al.* [19]

The data obtained from all parameters were subjected to two-way factorial under completely randomized design to determine the level of significance. [20]

Results

The current study aimed to investigate the functional and nutraceutical properties of almond oil against hypercholesterolemia. The study was divided into four phases: compositional analysis of milk, physicochemical analysis of almond milk, almond milk preparation, conducting animal trial to assess therapeutic potential of almond milk.

Raw milk analysis: The raw milk was analyzed for its moisture, fat, protein, pH, and acidity. The results of raw milk analysis revealed 86%, 5.8%, 3.8%, 6.89%, 0.23% of moisture, fat, protein, pH, and acidity (Table III).

Product Development: Creating food products involves a multifaceted and dynamic process influenced by scientific challenges, consumer preferences, convenience, affordability, demographics, and cultural norms. As part of this process, a product called flavored milk was developed by incorporating almond oil. These products underwent thorough analysis to assess their physicochemical properties and sensory attributes.

Table III: Milk compositional and chemical analysis

Moisture	86 %
Fat	5.8 %
Protein	3.8 %
pH	6.89 %
Acidity	0.23 %

Proximate composition analysis of flavored milk: The proximate analysis of milk was calculated as 84%, 5.5%, 4.1%, 6.7%, and 0.20% for moisture, fat, crude protein, pH, and acidity respectively. Similarly, the proximate composition of almond milk varied across different treatments, with four treatments in total. In the control group (T_0), the percentages were 3.4%, 3.07%, 86%, 0.19%, and 0.42% for fat, protein, moisture content, acidity, and pH respectively. In T_1, the percentages were 81.72%, 3.54%, 4.87%, 0.49%, 6.61%, and 0.22% for the same parameters. T_2 and T_3 had percentages of 79.42%, 3.79%, 5.86%, 0.55%, 6.68%, 0.25%, 14.37%, 20.08% and 77.50%, 4.42%, 6.63%, 0.65%, 6.62%, 0.31%, 14.97%, 21.86% respectively, for moisture, crude protein, crude fat, ash, pH, acidity, SNF, and TSS.

Moisture: Moisture content stands as a crucial and frequently assessed characteristic across various food products, serving several purposes such as meeting legal standards, complying with labeling regulations, ensuring economic viability, maintaining food quality, facilitating efficient processing operations, and ensuring storage stability. Analyzing moisture content is particularly vital in evaluating the quality of finished goods, as it not only ensures compliance with product specifications but also impacts textural attributes, solubility, and shelf life. [21].In the case of milk, the moisture content exhibited a notable decrease across all four treatments. The addition of almond oil to milk significantly influenced its moisture content, as evidenced by mean values of 85%, 82.66%, 80.33%, and 78.33% for T_0, T_1, T_2, and T_3 respectively. These findings align with those of Yada et al. [22] who similarly noted a decrease in the moisture content of almond milk with increasing almond oil content.

pH: The pH levels of milk exhibit a notable decrease across all four treatments, as indicated in Table 4. Treatment T_3 demonstrates the lowest pH value for milk, while the highest is observed in T_0. The incorporation of almond oil into milk significantly impacts its pH. Mean pH values for T_0, T_1, T_2, and T_3 are 6.79, 6.71, 6.68, and 6.62 respectively.

Acidity: The acidity levels of milk display a notable increase across all four treatments, as detailed in Table IV. Treatment T_3 exhibits the highest acidity value for milk, while the lowest is observed in T_0. The addition of almond oil to milk significantly influences its acidity. Percent acidity was calculated as 0.19%, 0.22%, 0.25%, and 0.31% for T_0, T_1, T_2, and T_3 respectively.

Fat: Mean values for fat content in flavored almond milk were presented in Table 4. T_3 exhibits the highest fat content for milk, while the lowest is observed in T_0. The addition of almond oil to milk significantly impacts its fat content. Mean values for treatments T_0, T_1, T_2, and T_3 are 3.15%, 4.87%, 5.86%, and 6.63% respectively.

Crude protein: Protein plays a crucial role in providing flavor, body, and texture to food products, making its determination highly important. Rich in essential amino acids, protein is

Table IV: Mean values for physicochemical analysis of almond milk.

Treatment	Moisture	Protein	Fat	Ash	pH	Acidity	SNF	TSS
T_0	84^{A}	3.54^{D}	3.15^{D}	0.38^{C}	6.79^{A}	0.19^{D}	13.52^{B}	16.05^{D}
T_1	81.72^{B}	3.64^{C}	4.87^{C}	0.49^{B}	6.71^{B}	0.22^{C}	13.92^{B}	17.78^{C}
T_2	79.42^{C}	3.79^{B}	5.86^{B}	0.55^{B}	6.68^{BC}	0.25^{B}	14.37^{AB}	20.08^{B}
T_3	77.50^{D}	4.42^{A}	6.63^{A}	0.65^{A}	6.62^{C}	0.31^{A}	14.97^{A}	21.86^{A}

essential for maintaining our health and contributes to the synthesis of stable air cells, which enhances the texture of the product. Additionally, it serves as an energy source for the body. The crude protein content in milk exhibits a significant increase across all four treatments. Mean values of protein content in flavored almond milk were presented in Table 4. T_3 exhibited the highest value (4.43%) whereas T0 showed the lowest (3.54%).

Ash: Mean ash content values for almond milk are provided in Table IV with T_3 exhibiting the highest value and T_0 the lowest. The addition of almond oil to milk significantly increased the ash content.

Solid not Fat (SNF) and Total Soluble Solids (TSS): The solid-not-fat (SNF) content of milk demonstrates a significant increase across all 4 treatments. Mean SNF values for almond milk are presented in Table 1, with T3 exhibiting the highest value and T_0 the lowest. The addition of almond oil to milk notably influences the SNF content, with mean values for T_0, T_1, T_2, and T_3 being 13.52%, 13.92%, 14.37%, and 14.97% respectively. Similarly, the total soluble solids (TSS) content of milk also displays a significant increase across all treatments. Mean TSS values for almond milk are detailed in Table 4, with T_3 displaying the highest value and T_0 the lowest.

Mean sharing different letter in a column are statistically significant (p>0.05)

T_0 = Control (Standardized milk with 2% fat)

T_1 = Standardized flavored milk (2% fat) with 3% almond oil

T_2 = Standardized flavored milk (2% fat) with 4% almond oil

T_3 = Standardized flavored milk (2% fat) with 5% almond oil

Sensory evaluation of flavored milk: Sensory analysis of almond milk with a 9-point hedonic scale consist of color, flavor, appearance, taste, and overall acceptability has been presented in Figure 1. It is illustrated in Figure 1 that the highest score for color was observed in T_0, whereas T_1, T_2, and T_3 showed highest score for flavor, taste, appearance, and overall acceptability. According to the hedonic scale responses, T_2 (4% almond oil) and T_3 (5% almond oil) received high scores. Previous research suggests that almond inclusion not only enhances the sensory properties of the almond milk but also decreases the fat level.

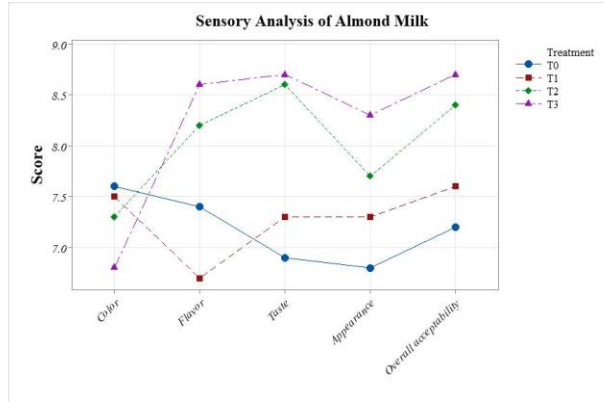

Figure 1: Sensorial analysis of almond milk. T_0: Control (Standardized milk with 2% fat), T_1: Standardized flavored milk (2% fat) with 3% almond oil, T_2: Standardized flavored milk (2% fat) with 4% almond oil, T_3: Standardized flavored milk (2% fat) with 5% almond oil

Table V: Mean values for cholesterol in blood

Treatments	Days		Mean
	0	45	
T_0	69.45	67.6	68.52^C
T_1	113.52	107.35	110.45^A
T_2	108.54	95.77	102.15^B
T_3	106.58	92.35	99.46^B
Mean	99.52^A	90.76^B	

Efficacy studies: A study was carried out to examine the potential health benefits of almond oil through an efficacy trial. Because it is easier to manage and analyze each parameter in an animal study than in human studies, where finding volunteers willing to adhere to a specific diet for an extended period is challenging, an animal study employing albino rats was undertaken for this purpose. The respective groups of rats were fed a specific diet to induce hypercholesterolemia for a defined period. Subsequently, a diet (functional and nutraceutical) was administered for a duration of 45 days to evaluate its potential to mitigating disorders related to lifestyles like hypercholesterolemia. Values were recorded at the beginning of the study, and at the end of the trial, sera collected from the rats were analyzed for lipid profile.

Cholesterol: The cholesterol levels observed over the span of 45 days yielded highly significant results, while the combined influence of treatment and duration did not show significant outcomes. Detailed mean cholesterol values are presented in Table V. Notably, treatment T_3 exhibited a considerable decrease in cholesterol levels in the blood samples. During the investigation, a significant reduction of cholesterol in T_1 (113.52- 107.35), T_2 (108.54- 95.77), and T_3 (106.58- 92.35) were seen compared to control group T_0 (69.45- 67.6). Whereas T_3 showed discernible reduction of cholesterol compared to T_1, T_2 groups.

High density lipoproteins (HDL): The findings indicate significant variations in HDL levels among all treatments. Over the 45-day period, HDL levels exhibited highly significant changes, although the combined impact of treatment and duration did not yield significant results. Detailed mean HDL values are presented in Table VI. Notably, treatment T_3 demonstrated an elevated HDL level in the blood samples. The HDL values in all four groups were observed and it was seen that T_3 showed the highest significant effects compared to T_1, T_2 and T_0 groups. This increase in HDL levels in T_3 can be attributed to the higher percentage of almond oil in the milk.

Low density lipoprotein (LDL): The average LDL value is detailed in Table 7. Blood samples indicated a significant reduction of LDL level in T_3 group dropping from 59.2 to 37.3 compared to T_2 (53.7-46.7), and T_1 (61.8 – 54.1), where a slight reduction of LDL observed. LDL levels in the blood ranged from 36.5 to 59.2 across all treatments.

Triglycerides: The triglyceride levels over 45 days displayed significant changes, while the combined influence of treatment and duration yielded nonsignificant outcomes. The average triglyceride value is presented in Table 8. Blood samples indicated slightly lower triglyceride levels in treatments T_2 and T_3. Triglyceride levels in the blood ranged from 96.8 to 117.8 across all four treatments. Treatment means were recorded as 96.8, 135.8, 129.6, and 117.3 for T_0, T_1, T_2, and T_3, respectively. T_3 exhibited the significant reduction in triglyceride levels due to the highest amount of oil in milk.

Mean sharing different letter in a column are statistically significant (p>0.05)

T_0 = Control (Standardized milk with 2% fat)

T_1 = Standardized flavored milk (2% fat) with 3% almond oil

T_2 = Standardized flavored milk (2% fat) with 4% almond oil

T_3 = Standardized flavored milk (2% fat) with 5% almond oil

Table VI: Mean values for high density lipoprotein (HDL) in blood.

Treatment	Day		Mean
	0	45	
T_0	37.5	39.4	38.45^{A}
T_1	21.3	25.3	23.3^{C}
T_2	20.8	26.2	23.5^{C}
T_3	18.8	34.1	26.45^{B}
Mean	24.6^{B}	31.25^{A}	

Mean sharing different letter in a column are statistically significant (p>0.05)

T_0 = Control (Standardized milk with 2% fat)

T_1 = Standardized flavored milk (2% fat) with 3% almond oil

T_2 = Standardized flavored milk (2% fat) with 4% almond oil

T_3 = Standardized flavored milk (2% fat) with 5% almond oil

Table VII: Mean values for LDL (low density lipoprotein) in blood.

Treatments	Days		Mean
	0	45	
T_0	36.5^{c}	38.6^{c}	37.55^{D}
T_1	61.8^{a}	54.1^{ab}	57.95^{A}
T_2	53.7^{ab}	46.7^{ab}	50.2^{B}
T_3	59.2^{ab}	37.3^{bc}	48.25^{C}
Mean	52.8^{A}	44.17^{B}	

Mean sharing different letter in a column are statistically significant (p>0.05)

T_0 = Control (Standardized milk with 2% fat)

T_1 = Standardized flavored milk (2% fat) with 3% almond oil

T_2 = Standardized flavored milk (2% fat) with 4% almond oil

T_3 = Standardized flavored milk (2% fat) with 5% almond oil

Table VIII: Mean values for triglycerides in blood

Treatments	**Days**		**Mean**
	0	**45**	
T_0	93.8	96.8	95.3^{C}
T_1	134.5	135.8	135.15^{A}
T_2	135.6	129.6	132.6^{AB}
T_3	138.7	117.3	128^{B}
Mean	125.65^{A}	119.87^{B}	

Mean sharing different letter in a column are statistically significant (p>0.05)

T_0 = Control (Standardized milk with 2% fat)

T_1 = Standardized flavored milk (2% fat) with 3% almond oil

T_2 = Standardized flavored milk (2% fat) with 4% almond oil

T_3 = Standardized flavored milk (2% fat) with 5% almond oil

Discussion

In the case of milk, the moisture content exhibited a notable decrease across all four treatments. The addition of almond oil to milk significantly influenced its moisture content, as evidenced by mean values of 85%, 82.66%, 80.33%, and 78.33% for T_0, T_1, T_2, and T_3 respectively. These findings align with those of Yada *et al.* [22], who similarly noted a decrease in the moisture content of almond milk with increasing almond oil content. Protein content of flavored milk was supported,[23] where they evaluated the chemical composition of various almond varieties grown worldwide, revealing substantial differences in macro and micro constituents among different almond varieties. The chemical analysis of five major US-grown almond varieties—Carmel, nonpareil, mission, neplus, and peerless—revealed moisture content ranging from 4.35% to 5.86%, protein content from 16.42% to 22.17%, lipid content from 53.59% to 56.05%, ash content from 2.69% to 2.93%, and pH ranging from 6.0 to 6.8%.

Percent acidity of flavored milk was supported by Yoo *et al.* [24] and align with the research conducted by Ahrens *et al.* [25], which reported a range of acidity content in almond oil from 1.38% to 3.55%. [26] Fat serves multiple functions in food, including contributing to taste, texture, and appearance. These findings are consistent with those of Yoo *et al.* [24] and are in line with the research conducted by Aherns *et al.* [25], which reported a range of fat content in almond oil. The addition of almond oil to milk significantly influences the protein content. These findings are supported by research conducted by Cherif *et al.* [27] The ash content of food indicates the residual inorganic material left after the organic components have been incinerated. However, the ash content obtained may not precisely mirror the original mineral composition of the food, as certain minerals may have altered during the process. [28] This finding is consistent with the observations of Ozcan *et al.* [29], who noted ash content ranging from 2.74% to 3.05% in various almond varieties. As mineral matter is inherently present in food, external factors such as temperature and humidity have minimal influence over time. The incorporation of almond oil into milk substantially impacts the TSS content. Comparable findings were reported by Aherns *et al.* [25], indicating that almond milk contained 10.2% fat, 4.3% protein, and 22.74% TSS. Sensorial characteristics are in consistent with those of Jia *et al.* [30]

Wang *et al.* [31] highlighted the suppressive effects of bioactive compounds in almonds on HMG CoA reductase, an enzyme which could synthesized and absorbed cholesterol. This mechanism helped to activate the LDL receptors and raised the amount of cholesterol in feces. This finding aligns with previous research conducted by Hyson *et al.* [32], where the addition of almond extract to the diets of healthy individuals led to a discernible increase in HDL cholesterol and a significant decrease in LDL cholesterol, triglycerides, and total cholesterol. Monounsaturated fats in almonds were found to be responsible for these effects, which enhanced energy levels by 54% without increasing the amount of saturated. Anwar *et al.* [33] in their 2013 study examined the antiatherogenic characteristics of almond oil on serum lipid profile of diabetic rats. Upon completion of the trial, blood samples were collected, revealing an improvement in HDL-C concentration from 45.3 ± 0.5 to 49.8 ± 1.3 mg/dl. This improvement was attributed to the activation of receptor cells, which facilitate the incorporation of excess LDL and triglycerides into the liver and adipose tissue, thereby preventing their circulation in the vascular system and the development of atherosclerosis. Low density lipoprotein findings align with those of Berryman *et al.* [34], who highlighted the role of bioactive almond components in reducing the risk of hypercholesterolemia by lowering LDL-C levels. They identified phytosterols as key bioactive components in almonds, which inhibit LDL-C oxidation (14.0 ± 3.8%) by binding to it, thereby reducing the incidence of coronary heart disorders. Additionally, the study emphasized the significant role of fiber and the fat content of almonds. They observed, specifically, that the monounsaturated (oleic acid 64–82%) and polyunsaturated fats found in almonds substitute the saturated fat and only slightly aid in the production of LDL cholesterol. Triglycerides findings are consistent with the research conducted by Emekli *et al.* [35], who examined the impact of nuts on rats' serum lipid profiles. Upon analysing blood samples for biochemical analysis at the end of the 12 weeks, they observed a decreasing trend in triglyceride levels, which ranged from 294.38 ± 27.41 to 282.75 ± 28.23.

Conclusion

The comprehensive discussion suggests that almonds hold significant utility as a nut, particularly in their conversion to almond oil and other pharmaceutical and dietary applications. Leveraging almond oil in therapeutic interventions can effectively mitigate lifestyle-related disorders such as hypercholesterolemia, as indicated by current research. Notably, rats administered with 4 and 5% almond oil in milk demonstrated improved serum lipid profiles. Thus, it is recommended to incorporate almonds into functional and nutraceutical food products as therapeutic agents for targeted populations.

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