fermentation

Article

Effect of Fermented Camel Milk Containing Pumpkin Seed Milk on the Oxidative Stress Induced by Carbon Tetrachloride in Experimental Rats

Magdy Ramadan Shahein [1], El Sayed Hassan Atwaa [2], Barakat M. Alrashdi [3], Mahytab Fawzy Ramadan [2], El Sayed Abd El-Sattar [4], Adel Abdel Hameed Siam [5], Mohamed A. Alblihed [6] and Ehab Kotb Elmahallawy [7,*]

1 Department of Food Science and Technology, Faculty of Agriculture, Tanta University, Tanta 31527, Egypt; magdrsh10@gmail.com
2 Food Science Department, Faculty of Agriculture, Zagazig University, Zagazig 44511, Egypt; elsayedhassanattwa@gmail.com (E.S.H.A.); ahetabhassanien@hotmail.com (M.F.R.)
3 Biology Department, College of Science, Jouf University, Sakaka 72388, Saudi Arabia; bmalrashdi@ju.edu.sa
4 Department of Food and Dairy Technology, Faculty of Technology and Development, Zagazig University, Zagazig 44511, Egypt; e.abdelsattar82@gmail.com
5 Department of Physiology, Faculty of Veterinary Medicine, Suez Canal University, Ismailia 41522, Egypt; adel_soliman@vet.suez.edu.eg
6 Department of Microbiology, College of Medicine, Taif University, P.O.Box. 11099, Taif 21944, Saudi Arabia; mabulihd@tu.edu.sa
7 Department of Zoonoses, Faculty of Veterinary Medicine, Sohag University, Sohag 82524, Egypt
* Correspondence: eehaa@unileon.es

Citation: Shahein, M.R.; Atwaa, E.S.H.; Alrashdi, B.M.; Ramadan, M.F.; Abd El-Sattar, E.S.; Siam, A.A.H.; Alblihed, M.A.; Elmahallawy, E.K. Effect of Fermented Camel Milk Containing Pumpkin Seed Milk on the Oxidative Stress Induced by Carbon Tetrachloride in Experimental Rats. *Fermentation* **2022**, *8*, 223. https://doi.org/10.3390/fermentation8050223

Academic Editor: Carlo Giuseppe Rizzello

Received: 30 March 2022
Accepted: 6 May 2022
Published: 13 May 2022

Abstract: Oxidative stress can lead to chronic inflammation, nephrotoxicity, and renal damage. The consumption of plant-based dairy alternatives has increased rapidly worldwide due to their health effects. Bioactive components from natural sources, such as plants, are effective in protecting against oxidative stress. The present study evaluated the physicochemical and sensory properties of fermented camel milk made from camel milk mixed with pumpkin seed milk. Fermented camel milk consists of camel milk mixed with 25% and 50% pumpkin seed milk. This blend (fermented camel milk containing 50% pumpkin seed milk) was evaluated as an antioxidant agent in oxidative stress induced rats. A total of thirty-two male adult albino rats of Sprague Dawley® Rat strain weighing 150–180 g were randomly divided into four groups ($n = 8$). The first group was solely administered the standard diet and served as the negative control. The other rats ($n = 24$), received a basal diet, including being intraperitoneally injected with carbon tetrachloride, with a single dose at a rate of 2 mL/kg body weight) as a model for oxidative stress. The oxidative stress rats were divided into three groups; the first group did not receive any treatment and served as the positive control. The second and third groups were administered 10 g/day fermented camel milk and fermented camel milk containing 50% pumpkin seed milk. The results revealed that mixing the camel milk with pumpkin seed milk was more effective in increasing the total solids, protein, ash, fiber, acidity, viscosity, phenolic content, and antioxidant activity. These enhancements were proportional to the mixing ratio. Fermented camel milk containing 50% pumpkin seed milk exhibited the highest scores for sensory properties compared with the other fermented camel milk treatments. The group of rats with oxidative stress treated with fermented camel milk containing 50% pumpkin seed milk showed a significant decrease ($p \leq 0.05$) in the levels of malondialdehyde (MDA), low-density lipoprotein (LDL), cholesterol (CL), triglycerides (TGs), AST, ALT, creatinine, and urea, and increased ($p \leq 0.05$) high-density lipoprotein (HDL) and total protein and albumin compared with rats with oxidative stress. Consumption of fermented camel milk containing 50% pumpkin seed milk by the oxidative stress rat groups caused significant improvement in all of these factors compared with the positive control group. This study revealed that the administration of fermented camel milk containing 50% pumpkin seed milk to rats with oxidative stress prevented disorders related to oxidative stress compared with the untreated oxidative stress group. Thus, incorporating fermented camel milk might play a beneficial role in patients with oxidative stress.

Keywords: oxidative stress; pumpkin seed milk; camel milk; physicochemical; carbon tetrachloride; rats

1. Introduction

Milk and its products are considered essential foods for human development and can benefit in the oxidative defense of consumers. Furthermore, milk and dairy products with protective properties have the potential to act as coadjuvants in metabolic disorders, intestinal health, conventional therapies, addressing cardiovascular diseases, and chemopreventive properties [1]. Among others, camel milk has high nutritional and health value as it contains immune proteins such as lysozyme (which is an antioxidant and anti-inflammatory compound) and aminoglobulins, but no beta-lactoglobulin, which may cause allergic reactions in some people. It contains iron, potassium, and vitamins C, E, and A [2,3]. Most camel milk is traditionally consumed fresh as raw milk or when soured. However, recently, fresh pasteurized camel milk has been made available in the Middle East and Arab lands [4]. Importantly, Camel milk contains more free amino acids and peptides than cow's milk [5]. Moreover, non-protein-bound amino acids in camel milk are easily digested by microorganisms, thus, camel milk has higher metabolic activity when used in the starter culture preparation [6]. Fermented camel milk has several health benefits: antioxidant activity, angiotensin-converting enzyme inhibitory activity, a hypocholesterolemic effect, antimicrobial activity, anti-diarrhea activity, and anticancer activity [7]. There is increased attention to improving fermented dairy products as a result of their health benefits [8–16].

Oxidative stress is an imbalance between the production of reactive oxygen species and their elimination by protective mechanisms, which can lead to chronic inflammation [17]. Oxidative stress can activate various transcription factors, thus leading to the differential expression of various genes involved in inflammatory pathways [18,19]. The inflammation triggered by oxidative stress causes many chronic diseases [20]. Polyphenols might be useful as adjuvant therapy based on their potential anti-inflammatory effect, associated with their antioxidant activity and inhibition of enzymes involved in the production of eicosanoids [21,22]. Moreover, the kidneys are vital organs that are essential for the excretion of metabolic wastes and maintaining chemical homeostasis, among other functions [23]. Several studies have reported oxidative stress as a potential cause of different forms of renal damage and nephrotoxicity. Increased oxidative stress caused by free radical generation is a likely result of inflammatory responses [24]. Adding supplementary antioxidants from natural sources, such as plants, is efficient in protecting against oxidative stress. This may prompt more food manufacturers to replace synthetic antioxidants with ingredients containing natural antioxidative compounds. Therefore, natural additives have gained increased attention as they pose no health risk to consumers [25]. The antioxidant protection mechanism acting against the reactions of free radicals comprises enzymatic and non-enzymatic elements, part of which are synthesized in plants exclusively, and the body can obtain them only from foods [26]. Several antioxidants, e.g., flavonoids, phenolic acids, tannins, vitamin C, and vitamin E, interact synergistically with other reducing compounds and have diverse biological properties, such as anti-inflammatory, anti-carcinogenic, and anti-atherosclerotic effects [21]. Moreover, foods that are rich in bioactive compounds are advantageous as they boost the immune system.

It is noteworthy to state that cereals and their components have been accepted as functional foods as they provide the vitamins, dietary fiber, protein, antioxidants, energy, and minerals required for human health [8,10–16]. Moreover, cereals can be fermentable substances for the growth of probiotic bacteria [27,28]. Pumpkin (*Cucurbita pepo* L.) is a plump nutritious vegetable that belongs to the *Cucurbitaceae* family, with several varieties grown throughout the world, e.g., from South–Central America, to Mexico, Argentina, Chile, Europe, Asia (India and China), and Western America [29–31]. The pumpkin seed oil has been used since the end of the 19th century to treat urinary tract problems [32]. Pumpkin contains various biologically active components, such as sterols, proteins, peptides, polysaccharides, para-aminobenzoic acid [33], carotenoids, and γ-aminobutyric acid [34]. In addition, pumpkin seeds have a high protein content and essential fatty acids [35], mainly linoleic acid, stearic acid, oleic acid, and palmitic acid. Furthermore, it contains non-essential amino acids and phytosterols, e.g., sitosterol and stigmasterol [36]. Moreover, pumpkin seeds contain tocopherol (vitamin E), microelements (Na, K, and Cr), [37,38], and phenolic compounds, such as coumarins, flavonoids, pigments, pyrazine, and triterpenoids [39–41]. Importantly, a pumpkin seed extract promotes wound healing and has anti-arthritis, hair-growth-stimulating [42], anthelmintic [43], antitumor [44], hepatoprotective [45], and antioxidant effects [46]. In addition, the therapeutic activities of pumpkin seed extracts include the relief of symptoms associated with urinary bladder complications and prostate disorders [47]. A previous study [48] explained that pumpkin seeds and their different types are of high nutritional value as they are a good source of protein, fats, fiber, minerals, squalene, tocopherols and β-carotene, and therefore can be used as a good nutritional source in the food industry. Pumpkin seeds also have good antimicrobial properties and can be used for medicinal purposes. The chromatographic analysis showed Pumpkin seeds contain many phenolic compounds such as protocatechuic acid, myricetin, injectable acid, vanillin, quercetin, hydroxybenzoic acid, apigenin, etc., which are famous for their anti-inflammatory and anti-cancer activity. Revising the available literature, no information is available about the potential effect of adding pumpkin seed milk to fermented camel milk manufacturing. The present study aimed to investigate the impact of incorporating pumpkin seed milk into fermented camel milk manufacture, to fortify different proportions of fermented camel milk regarding its functional properties. Furthermore, the fortified fermented camel milk was tested for its therapeutic effect as a functional food in rats with high oxidative stress induced by carbon tetrachloride.

2. Material and Methods

2.1. Materials and Reagents

Fresh bulk camel milk was obtained from the Desert Research Center, Dokki, Egypt. Pumpkin seeds were obtained from the Crops Research Institute, Agricultural Research Center, Ministry of Agriculture Giza, Egypt. All solvents used for extraction and analyses were of analytical grade. Folin–Ciocalteu, gallic acid, and 2,2-diphenyl-1-pic-rylhydrazyl (DPPH) were purchased from Sigma (St. Louis, MO, USA). The kits that were used for the biochemical analyses were from Gamma Trade Company (Cairo, Egypt). The protocol reported by Reeves et al. 1993 [49] was used to prepare the standard basal diet. The yogurt cultures that were added included *Lactobacillus delbruekii* subsp. *bulgaricus* and *Streptococcus salivarius* subsp. *thermophilus* were purchased from the Ain Shams University, Agriculture Faculty, Microbiological Resources Center, Egypt.

2.2. Preparation of Pumpkin Seed Milk (PSM)

PSM was prepared as per Hassan et al., 2012, with some modifications [50]. Seeds (100 g) were cleaned, dehulled, and soaked in water at a 1:4 w/v ratio overnight at room temperature. The soaked seeds were blended in a grinder. The resulting emulsion was then filtered through a double-layer muslin cloth, boiled for 5 min with constant stirring, cooled to room temperature (22 °C), and kept at a refrigerator temperature before product formulation was used within 24 h.

2.3. Fermented Camel Milk Manufacture

Fermented camel milk was manufactured according to the method reported by Tamime and Robinson, 1999 [51]. Raw camel milk was divided into three equal portions; the first portion served as the control (C), the second portion was mixed with 25.0% PSM (T1), and the third portion was mixed with 50.0% PSM (T2). The milk used for all treatments was homogenized at 55 °C–60 °C for 2 min using a high-speed mixer (22,000 rpm), heat-treated in a thermostatically controlled water bath at 85 °C for 30 min, cooled to 42 °C in an ice bath, inoculated with 5% (w/v) yogurt culture, and incubated at 42 °C until a firm curd was obtained (around 12 h). The curd was then refrigerated at 4 °C overnight, stirred using the mixer, stored at 4 °C ± 1 °C, and analyzed 1 day after manufacture for its physicochemical and sensory properties.

2.4. Chemical Composition, Physicochemical Analysis, and Sensory Evaluation of the Fermented Camel Milk Treatments

In the fermented camel milk samples, the total solids, fat, protein, ash amounts, and titratable acidity were determined as described elsewhere [52]. The total solids of camel milk, pumpkin seed milk and fermented camel milk samples were determined using a drying oven for 24 h at 100 °C. The percentage of moisture content was calculated by the following formula.

$$\% \text{ moisture} = W1 - (W2 \times 100)/W1$$

where, W1 = initial weight of sample; W2 = weight of the dried sample

$$\text{Total solids } \% = 100 - \text{moisture content}$$

For fat content of camel milk and fermented camel milk samples, the Gerber method was applied, whereas for pumpkin seed milk, the Soxhelt apparatus method was used. The total nitrogen content (TN) was determined using the micro-Kjeldahl method, and protein content was calculated by multiplying the percentage of TN by 6.38 for milk components and 6.25 for pumpkin seed milk. To determine ash content, a 5 g sample was heated in a muffle furnace at 550 °C overnight. The titratable acidity, expressed as a percentage of lactic acid (%), was determined by mixing 10 g of fermented camel milk with 10 mL of distilled water and titrating with 0.1 N NaOH using phenolphthalein as an indicator to an end-point of faint pink color. Their pH values were monitored using a pH meter equipped with a glass electrode (HANNA, Instrument, Portugal). The viscosity of the fermented camel milk was determined according to Aryana, 2003 [53] using Rotational Viscometer Type Lab. Line Model 5437. Measurements were determined at a temperature of 30 °C after 15 s; the results were expressed as centipoise (cP). The total phenolic content (TPC; as mg GAE (gallic acid equivalents)/100 g) and antioxidant (AO) activity (%) of the prepared fermented camel milk treatments were assessed according to Maksimović et al. 2008 [54] and Apostolidis et al. 2007 [55], respectively. The sensory evaluation of the fermented camel milk treatments was carried out by ten trained panelists as described elsewhere [28].

2.5. Experimental Design of the Biological Study

A total of 32 male adult Sprague Dawley albino rats (weight range, 150–180 g) were provided by the Agricultural Research Center of Giza (Egypt) and housed in wire cages at 25 °C. After acclimation to a basal diet for 7 days, they were divided randomly into four groups (eight rats n each group), as follows: eight rats were kept as the normal control and were fed a standard diet as the negative control (group 1). Twenty-four rats were intraperitoneally injected with a single dose of carbon tetrachloride at a 2 mL/kg of body weight (oxidative stress rats), as described elsewhere [56]. Oxidative stress rats were then organized into three groups (eight rats in each group), as follows: oxidative stress rats as a positive control (group 2), oxidative stress rats fed a basal diet with 10 g/day plain fermented camel milk via an epigastric tube (group 3), and oxidative stress rats fed a basal diet with 10 g/day PSM-fermented camel milk (fermented camel milk manufactured from blended milk; i.e., 50% camel milk and 50% PSM) via a epigastric tube (group 4).

2.6. Biochemical Analysis

After five weeks, the rats were lightly anesthetized with diethyl ether. Blood samples were collected from the hepatic portal vein, submitted to centrifugation (to separate the serum) at 3000 rpm for 15 min, and stored at −40 °C. Caraway's method [57] was used to determine serum uric acid. In contrast, the serum creatinine level was measured using Bonsens' method [58], and serum urea was determined according to Marsch et al., 1965 [59].

Malondialdehyde (MDA) was determined in the serum according to Kei, 1978 [60]. The alanine amino transferase (ALT) and aspartate amino transferase (AST) enzymes were measured according to the methods described by Bergmeyer and Harder, 1986 [61]. The total cholesterol was determined according to the method of the Enzymatic Colorimeter [62]. Total lipids and triglycerides were determined according to the method of Devi and Sharma, 2004 [63]. LDL-cholesterol was calculated using the Friedewald formula [64], as follows:

$$\text{LDL-cholesterol} = \text{total cholesterol} - (\text{HDL-cholesterol}) - (\text{triglycerides}/5) \quad (1)$$

2.7. Statistical Analysis

The obtained data were statistically evaluated using analysis of variance, as reported by McClave and Benson, 1991 [65]. All data were subjected to one-way analysis of variance (ANOVA) software. Significant treatment means were separated by Duncan's new multiple range tests. Differences were considered significant at ($p \leq 0.05$).

3. Results and Discussion

3.1. Chemical Composition of Raw Camel Milk and PSM

Table 1 presents the approximate chemical composition of the PSM compared with the camel milk. The PSM had a higher protein content (7.84% vs. 3.94%), whereas it had a much higher fat content (6.90%) than camel milk (3.12%). The PSM was characterized by the unique presence of fibers and much higher levels of TPC (mg GAE/100 g) and DPPH inhibition % activity than present in the camel milk, i.e., 234.20 and 56.7 vs. 4.60 and 12.86, respectively. These results of approximate chemical composition are in line with those reported previously for PSM and camel milk [50,66]. The low TPC detected in camel milk was consistent with Bouhaddaoui et al., 2019 [67], who reported a level of 35.54 mg GAE/L. The high phenol content of PSM agreed with Peiretti et al., 2017 [68], who reported that pumpkin seeds contain a higher total phenolic level.

Table 1. Chemical composition of fresh camel milk and pumpkin seed milk.

Components (%)	Camel Milk	Pumpkin Seed Milk
Total Solids	14.14 ± 0.46^{b}	21.60 ± 1.04^{a}
Protein	3.94 ± 0.14^{b}	7.84 ± 0.82^{a}
Fat	3.12 ± 0.18^{b}	6.90 ± 0.55^{a}
Ash	0.82 ± 0.05^{b}	0.92 ± 0.04^{a}
Fiber	0.00^{b}	3.42 ± 0.24^{a}
Total phenolic content (mg/100 g)	3.62 ± 0.28^{b}	234.20 ± 5.62^{a}
DPPH inhibition % activity	12.86 ± 0.92^{b}	56.70 ± 2.14^{a}

Values (means ± SD) with different superscript letters are statistically significantly different ($p \leq 0.05$).

3.2. Chemical, Physicochemical, and Phytochemical Properties of Fermented Camel Milk Containing PSM

Table 2 shows the chemical composition of fermented camel milk containing PSM. Control fermented camel milk had a lower total solid (TS), protein, fat, ash, and fiber content. This difference was significant ($p \leq 0.05$) compared with the fermented camel milk containing PSM treatments. The TS, protein, fat, ash, and fiber content of fermented camel milk containing PSM increased gradually by increasing the mixing ratio, which was attributed to the high TS, protein, fat, ash, and fiber content of the prepared PSM compared with the camel milk used in this study (Table 1). These results are in agreement with those reported by Atwaa et al., 2020 [28], who found that partial replacement of camel milk with oat milk, of up to 40%, increased the TS, protein, ash, and carbohydrate content of the resultant fermented camel milk compared with camel milk yogurt. Moreover, the data illustrated in Table 2 indicated that the titratable acidity (TA) of the control fermented camel milk had the lowest value, which might be attributable to the high level of antimicrobial components, such as lysozyme, lactoferrin, and immunoglobulins in camel milk, which decreased the viability of the starter culture [69]. The acidity of fermented camel milk made from camel milk mixed with PSM increased gradually by increasing the mixing ratio; this may be attributed to the hypothesis that the PSM fermentable substance improves the viability of the starter culture [70].

Table 2. Chemical, physicochemical, and phytochemical properties of fermented camel milk containing pumpkin seed milk.

Items	Treatments		
	C	**T_1**	**T_2**
Chemical composition (%)			
Total Solids	14.22 ± 0.86 [c]	15.94 ± 0.74 [b]	16.54 ± 0.48 [a]
Protein	3.98 ± 0.32 [c]	4.92 ± 0. 48 [b]	5.86 ± 0.28 [a]
Fat	3.18 ± 0.24 [c]	4.04 ± 0.12 [b]	4.96 ± 0.32 [a]
Ash	0.85 ± 0.06 [c]	0.87 ± 0.04 [b]	0.90 ± 0.03 [a]
Fiber	0.00 [c]	0.82 ± 0.03 [b]	1.55 ± 0.09 [a]
Physicochemical properties			
Acidity [as lactic acid percentage (%)]	0.75 ± 0.04 [c]	0.80 ± 0.02 [b]	0.84 ± 0.03 [a]
pH values	4.82 ± 0.06 [a]	4.76 ± 0.04 [b]	4.68 ± 0.05 [c]
Viscosity (cP)	2160 ± 54 [c]	2420 ± 84 [b]	2590 ± 98 [a]
Phytochemical properties			
Total phenolic content (mg/100 g)	4.06 ± 0.22 [c]	58.54 ± 1.64 [b]	108.65 ± 4.42 [a]
DPPH inhibition % activity	13.40 ± 0.34 [c]	15.24 ± 0.56 [b]	18.12 ± 0.74 [a]

Values (means ± SD) with different superscript letters are statistically significantly different ($p \leq 0.05$). C: fermented camel milk made from camel milk as a control (C); T_1: fermented camel milk made from camel milk mixed with 25% pumpkin seed milk; T_2: fermented camel milk made from camel milk mixed with 50% pumpkin seed milk; cP:centipoise (Unit of dynamic viscosity).

Furthermore, the addition of vegetarian milk to camel milk reduces the concentration of the components of camel milk. The pH values of all treatments exhibited an opposite trend to that of TA. Similar results were obtained by Atwaa et al., 2020 [28], who found that partial replacement of camel milk with oat milk increased the TA and decreased the pH values of the resultant camel milk yogurt. The mixing of camel milk with PSM greatly increased the viscosity of fermented camel milk. Dabija et al., 2018 [70] and Johari et al., 2021 [71] reported similar results. They observed that the addition of pumpkin seed powder to milk caused an increase in the viscosity of the yogurt gel. Moreover, the addition of oat milk to camel milk caused an increase in the viscosity of fermented camel milk [28].

The TPC of fermented camel milk made from camel milk mixed with PSM was increased by increasing the mixing ratio compared with the control fermented camel milk. This might be attributed to the higher TPC of PSM vs. camel milk [72,73]. These results are consistent with those reported by Barakat and Hassan, 2017 [74], who found that the TPC and radical scavenging activity of yogurt increased when milk was fortified with different types of pumpkin. In addition, Atwaa et al., 2020 [28] found that adding oat milk to camel milk increased the TPC and radical scavenging activity of fermented camel milk.

3.3. Sensory Properties of Fermented Camel Milk Containing PSM

The data presented in Table 3 showed that mixing camel milk with PSM greatly increased the sensory attributes of the resultant fermented camel milk, particularly its flavor and, body and texture, compared with the control fermented camel milk. Moreover, this improvement was proportional to the mixing ratio. The control fermented camel milk had the lowest score for sensory properties, which may be attributed to the weak body and texture and inferior flavor of the curd produced from camel milk [73]. This finding is consistent with the observation reported by Atwaa et al., 2020 [28], who found that fortification of camel milk with oat milk increased the sensory attribute scores of the resultant fermented camel milk. Dabija et al., 2018 [70] and Johari et al., 2021 [71] observed that the addition of pumpkin seed powder to milk caused an improvement in the sensory attributes of the resultant yogurt.

Table 3. Sensory properties of fermented camel milk containing pumpkin seed milk.

Attributes	Treatments		
	C	T_1	T_2
Flavor (60)	40.7 ± 2.30 ^{c}	44.3 ± 2.60 ^{b}	46.2 ± 3.12 ^{a}
Body and Texture (30)	22.2 ± 1.18 ^{c}	25.4 ± 1.74 ^{b}	27.9 ± 1.58 ^{a}
Appearance (10)	7.8 ± 0.50 ^{c}	8.4 ± 0.82 ^{b}	9.2 ± 0.60 ^{a}
Total Scores (100)	70.7 ± 3.10 ^{c}	78.1 ± 3.24 ^{b}	83.3 ± 3.46 ^{a}

Values (means ± SD) with different superscript letters are statistically significantly different ($p \leq 0.05$).

3.4. Effect of Fermented Camel Milk Containing PSM on Final Weight and Body Weight Gain in Rats with Oxidative Stress

The data presented in Table 4 showed that final weight (FW) and body weight gain (BWG) was significantly ($p < 0.05$) affected by the treatments. Using 10 g/day fermented camel milk containing 50% PSM in oxidative stress rats induced the best values of FW (216.4 g) and BWG (21.80%) compared with the positive control group, which had an FW f 202.6 g and a BWG f 16.28%. This improvement may be ascribed to the high vitamin, mineral, and antioxidant content of PSM, which may protect the body's cells from the damage caused by free radicals [72]. Accordingly, Barakat and Mahmoud, 2011 [75], and Ghahremanloo et al., 2018 [76], found that adding pumpkin seed or pumpkin seed extract to the diet of obese rats promoted a significant increment ($p \leq 0.05$) in the BWG and enhanced their nutritional status compared with non-treated rats (positive control group). Dikhanbayeva et al., 2021 [77] found that the addition of camel milk curd masses to rat diets promoted a significant increment ($p \leq 0.05$) in the BWG and enhanced the nutritional status compared with the control group. Therefore, the group fed with fermented camel milk containing PSM exhibited the best outcomes as a result of the combined action of fermented camel milk and PSM.

Table 4. Final weight and body weight gain of oxidative stress rats treated with fermented camel milk containing pumpkin seed milk.

Group	Parameters			
	Initial Weight (g)	Final Weight (g)	Percentage of Increase in Body Weight (%)	BWG (%)
Group (1)	168.2 ± 3.8 ^{a}	222.3 ± 4.8 ^{a}	32.16 ± 1.3 ^{a}	24.33 ± 1.5 ^{a}
Group (2)	169.6 ± 2.9 ^{a}	202.6 ± 3.5 ^{d}	19.45 ± 0.96 ^{d}	16.28 ± 1.4 ^{d}
Group (3)	168.3 ± 4.3 ^{a}	210.5 ± 4.8 ^{c}	25.07 ± 1.7 ^{c}	20.04 ± 1.6 ^{c}
Group (4)	169.1 ± 4.6 ^{a}	216.4 ± 4.4 ^{b}	27.97 ± 1.6 ^{b}	21.80 ± 1.2 ^{b}

Mean values of six rats ± SD. a–d of the small letters in the same column are significantly different at $p \leq 0.05$). Group (1), non-treated non-oxidative stress rats (negative control). Group (2), oxidative stress rats (positive control). Group (3), oxidative stress rats treated with fermented camel milk. Group (4), oxidative stress rats treated with fermented camel milk containing 50% pumpkin seed milk.

3.5. Effect of Fermented Camel Milk Containing PSM on the Serum Lipid Profile of Oxidative Stress Rats

As depicted in Table 5, among the various groups of rats with oxidative stress, group 4 (treated with fermented camel milk containing PSM) had the lowest total cholesterol (75.6 mg/dL) compared with the positive control group, which had the highest total cholesterol (92.4 mg/dL). Regarding the triacylglyceride and LDL levels, the positive control group had the highest content of these components (103.4 and 43.12 mg/dL, respectively) compared with rats treated with fermented camel milk containing PSM, which exhibited a significant decrease in triacylglyceride and LDL content (78.50 and 23.50 mg/dL, respectively). In contrast, HDL content in the positive control group was the lowest (28.60 mg/dL) compared with the other groups. A diet supplemented with fermented camel milk containing PSM increased HDL levels significantly (33.40 mg/dL). Moreover, treatment with fermented camel milk containing PSM decreased total cholesterol, triglycerides, and LDL-c significantly compared with the positive control group. Pumpkin seed exerts a remarkable hypolipidemic effect by reducing total cholesterol and LDL, associated with a significant elevation of HDL [75,76,78]. Carbon tetrachloride causes oxidative stress, with a consequent increment in the formation of reactive oxygen species, which can promote the oxidation of pivotal cellular components (e.g., membrane lipids, proteins, and DNA), leading to cellular damage [79]. Therefore, the beneficial hypolipidemic effect on the lipid profile of the pumpkin seed observed here may be attributed to the phenolic compounds (as natural antioxidants) present in pumpkin seeds [68]. This hypolipidemic action has also been ascribed to the modulation of the lipid metabolism by phenolics and flavonoids, leading to a decrease in total cholesterol, triglycerides, and LDL (not associated with the increase in HDL levels) by upregulating the hepatic peroxisome proliferator-activated receptor α (PPAR-α) [80]. In addition, the high insulin concentration in camel milk can cause the activation of the lipoprotein lipase enzyme [81]. Ashraf et al., 2021 [82] reported that the high mineral content of camel milk (sodium, potassium, zinc, copper, and magnesium), as well as its high vitamin C level, may act as antioxidant agents by removing free radicals. Similar results were obtained by Ghahremanloo et al., 2018 [76] and Dikhanbayeva et al., 2021 [77], who found that pumpkin seed or fermented camel milk had a hypocholesterolemic effect. The group fed with fermented camel milk containing PSM exhibited the best outcomes regarding lipid profiles due to the combined action of fermented camel milk and PSM.

Table 5. Effect of fermented camel milk containing pumpkin seed milk on the lipid profile of experimental rats.

Groups	Parameters			
	Total Cholesterol (TC) (mg/dL)	Triglycerides (TG) (mg/dL)	HDL (mg/dL)	LDL (mg/dL)
Group (1)	70.6 ± 2.5^{d}	80.2 ± 2.2^{c}	39.4 ± 1.8^{a}	15.16 ± 1.1^{d}
Group (2)	92.4 ± 3.2^{a}	103.4 ± 3.2^{a}	28.6 ± 1.5^{d}	43.12 ± 1.3^{a}
Group (3)	85.8 ± 2.8^{b}	87.3 ± 2.4^{b}	32.9 ± 1.7^{c}	35.44 ± 1.2^{b}
Group (4)	75.6 ± 2.3^{c}	78.5 ± 2.6^{c}	36.4 ± 1.6^{b}	23.50 ± 1.6^{c}

Mean values of six rats ± SD. a–d of the small letters in the same column are significantly different at $p \leq 0.05$.

3.6. Effect of Fermented Camel Milk Containing PSM on Liver Function Parameters in Oxidative Stress Rats

The data illustrated in Table 6 showed that the untreated group (positive control) showed a significant ($p \leq 0.05$) increase in ALT and AST and a decrease in total protein and albumin compared with the normal control group. Conversely, the rats treated with fermented camel milk containing PSM showed a significant ($p \leq 0.05$) increase in total protein and albumin and decreased ALT and AST compared with the positive control group. This decrease in aminotransferase enzymes and restoration of various vital functions by hepatocytes can be attributed to the high content of phenolic and bioactive compounds in pumpkin seed, such as phenolic acids and flavonoids. They help the integrity of the plasma membrane in hepatocytes and protect it from damage and the release of the cytosol loaded with these enzymes [68]. Moreover, camel milk's high vitamin C and mineral content may act as antioxidants, restoring aminotransferase enzymes' values [83]. These results are in harmony with Ghahremanloo et al., 2018 [76] and Dikhanbayeva et al., 2021 [77], who reported that pumpkin seed or fermented camel milk significantly reduced the levels of ALT and AST and increased TP and albumin levels in the serum. Our results are also in agreement with a previous study [84], which found that the administration of pumpkin seed oil in sodium nitrate (NaNO3)-induced oxidative damage in rats restored most hematological and biochemical parameters, including ALT, AST, TP and albumin, to their normal level. Given the above findings, the rats fed with fermented camel milk containing PSM exhibited the best results regarding liver function parameters due to the combined action of fermented camel milk and PSM.

Table 6. Effect of fermented camel milk containing pumpkin seed milk on liver function parameters in experimental rats.

Group	Aspartate Aminotransferase (AST U/L)	Alanine Aminotransferase (ALT U/L)	Total Protein (g/dL)	Total Albumin (g/dL)
Group (1)	37.2 ± 1.14^{d}	44.2 ± 2.12^{d}	6.22 ± 0.48^{a}	3.94 ± 0.22^{a}
Group (2)	82.5 ± 2.30^{a}	85.6 ± 3.25^{a}	5.54 ± 0.62^{c}	2.82 ± 0.17^{c}
Group (3)	44.6 ± 1.62^{b}	52.9 ± 2.14^{b}	5.86 ± 0.35^{b}	3.18 ± 0.20^{b}
Group (4)	39.8 ± 1.26^{c}	46.8 ± 3.10^{c}	6.04 ± 0.32^{ab}	3.74 ± 0.24^{a}

Mean values of six rat's ± SD. a–d of the small letters in the same column are significantly different at ($p \leq 0.05$).

3.7. Effect of Fermented Camel Milk Containing PSM on Kidney Function and Oxidative Stress Markers in Oxidative Stress Rats

The data presented in Table 7 showed that the positive control group had a significant increase in creatinine and urea levels ($p < 0.05$) compared with the normal control group. In contrast, the group treated with fermented camel milk containing PSM showed a significant decrease in creatinine and urea compared with the positive control group. The creatinine and urea levels were reduced in the negative control groups, rats treated with fermented camel milk containing PSM, and rats treated with fermented camel milk compared with the positive control group. This conspicuous change may be partially attributed to the high content of bioactive components in pumpkin seeds, such as minerals, vitamins, and phenolic and flavonoid compounds, which may indirectly reduce uric acid levels and keep the kidney protected from the disorders potentially caused by oxidative stress [72]. These bioactive components act as superoxide scavengers, resulting in suppressing reactive oxygen species and uric acid formation [85]. Moreover, plant-based nutrition yielded a lower mean uric acid serum concentration than animal-based nutrition [86]. A previous study [87] reported that a higher plant protein intake was significantly associated with a lower risk of prevalent CKD. A 20 g increase in plant protein intake reduced the risk of developing CKD by 16%. In addition, the high mineral content of camel milk (sodium, potassium, zinc, copper, and magnesium) and its high vitamin C level may act as antioxidants, thereby affording an improvement in kidney function [77,81]. Furthermore, the positive control group exhibited the highest mean value of MDA (68.40 µmol/L) compared with the negative control, which showed the lowest value (43.60 µmol/L).

Table 7. Effect of fermented camel milk containing pumpkin seed milk on kidney function and oxidative stress markers in experimental rats.

Group	Creatinine (mg/dL)	Urea (mg/dL)	Malondialdehyde (MDA) (µmol/L)
Group (1)	0.48 ± 0.04 [d]	16.2 ± 0.42 [d]	43.60 ± 1.5 [d]
Group (2)	0.84 ± 0.06 [a]	25.8 ± 0.25 [a]	68.40 ± 2.2 [a]
Group (3)	0.66 ± 0.03 [b]	21.3 ± 0.34 [b]	51.92 ± 1.6 [b]
Group (3)	0.54 ± 0.02 [c]	18.2 ± 0.22 [c]	46.38 ± 1.2 [c]

Mean values of six rats ± SD. a–d of the small letters in the same column are significantly different at $p \leq 0.05$.

In addition, there was a significant decrease in MDA in the group treated with fermented camel milk containing PSM (46.38 µmol/L). MDA exerts an adverse effect as it alters the cell membranes structure and function [88]. The increase in the MDA level can lead to oxidative mechanisms, high cytotoxicity, and inhibitory actions. MDA acts as a tumor promoter and co-carcinogenic agent [89]. These results are in harmony with the finding of Barakat and Mahmoud, 2011 [75], who reported that pumpkin seed supplementation effectively decreased the level of creatinine, urea, and malondialdehyde. Previous studies [90,91] have found that fermented camel milk supplementation decreased creatinine, urea, and malondialdehyde compared with the positive control group. Therefore, the group fed with fermented camel milk containing PSM exhibited the best results regarding kidney function and decreased oxidative stress markers as a result of the combined action of fermented camel milk and PSM.

4. Conclusions

Based on the information provided above, the mixing of camel milk with PSM improved the chemical, antioxidant, viscosity, and sensory properties of fermented camel milk, and these improvements were proportional to a mixing ratio up to 50%, which improved the technological problems associated with camel milk such as weak texture and salty taste, and increased the content of phenolic components and antioxidants dietary fiber, thus increasing the nutritional value and healthy benefits to fermented camel milk. The consumption of fermented camel milk containing 50% PSM in oxidative stress rats caused a significant decrease in the levels of malondialdehyde (MDA), low-density lipoprotein (LDL), and cholesterol (CL), triglycerides (TGs), AST, ALT, creatinine, and urea. It increased high-density lipoprotein (HDL), total protein, and albumin compared with the non-treated group. The present findings suggest that more extensive studies of PSM are warranted. Incorporating it into dairy products may be beneficial, particularly in non-traditional milk, which has an undesirable taste.

Author Contributions: M.R.S., E.S.H.A., E.S.A.E.-S. and M.F.R. were involved in the conception of the research idea and methodology design, supervision, and performed data analysis and interpretation. B.M.A., A.A.H.S., M.A.A. and E.K.E. were involved in methodology and drafted and prepared the manuscript for publication and revision. The funders had no role in data collection and analysis, decision to publish, or preparation of the manuscript. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by Saudi Ministry of Environment, Water, and Agriculture, facilitated by General Administration of Animal Productions.

Institutional Review Board Statement: This study was conducted with the approval of the approval of the Faculty of Veterinary Medicine, Suez Canal University and the institutional Review Board Number 2022003.

Informed Consent Statement: Not applicable.

Data Availability Statement: The data that support the findings of this study are available on request from the corresponding author.

Acknowledgments: The authors thank the Taif University Researchers supporting project number (TURSP-2020/93), Taif University, Taif, Saudi Arabia, for support.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Khan, I.T.; Nadeem, M.; Imran, M.; Ullah, R.; Ajmal, M.; Jaspal, M.H. Antioxidant properties of Milk and dairy products: A comprehensive review of the current knowledge. *Lipids Health Dis.* **2019**, *18*, 41. [CrossRef] [PubMed]
2. Salem, S.; Meead, G.; El-Rashody, F.M. Physicochemical and sensory properties of ice cream made from camel milk and fortified with dates products. *Int. J. Humanit. Arts Med. Sci.* **2017**, *5*, 29–40.
3. Khalesi, M.; Salami, M.; Moslehishad, M.; Winterburn, J.; Moosavi-Movahedi, A.A. Biomolecular content of camel milk: A traditional superfood towards future healthcare industry. *Trends Food Sci. Technol.* **2017**, *62*, 49–58. [CrossRef]
4. Kaskous, S. Importance of camel milk for human health. *Emir. J. Food Agric.* **2016**, *28*, 158–163. [CrossRef]
5. Meena, S.; Rajput, Y.; Sharma, R. Comparative fat digestibility of goat, camel, cow and buffalo milk. *Int. Dairy J.* **2014**, *35*, 153–156. [CrossRef]
6. Mudgil, P.; Kamal, H.; Yuen, G.C.; Maqsood, S. Characterization and identification of novel antidiabetic and anti-obesity peptides from camel milk protein hydrolysates. *Food Chem.* **2018**, *259*, 46–54. [CrossRef]
7. Solanki, D.; Hati, S. Fermented camel milk: A Review on its bio-functional properties. *Emir. J. Food Agric.* **2018**, *30*, 268–274.
8. Atwaa, E.S.H.; Shahein, M.R.; El-Sattar, E.S.A.; Hijazy, H.H.A.; Albrakati, A.; Elmahallawy, E.K. Bioactivity, Physicochemical and Sensory Properties of Probiotic Yoghurt Made from Whole Milk Powder Reconstituted in Aqueous Fennel Extract. *Fermentation* **2022**, *8*, 52. [CrossRef]
9. Shori, A.B. Camel milk and its fermented products as a source of potential probiotic strains and novel food cultures: A mini review. *PharmaNutrition* **2017**, *5*, 84–88. [CrossRef]
10. Shahein, M.R.; Atwaa, E.S.H.; El-Zahar, K.M.; Elmaadawy, A.A.; Hijazy, H.H.A.; Sitohy, M.Z.; Albrakati, A.; Elmahallawy, E.K. Remedial Action of Yoghurt Enriched with Watermelon Seed Milk on Renal Injured Hyperuricemic Rats. *Fermentation* **2022**, *8*, 41. [CrossRef]
11. Swelam, S.; Zommara, M.A.; Abd El-Aziz, A.E.-A.M.; Elgammal, N.A.; Baty, R.S.; Elmahallawy, E.K. Insights into Chufa Milk Frozen Yoghurt as Cheap Functional Frozen Yoghurt with High Nutritional Value. *Fermentation* **2021**, *7*, 255. [CrossRef]
12. Beltrán-Barrientos, L.; Hernández-Mendoza, A.; Torres-Llanez, M.; González-Córdova, A.; Vallejo-Córdoba, B. Invited review: Fermented milk as antihypertensive functional food. *J. Dairy Sci.* **2016**, *99*, 4099–4110. [CrossRef] [PubMed]
13. Shahein, M.R.; Atwaa, E.S.H.; Radwan, H.A.; Elmeligy, A.A.; Hafiz, A.A.; Albrakati, A.; Elmahallawy, E.K. Production of a Yogurt Drink Enriched with Golden Berry (*Physalispubescens* L.) Juice and Its Therapeutic Effect on Hepatitis in Rats. *Fermentation* **2022**, *8*, 112. [CrossRef]
14. Elkot, W.F.; Ateteallah, A.H.; Al-Moalem, M.H.; Shahein, M.R.; Alblihed, M.A.; Abdo, W.; Elmahallawy, E.K. Functional, Physicochemical, Rheological, Microbiological, and Organoleptic Properties of Synbiotic Ice Cream Produced from Camel Milk Using Black Rice Powder and Lactobacillus acidophilus LA-5. *Fermentation* **2022**, *8*, 187. [CrossRef]
15. Shahein, M.R.; Atwaa, E.S.H.; Elkot, W.F.; Hijazy, H.H.A.; Kassab, R.B.; Alblihed, M.A.; Elmahallawy, E.K. The Impact of Date Syrup on the Physicochemical, Microbiological, and Sensory Properties, and Antioxidant Activity of Bio-Fermented Camel Milk. *Fermentation* **2022**, *8*, 192. [CrossRef]
16. Shahein, M.R.A.; Atwaa, E.S.H.; Babalghith, A.O.; Alrashdi, B.M.; Radwan, H.A.; Umair, M.; Abdalmegeed, D.; Mahfouz, H.; Dahran, N.; Cacciotti, I.; et al. Impact of incorporation of Hawthorn (*C. oxyanatha*) leaves aqueous extract on yogurt properties and its therapeutic effects against oxidative stress in Rats induced by carbon tetrachloride. *Fermentation* **2022**, *8*, 200.
17. Hussain, T.; Tan, B.; Yin, Y.; Blachier, F.; Tossou, M.C.; Rahu, N. Oxidative stress and inflammation: What polyphenols can do for us? *Oxidative Med. Cell. Longev.* **2016**, *2016*, 7432797. [CrossRef]
18. Ďuračková, Z. Some current insights into oxidative stress. *Physiol. Res.* **2010**, *59*, 459–469. [CrossRef]
19. Kumar, S.; Pandey, A.K. Free radicals: Health implications and their mitigation by herbals. *Br. J. Med. Med. Res.* **2015**, *7*, 438–457. [CrossRef]
20. Kim, Y.S.; Young, M.R.; Bobe, G.; Colburn, N.H.; Milner, J.A. Bioactive food components, inflammatory targets, and cancer prevention. *Cancer Prev. Res.* **2009**, *2*, 200–208. [CrossRef]
21. Cosme, P.; Rodríguez, A.B.; Espino, J.; Garrido, M. Plant phenolics: Bioavailability as a key determinant of their potential health-promoting applications. *Antioxidants* **2020**, *9*, 1263. [CrossRef] [PubMed]
22. Vuolo, M.M.; Lima, V.S.; Junior, M.R.M. Phenolic compounds: Structure, classification, and antioxidant power. In *Bioactive Compounds*; Elsevier: Amsterdam, The Netherlands, 2019; pp. 33–50.
23. Samuel, S.A.; Francis, A.O.; Anthony, O.O. Role of the kidneys in the regulation of intra-and extra-renal blood pressure. *Ann. Clin. Hypertens.* **2018**, *2*, 48–58.
24. Tomsa, A.M.; Alexa, A.L.; Junie, M.L.; Rachisan, A.L.; Ciumarnean, L. Oxidative stress as a potential target in acute kidney injury. *PeerJ* **2019**, *7*, e8046. [CrossRef] [PubMed]
25. Shahidi, F.; Ambigaipalan, P. Phenolics and polyphenolics in foods, beverages and spices: Antioxidant activity and health effects—A review. *J. Funct. Foods* **2015**, *18*, 820–897. [CrossRef]

26. Butnariu, M.; Grozea, I. Antioxidant (Antiradical) Compounds. *J. Bioequivalence Bioavailab.* **2012**, *4*, 6. [CrossRef]
27. Charalampopoulos, D.; Wang, R.; Pandiella, S.; Webb, C. Application of cereals and cereal components in functional foods: A review. *Int. J. Food Microbiol.* **2002**, *79*, 131–141. [CrossRef]
28. Atwaa, E.; Hassan, M.; Ramadan, M.F. Production of probiotic stirred yoghurt from camel milk and oat milk. *J. Food Dairy Sci.* **2020**, *11*, 259–264. [CrossRef]
29. Dotto, J.M.; Chacha, J.S. The potential of pumpkin seeds as a functional food ingredient: A review. *Sci. Afr.* **2020**, *10*, e00575. [CrossRef]
30. Staff, T.P. *PDR for Herbal Medicines*; Physician's Desk Reference (PDR): New York, NY, USA, 2004.
31. Stovel, D. *Pumpkin, a Super Food for All 12 Months of the Year*; Storey Publishing: North Adams, MA, USA, 2012.
32. Csikós, E.; Horváth, A.; Ács, K.; Papp, N.; Balázs, V.L.; Dolenc, M.S.; Kenda, M.; Kočevar Glavač, N.; Nagy, M.; Protti, M. Treatment of Benign Prostatic Hyperplasia by Natural Drugs. *Molecules* **2021**, *26*, 7141. [CrossRef]
33. Caili, F.; Huan, S.; Quanhong, L. A review on pharmacological activities and utilization technologies of pumpkin. *Plant Foods Hum. Nutr.* **2006**, *61*, 70–77. [CrossRef]
34. Matus, Z.; Molnár, P.; Szabó, L.G. Main carotenoids in pressed seeds (*Cucurbitae semen*) of oil pumpkin (*Cucurbita pepo* convar. *pepo var. styriaca). Acta Pharm. Hung.* **1993**, *63*, 247–256. [PubMed]
35. Aktaş, N.; Uzlaşır, T.; Tuncil, Y.E. Pre-roasting treatments significantly impact thermal and kinetic characteristics of pumpkin seed oil. *Thermochim. Acta* **2018**, *669*, 109–115. [CrossRef]
36. Glew, R.; Glew, R.; Chuang, L.-T.; Huang, Y.-S.; Millson, M.; Constans, D.; Vanderjagt, D. Amino acid, mineral and fatty acid content of pumpkin seeds (*Cucurbita* spp.) and Cyperus esculentus nuts in the Republic of Niger. *Plant Foods Hum. Nutr.* **2006**, *61*, 49–54. [CrossRef] [PubMed]
37. Yadav, M.; Jain, S.; Tomar, R.; Prasad, G.; Yadav, H. Medicinal and biological potential of pumpkin: An updated review. *Nutr. Res. Rev.* **2010**, *23*, 184–190. [CrossRef] [PubMed]
38. Broznić, D.; Čanadi Jurešić, G.; Milin, Č. Involvement of α-, γ-and δ-tocopherol isomers from pumpkin (*Cucurbita pepo* L.) seed oil or oil mixtures in the biphasic DPPH disappearance kinetics. *Food Technol. Biotechnol.* **2016**, *54*, 200–210. [CrossRef] [PubMed]
39. Naziri, E.; Mitić, M.N.; Tsimidou, M.Z. Contribution of tocopherols and squalene to the oxidative stability of cold-pressed pumkin seed oil (*Cucurbita pepo* L.). *Eur. J. Lipid Sci. Technol.* **2016**, *118*, 898–905. [CrossRef]
40. Abou-Zeid, S.M.; AbuBakr, H.O.; Mohamed, M.A.; El-Bahrawy, A. Ameliorative effect of pumpkin seed oil against emamectin induced toxicity in mice. *Biomed. Pharmacother.* **2018**, *98*, 242–251. [CrossRef]
41. Aghaei, S.; Nikzad, H.; Taghizadeh, M.; Tameh, A.A.; Taherian, A.; Moravveji, A. Protective effect of Pumpkin seed extract on sperm characteristics, biochemical parameters and epididymal histology in adult male rats treated with Cyclophosphamide. *Andrologia* **2014**, *46*, 927–935. [CrossRef]
42. Cho, Y.H.; Lee, S.Y.; Jeong, D.W.; Choi, E.J.; Kim, Y.J.; Lee, J.G.; Yi, Y.H.; Cha, H.S. Effect of pumpkin seed oil on hair growth in men with androgenetic alopecia: A randomized, double-blind, placebo-controlled trial. *Evid. Based Complementary Altern. Med.* **2014**, *2014*, 549721. [CrossRef]
43. Ezea, B.O.; Ogbole, O.O.; Ajaiyeoba, E.O. In vitro anthelmintic properties of root extracts of three Musa species. *J. Pharm. Bioresour.* **2019**, *16*, 145–151. [CrossRef]
44. Jayaprakasam, B.; Seeram, N.P.; Nair, M.G. Anticancer and antiinflammatory activities of cucurbitacins from Cucurbita andreana. *Cancer Lett.* **2003**, *189*, 11–16. [CrossRef]
45. Balbino, S.; Dorić, M.; Vidaković, S.; Kraljić, K.; Škevin, D.; Drakula, S.; Voučko, B.; Čukelj, N.; Obranović, M.; Ćurić, D. Application of cryogenic grinding pretreatment to enhance extractability of bioactive molecules from pumpkin seed cake. *J. Food Process Eng.* **2019**, *42*, e13300. [CrossRef]
46. Fruhwirth, G.O.; Wenzl, T.; El-Toukhy, R.; Wagner, F.S.; Hermetter, A. Fluorescence screening of antioxidant capacity in pumpkin seed oils and other natural oils. *Eur. J. Lipid Sci. Technol.* **2003**, *105*, 266–274. [CrossRef]
47. Alhakamy, N.A.; Fahmy, U.A.; Ahmed, O.A. Attenuation of benign prostatic hyperplasia by optimized tadalafil loaded pumpkin seed oil-based self nanoemulsion: In vitro and in vivo evaluation. *Pharmaceutics* **2019**, *11*, 640. [CrossRef]
48. Singh, A.; Kumar, V. Nutritional, phytochemical, and antimicrobial attributes of seeds and kernels of different pumpkin cultivars. *Food Front.* **2022**, *3*, 182–193. [CrossRef]
49. Reeves, P.G. Purified diets for laboratory rodents: Final report of the American Institute of Nutrition ad hoc writing committee on the reformation of the AIN-76A rodent diet. *J. Nutr.* **1993**, *123*, 1939–1951. [CrossRef]
50. Hassan, A.A.; Aly, M.; El-Hadidie, S.T. Production of cereal-based probiotic beverages. *World Appl. Sci. J.* **2012**, *19*, 1367–1380.
51. Tamime, A.; Robinson, R. *Yoghurt. Science and Technology*; Woodhead Publishing Limited England: Cambridge, UK, 1999.
52. Chemists, A.; Horwitz, W. *Official Methods of Analysis*; Association of Official Analytical Chemists: Washington, DC, USA, 1975; Volume 222.
53. Aryana, K.J. Folic acid fortified fat-free plain set yoghurt. *Int. J. Dairy Technol.* **2003**, *56*, 219–222. [CrossRef]
54. Maksimović, Z.; Malenčić, Đ.; Kovačević, N. Polyphenol contents and antioxidant activity of Maydis stigma extracts. *Bioresour. Technol.* **2005**, *96*, 873–877. [CrossRef]
55. Apostolidis, E.; Kwon, Y.-I.; Shetty, K. Inhibitory potential of herb, fruit, and fungal-enriched cheese against key enzymes linked to type 2 diabetes and hypertension. *Innov. Food Sci. Emerg. Technol.* **2007**, *8*, 46–54. [CrossRef]

56. Kujawska, M.; Ignatowicz, E.; Murias, M.; Ewertowska, M.; Mikołajczyk, K.; Jodynis-Liebert, J. Protective effect of red beetroot against carbon tetrachloride-and N-nitrosodiethylamine-induced oxidative stress in rats. *J. Agric. Food Chem.* **2009**, *57*, 2570–2575. [CrossRef] [PubMed]
57. Caraway, W.T. Determination of uric acid in serum by a carbonate method. *Am. J. Clin. Pathol.* **1955**, *25*, 840–845. [CrossRef] [PubMed]
58. Bonsens, K.; Taussky, D. Determination of serum creatinine. *J. Chem. Inv.* **1984**, *27*, 648–660.
59. Marsh, W.H.; Fingerhut, B.; Miller, H. Automated and manual direct methods for the determination of blood urea. *Clin. Chem.* **1965**, *11*, 624–627. [CrossRef] [PubMed]
60. Kei, S. Serum lipid peroxide in cerebrovascular disorders determined by a new colorimetric method. *Clin. Chim. Acta* **1978**, *90*, 37–43. [CrossRef]
61. Bergmeyer, H.; Harder, M. A colorimetric method of the determination of serum glutamic oxaloacetic and glutamic pyruvic transaminase. *Clin. Biochem.* **1986**, *24*, 1–488.
62. Dougnon, T.V.; Bankolé, H.S.; Klotoé, J.R.; Sènou, M.; Fah, L.; Koudokpon, H.; Akpovi, C.; Dougnon, T.J.; Addo, P.; Loko, F. Treatment of hypercholesterolemia: Screening of Solanum macrocarpon Linn (Solanaceae) as a medicinal plant in Benin. *Avicenna J. Phytomed.* **2014**, *4*, 160–169. [CrossRef]
63. Devi, R.; Sharma, D. Hypolipidemic effect of different extracts of Clerodendron colebrookianum Walp in normal and high-fat diet fed rats. *J. Ethnopharmacol.* **2004**, *90*, 63–68. [CrossRef]
64. Friedewald, W.T.; Levy, R.I.; Fredrickson, D.S. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin. Chem.* **1972**, *18*, 499–502. [CrossRef]
65. McClave, J.; Benson, P. *Statistical for Business and Economics*; Maxwell Macmillan International editions; Dellen Publishing Co.: New York, NY, USA, 1991; Volume 1991, pp. 272–295.
66. Fouzia, R.; Noureddine, S.; Mebrouk, K. Evaluation of the factors affecting the variation of the physicochemical composition of Algerian camel's raw milk during different seasons. *Adv. Environ. Biol.* **2013**, *7*, 4879–4884.
67. Bouhaddaoui, S.; Chabir, R.; Errachidi, F.; El Ghadraoui, L.; El Khalfi, B.; Benjelloun, M.; Soukri, A. Study of the biochemical biodiversity of camel milk. *Sci. World J.* **2019**, *2019*, 2517293. [CrossRef] [PubMed]
68. Peiretti, P.G.; Meineri, G.; Gai, F.; Longato, E.; Amarowicz, R. Antioxidative activities and phenolic compounds of pumpkin (*Cucurbita pepo*) seeds and amaranth (*Amaranthus caudatus*) grain extracts. *Nat. Prod. Res.* **2017**, *31*, 2178–2182. [CrossRef] [PubMed]
69. Galeboe, O.; Seifu, E.; Sekwati-Monang, B. Production of camel milk yoghurt: Physicochemical and microbiological quality and consumer acceptability. *Int. J. Food Stud.* **2018**, *7*, 51–63. [CrossRef]
70. Dabija, A.; Codină, G.G.; Gâtlan, A.-M.; Sănduleac, E.T.; Rusu, L. Effects of some vegetable proteins addition on yogurt quality. *Sci. Study Res. Chem. Chem. Eng. Biotechnol. Food Ind.* **2018**, *19*, 181–192.
71. Johari, S.; Hosseini Ghaboos, S.H.; Shahi, T. Investigation on the rheological properties of fortified yogurt containing pumpkin powder. *Food Sci. Technol.* **2021**, *18*, 349–358. [CrossRef]
72. Peng, M.; Lu, D.; Liu, J.; Jiang, B.; Chen, J. Effect of Roasting on the Antioxidant Activity, Phenolic Composition, and Nutritional Quality of Pumpkin (*Cucurbita pepo* L.) Seeds. *Front. Nutr.* **2021**, *8*, 647354. [CrossRef]
73. Soliman, T.N.; Shehata, S.H. Characteristics of fermented camel's milk fortified with kiwi or avocado fruits. *Acta Sci. Pol. Technol. Aliment.* **2019**, *18*, 53–63.
74. Barakat, H.; Hassan, M.F. Chemical, nutritional, rheological, and organoleptical characterizations of stirred pumpkin-yoghurt. *Food Nutr. Sci.* **2017**, *8*, 746–759. [CrossRef]
75. Barakat, L.A.; Mahmoud, R.H. The antiatherogenic, renal protective and immunomodulatory effects of purslane, pumpkin and flax seeds on hypercholesterolemic rats. *North Am. J. Med. Sci.* **2011**, *3*, 411–417. [CrossRef]
76. Ghahremanloo, A.; Hajipour, R.; Hemmati, M.; Moossavi, M.; Mohaqiq, Z. The beneficial effects of pumpkin extract on atherogenic lipid, insulin resistance and oxidative stress status in high-fat diet-induced obese rats. *J. Complementary Integr. Med.* **2018**, *15*, 20170051. [CrossRef]
77. Dikhanbayeva, F.; Zhaxybayeva, E.; Smailova, Z.; Issimov, A.; Dimitrov, Z.; Kapysheva, U.; Bansal, N. The effect of camel milk curd masses on rats blood serum biochemical parameters: Preliminary study. *PLoS ONE* **2021**, *16*, e0256661. [CrossRef] [PubMed]
78. Makni, M.; Fetoui, H.; Gargouri, N.; Garoui, E.M.; Jaber, H.; Makni, J.; Boudawara, T.; Zeghal, N. Hypolipidemic and hepatoprotective effects of flax and pumpkin seed mixture rich in ω-3 and ω-6 fatty acids in hypercholesterolemic rats. *Food Chem. Toxicol.* **2008**, *46*, 3714–3720. [CrossRef] [PubMed]
79. Yilmaz, S.; Kaya, E.; Kisacam, M.A. The effect on oxidative stress of aflatoxin and protective effect of lycopene on aflatoxin damage. *Aflatoxin Control Anal. Detect. Health Risks* **2017**, *30*, 67–90.
80. Zeni, A.L.B.; Moreira, T.D.; Dalmagro, A.P.; Camargo, A.; Bini, L.A.; Simionatto, E.L.; Scharf, D.R. Evaluation of phenolic compounds and lipid-lowering effect of Morus nigra leaves extract. *An. Acad. Bras. Ciências* **2017**, *89*, 2805–2815. [CrossRef] [PubMed]
81. Agrawal, R.P.; Saran, S.; Sharma, P.; Gupta, R.P.; Kochar, D.K.; Sahani, M.S. Effect of camel milk on residual β-cell function in recent onset type 1 diabetes. *Diabetes Res. Clin. Pract.* **2007**, *3*, 494–495. [CrossRef] [PubMed]

82. Ashraf, A.; Mudgil, P.; Palakkott, A.; Iratni, R.; Gan, C.-Y.; Maqsood, S.; Ayoub, M.A. Molecular basis of the anti-diabetic properties of camel milk through profiling of its bioactive peptides on dipeptidyl peptidase IV (DPP-IV) and insulin receptor activity. *J. Dairy Sci.* **2021**, *104*, 61–77. [CrossRef]
83. Mailam, A. A Comparative Study On Antidiabetic Effect Of Buffalo And Camel Fermented Milk In Induced Diabetic Rats. *Adv. Food Sci.* **2017**, *39*, 124–132.
84. Rouag, M.; Berrouague, S.; Djaber, N.; Khaldi, T.; Boumendjel, M.; Taibi, F.; Abdennour, C.; Boumendjel, A.; Messarah, M. Pumpkin seed oil alleviates oxidative stress and liver damage induced by sodium nitrate in adult rats: Biochemical and histological approach. *Afr. Health Sci.* **2020**, *20*, 413–425. [CrossRef]
85. Lin, S.; Zhang, G.; Liao, Y.; Pan, J.; Gong, D. Dietary flavonoids as xanthine oxidase inhibitors: Structure–affinity and structure–activity relationships. *J. Agric. Food Chem.* **2015**, *63*, 7784–7794. [CrossRef]
86. Chiu, T.H.; Liu, C.-H.; Chang, C.-C.; Lin, M.-N.; Lin, C.-L. Vegetarian diet and risk of gout in two separate prospective cohort studies. *Clin. Nutr.* **2020**, *39*, 837–844. [CrossRef]
87. Alvirdizadeh, S.; Yuzbashian, E.; Mirmiran, P.; Eghtesadi, S.; Azizi, F. A prospective study on total protein, plant protein and animal protein in relation to the risk of incident chronic kidney disease. *BMC Nephrol.* **2020**, *21*, 489. [CrossRef]
88. Nair, G.G.; Nair, C.K.K. Radioprotective effects of gallic acid in mice. *BioMed Res. Int.* **2013**, *2013*, 953079. [CrossRef] [PubMed]
89. Koc, M.; Taysi, S.; Buyukokuroglu, M.E.; Bakan, N. Melatonin protects rat liver against irradiation-induced oxidative injury. *J. Radiat. Res.* **2003**, *44*, 211–215. [CrossRef] [PubMed]
90. Hamed, H.; Gargouri, M.; Boulila, S.; Chaari, F.; Ghrab, F.; Kallel, R.; Ghannoudi, Z.; Boudawara, T.; Chaabouni, S.; El Feki, A. Fermented camel milk prevents carbon tetrachloride induced acute injury in kidney of mice. *J. Dairy Res.* **2018**, *85*, 251–256. [CrossRef] [PubMed]
91. El-Zahar, K.M.; Hassan, M.F.; Al-Qaba, S.F. Protective Effect of Fermented Camel Milk Containing Bifidobacterium longum BB536 on Blood Lipid Profile in Hypercholesterolemic Rats. *J. Nutr. Metab.* **2021**, *2021*, 1557945. [CrossRef]