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Research Article

Variation of Fatty Acids and Antioxidants Contents of Vegetarian Rayeb Milk as Affected by Fortification with Natural and Artificial Sweeteners

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ARTICLE INFO

Article history:
Received: 25 April, 2020
Accepted: 16 May, 2020
Published: 15 June, 2020

Keywords:
Rayeb milk
sesame milk
omega-6
omega-3
omega-9

ABSTRACT

Rayeb milk (bio-stirred yogurt) samples were prepared from cow milk sesame milk or cow and sesame milk mixture (1:1) with or without adding sucrose (5%), honey (5%), fructose (2.5%) and sorbitol (1.5%) and using ABT-5 culture. Results showed levels of saturated fatty acids (SFA), short chain fatty acids (SCFA) and medium chain fatty acids (MCFA were lower whereas values of unsaturated fatty acids (USFA), monounsaturated fatty acids (MUSFA), polyunsaturated fatty acids (PUSFA) and long chain fatty acids (LCFA) were higher in sesame milk Rayeb than that of Rayeb manufactured from cow milk. Rayeb made from cow and sesame mixture had higher levels of antioxidant activity than Rayeb prepared from cow milk or sesame milk. The acidity, total solids and total volatile fatty acids values of Rayeb milk treatments contained sweeteners were higher than that of control. The addition of sweeteners decreased SFA, SCFA and MCFA and increased USFA, MUSFA, PUSFA, LCFA and antioxidant activity values of Rayeb milk. Essential fatty acids, linoleic acid (omega-6), α-linolenic acid (omega-3) and oleic acid (omega-9) greatly increased in Rayeb made from cow and sesame milk mixture. Adding sweeteners had the same effect. Fortification of Rayeb milk with sweeteners highly improved the smell, taste, mouth feel, texture and body evaluation scores.

Introduction

Foods play very important role in health and disease. Hippocrates was on the right path when he said, “Let food be thy medicine and medicine be thy food”. Now we might change that to "Let functional food be thy medicine. Functional food can be generally classified as a natural food with improved composition by employing particular agronomical conditions, food including a health-promoting component, food from which a component has been removed to produce less adverse effects on health, food in which the nature of one or more of its components has been chemically improved for obtaining health benefits and food in which the bioavailability of one or more of its components has been increased to improve the assimilation of a health promoting component [1]. Nowadays most popular functional food ingredients worldwide are probiotics, prebiotics and symbiotics, dietary fiber, omega 3 fatty acids, oleic acids and phytosterols, phytoestrogens and phenolic compounds [1]. Milk and dairy products provide all the necessary nutrients for healthy living. Fermented milk products, like yoghurt and probiotic foods based on lactobacilli and bifidobacteria, have gained popularity as functional foods and their consumption is on the increase throughout the world. Bioactive ingredients, including probiotics, are now being used in many other food applications [2].

On the other hand, an unhealthy diet and some eating behaviors have been linked to high risk of obesity and finally to Type 2 Diabetes (T2D) [3]. Several epidemiologic studies reveal a parallel increase of the twin epidemics of obesity and diabetes which is a chronic disease characterized by derangement in glucose metabolism and abnormalities in fat and protein metabolism [4]. Food for Specific Groups regulation (Regulation (EU) No 609/2013) abolished the concept of diabetic foods. Consequently, the development and design of functional foods for reducing the risk of chronic diseases such as diabetes and disability,

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http://dx.doi.org/10.31487/j.CMR.2020.01.05

which refers to diabetes occurring in the context of obesity, have a key role for achieving a global sustainable health [1].

The sweetened foods have very important critical role in diabetes and obesity patients' life. Over the past 10 years a number of large observational studies have found positive associations between sugar-sweetened beverages consumption and long-term weight gain and development of T2D and related metabolic conditions [5]. Nonnutritive sweeteners have been utilized in the diet of diabetic patients an agent to replace glucose and sucrose. The nutritive sweeteners fructose, xylitol, and sorbitol are being considered as possible alternatives for glucose and sucrose [6]. Sugar alcohols, including sorbitol, have been approved by the Food and Drug Administration as generally recognized as safe or as food additives and are used by food manufacturers to fully or partially replace added sugars in foods, as well as to serve as bulking agents. In studies comparing sugar alcohols to similar amounts of fructose, sucrose, or glucose in individuals with diabetes, the sugar alcohols produce significantly lower postprandial glucose responses [7]. Therefore, the aim of this investigation was to study the possibility of manufacture sweetened functional dairy food suitable for healthy people or diabetics or obesity.

Materials and Methods

I Materials

Raw cow milk was obtained from Animal Production Research Institute, Agriculture Research Center. Sugar, honey and sesame (*Sesamum indicum*) were purchased from a local grocery in Damiette Governorate. Fructose and sorbitol were obtained from El-Gomhouria Chemical Company, Egypt. ABT-5 culture which consists of *S. thermophilus*, *L. acidophilus* + *Bifidobacterium* was obtained d from Chr. Hansen's Lab A/S Copenhagen, Denmark. Starter cultures were in freeze-dried direct-to-vat set form and stored at -18°C until used.

II Methods

i Preparation of Sesame Milk

Firstly, the decorticated sesame seed was grinded then hot water (75°C) was added (1 sesame: 2 water). The mixture was blended in a blender at high speed for 10 min. The obtained sesame milk was filtered through cheesecloth to separate coarse particles. For pasteurization, sesame milk was filled in a beaker and heated to 90°C/5 min in boiling water bath with manual stirring then sesame milk was cooled to room temperature. Sesame milk had total acidity 0.21%, pH 6.43, E_h 38.4, TS 11.42%, protein 2.63% and fat 5.9%.

ii Preparation of Rayeb Milk Made from Cow and Sesame Milk Mixture

Seven treatments of Rayeb milk were made from cow's milk, sesame milk and various sweeteners mixtures as follows:

A: Rayeb milk made from cow milk

B: Rayeb milk made from sesame milk

C: Rayeb milk made from 50% cow milk + 50% sesame milk

D: Rayeb milk made from 50% cow milk + 50% sesame milk + 5% sucrose

E: Rayeb milk made from 50% cow milk + 50% sesame milk + 5% honey

F: Rayeb milk made from 50% cow milk + 50% sesame milk + 2.5% fructose

G: Rayeb milk made from 50% cow milk + 50% sesame milk + 1.5% sorbitol

After pasteurization, cow milk was mixed with sesame milk. Immediately, milk of various treatments was cooled to 40°C, fortified with sweeteners, inoculated with cultures (0.1 g/L of milk mix), incubated at 40°C for fully coagulation, and stored at 4°C overnight. Once blended for three min and divided to three parts transferred to three jars which preserved at 4°C for 14 days. Rayeb milk samples were analyzed when fresh and after 7 and 14 days of refrigerated storage.

iii Chemical Analysis

Total solids, fat and total nitrogen contents of Rayeb milk samples were determined according to AOAC [8]. Titratable acidity (as lactic acid %) was measured by titrating 10g of sample mixed with 10ml of boiling distilled water against 0.1 N NaOH using a 0.5% phenolphthalein indicator to an end point of faint pink color. pH of the sample was measured at 17 to 20°C using a pH meter (Corning pH/ion analyzer 350, Corning, NY) after calibration with standard buffers (pH 4.0 and 7.0). Redox potential was measured with a platinum electrode (model P14805-SC-DPAS-K8S/325; Ingold (now Mettler Toledo), Urdorf, Switzerland) connected to a pH meter (model H 18418; Hanna Instruments, Padova, Italy). Total volatile fatty acids (TVFA) were determined according to Kosikowiski [9]. The antioxidant activity of Rayeb milk was measured in terms of hydrogen donating or radical scavenging ability, using the stable radical DPPH as described by Olivera *et al.* [10].

iv Determination of Fatty Acids Composition

The extraction of milk fat was done using the method of Rose-Gottlieb using diethyl ether and petroleum ether (Methodenbuch, Bd. VI VDLUFA-Verlag, Darmstadt, 1985). After that, the solvents were evaporated on a vacuum rotary evaporator. For obtaining methyl esters of the fatty acids, sodium methylate (CH_3ONa) was used [11]. The fatty acid composition of Rayeb milk was determined by gas chromatography "Pay-Unicam 304" with flame ionization detector and column EC^{TM}-WAX, 30 m, ID 0.25 mm, Film:0,25 μm.

v Sensory Properties Judging

The sensory properties of Rayeb milk samples were determined according to Tunde-Akintunde and Souley [12].

vi Statistical Analysis

The obtained results were statistically analyzed using a software package based on analysis of variance [13]. When F-test was significant, least significant difference (LSD) was calculated according to Duncan for the comparison between means [14]. The data presented, in the tables, are the mean of 3 experiments.

Results and Discussion

I Effect of Adding Different Types of Sweeteners on the Chemical Composition of Rayeb Milk Made from Cow and Sesame Milk

The changes in the titratable acidity, pH, E_h and total solids (TS) of Rayeb milk during storage period are tabulated in (Table 1). The values of titratable acidity and E_h gradually increased throughout cold storage in different Rayeb milk treatments. The results of the pH followed an opposite trend. This may be due to lactose fermentation, which produces lactic and acetic acid during fermentation. These results are agreed with those reported by Hamad *et al.* [15]. In fresh samples and during storage period, the acidity and E_h values of Rayeb made from sesame milk (sample B) were higher than that of Rayeb manufactured form cow milk or mixture of cow and sesame milk (samples A and C respectively). Also, the increasing in titratable acidity or E_h values was higher in sesame milk Rayeb than that detected in cow milk one.

On the other side, the acidity and E_h levels of Rayeb milk treatments contained 5% sucrose, 5% honey, 2.5% fructose and 1.5% sorbitol were significantly ($P<0.05$) higher than that of control. Adding honey recorded the highest increase among various sweeteners. This is may be due to the honey content of fructooligosacchrides [16]. Giyarto reported the significant increase in titratable acidity during storage of fermented peanut milk fortified with sugar, but not for the plain fermented peanut milk [17]. High level of sugar supports the metabolism activity of lactic acid bacteria during storage. Acidity in the fermented peanut milk drink at day 28 was twice of that at the day 0. This increase in acidity was presumably attributed to continued fermentation by lactic acid bacteria during storage period.

Table 1: Effect of adding different types of sweeteners on physiochemical composition of Rayeb milk.

Properties	Treatments	Storage period (days)			Means
		Fresh	7	14	
Acidity %	A	0.70	0.90	1.04	0.88^{b}
	B	0.75	0.94	1.10	0.93^{ab}
	C	0.73	0.94	1.11	0.93^{ab}
	D	0.76	0.98	1.17	0.97^{a}
	E	0.78	1.03	1.20	1.00^{a}
	F	0.75	0.94	1.10	0.93^{ab}
	G	0.74	0.94	1.09	0.92^{ab}
	Means	0.74^{C}	0.95^{B}	1.11^{A}	
pH values	A	4.86	4.70	4.59	4.72^{a}
	B	4.73	4.61	4.42	4.59^{ab}
	C	4.78	4.62	4.40	4.60^{ab}
	D	4.72	4.58	4.37	4.56^{b}
	E	4.71	4.50	4.33	4.51^{b}
	F	4.74	4.63	4.43	4.60^{ab}
	G	4.76	4.62	4.43	4.60^{ab}
	Means	4.76^{A}	4.61^{B}	4.42^{C}	
E_h mV	A	151	171	188	170.00^{d}
	B	161	184	202	182.33^{c}
	C	157	185	205	182.33^{c}
	D	164	187	216	189.00^{b}
	E	166	195	221	194.00^{a}
	F	161	185	202	182.66^{c}
	G	159	185	201	181.67^{c}
	Means	159.9^{C}	184.6^{B}	205^{A}	
TS %	A	13.38	13.41	13.40	13.39^{b}
	B	11.77	11.80	11.79	12.73^{c}
	C	12.23	12.21	12.24	12.23^{c}
	D	16.54	16.50	16.52	16.52^{a}
	E	16.24	16.27	16.23	16.25^{a}
	F	13.34	13.40	13.41	13.38^{b}
	G	13.27	13.29	13.30	13.29^{b}
	Means	13.82^{A}	13.84^{A}	13.84^{A}	

^{abcde} Letters indicate significant differences between Rayeb milk treatments.
^{ABCD} Letters indicate significant differences between storage times.

Using sesame milk in Rayeb making decreased TS content compared to that made from cow milk. Total solids values of fresh treatments A, B and C were 13.38, 11.77 and 12.23% respectively. From data in (Table 1), it can be observed that there is a substantial effect of the presence of sweeteners on TS levels of Rayeb milk. Significant increases in TS contents were found with supplementation of Rayeb milk with sweeteners especially sucrose and honey. Similar results were reported by Ammar *et al.* [18].

Results cleared in (Table 2) illustrate the impact of using sesame milk and fortification of Rayeb milk with sweeteners on fat, total nitrogen (TN), total protein (TP) and total volatile fatty acids (TVFA) contents during the storage period. Rayeb made from sesame milk had the highest fat content which may be due to high fat content of sesame milk used in Rayeb manufacture. The fat values of fresh samples A and B were 3.9 and 6.1%. On the other side, fat levels were slightly lower in Rayeb milk samples contained sucrose, honey and fructose whereas adding sorbitol had no clear effect on fat content of Rayeb milk. Generally, total solids and fat values were almost constant in various Rayeb milk samples during storage period.

Table 2: Effect of adding different types of sweeteners on fat, TN, TP and TVFA contents of Rayeb milk.

Properties	Treatments	Storage period (days)			Means
		Fresh	7	14	
Fat %	A	3.9	3.9	3.8	3.86^{d}
	B	6.1	6.0	6.1	6.06^{a}
	C	5.0	5.1	5.1	5.06^{b}
	D	4.7	4.8	4.7	4.73^{c}
	E	4.7	4.7	4.7	4.70^{c}
	F	4.9	5.0	5.0	4.96^{b}
	G	5.0	5.0	5.1	5.06^{b}
	Means	4.9^{A}	4.9^{A}	4.9^{A}	
TN %	A	0.539	0.540	0.541	0.540^{a}
	B	0.420	0.421	0.416	0.419^{c}
	C	0.491	0.489	0.488	0.489^{ab}
	D	0.462	0.465	0.463	0.463^{b}
	E	0.463	0.460	0.464	0.462^{b}
	F	0.475	0.477	0.478	0.477^{ab}
	G	0.485	0.489	0.487	0.487^{ab}
	Means	0.476^{A}	0.477^{A}	0.477^{A}	
TP %	A	3.44	3.45	3.45	3.45^{a}
	B	2.63	2.69	2.60	2.64^{ab}
	C	3.13	3.12	3.11	3.12^{a}
	D	2.95	2.97	2.95	2.96^{ab}
	E	2.95	2.93	2.96	2.95^{ab}
	F	3.03	3.04	3.05	3.04^{a}
	G	3.09	3.12	3.11	3.11^{a}
	Means	3.03^{A}	3.05^{A}	3.03^{A}	
TVFA*	A	8.2	12.6	14.1	11.6^{b}
	B	8.0	12.5	14.2	11.6^{b}
	C	8.1	12.5	14.1	11.6^{b}
	D	10.2	14.9	16.8	13.9^{a}
	E	10.9	15.5	17.8	14.7^{a}
	F	9.7	14.2	16.4	13.4^{ab}
	G	9.4	13.8	16.0	13.1^{ab}
	Means	9.2^{C}	13.7^{B}	15.6^{A}	

^{abcde} Letters indicate significant differences between Rayeb milk treatments.
^{ABCD} Letters indicate significant differences between storage times.
*expressed as ml 0.1 NaOH 100 g^{-1} Rayeb milk.

Conversely to fat contents, TN and TP concentrations of Rayeb milk made from sesame milk were lower than those of cow milk Rayeb. Levels of TN and TP of Rayeb manufactured from mixture of cow and sesame milk (50:50) were lower than of cow milk Rayeb and higher than sesame milk one. At the end of storage period, values of TN were 0.539, 0.420 and 0.491% for samples A, B and C respectively. The addition of sweeteners to milk used in Rayeb preparation slightly lowered TN and TP values. General spiking, TN and TP levels of various Rayeb milk treatments nearly didn't change during storage period. Levels of TVFA of Rayeb made from cow milk, sesame milk or mixture of cow and sesame milk were close to each other. The addition of sweeteners to

Rayeb milk increased TVFA concentrations which refer to the stimulation effect of these sweeteners on culture bacteria. Similar notes were recorded by Chick *et al.* [19].

As a result of starter bacteria growth and secretion of lipase, TVFA values gradually increased in various Rayeb milk treatments during storage period. The obtained results were in accordance with that found by Badawi *et al.* who noticed that the acidity and TVFA of stirred yoghurts increased while pH decreased all through storage [20].

II Free Fatty Acids Content (FFA) of Rayeb Milk

The FFA profile in fresh Rayeb milk samples was illustrated in (Tables 3 & 4).

Table 3: Effect of adding different types of sweeteners on free fatty acids (%) content of fresh Rayeb milk.

Fatty acids	C	Treatments						
		A	B	C	D	E	F	G
		Saturated fatty acids (SFA) %						
Caprylic	8:0	0.46	0.37	0.14	0.21	0.13	0.19	0.23
Capric	10:0	2.46	1.11	1.56	1.50	1.11	1.52	1.61
Undecanoic	11:0	-	0.12	0.10	-	-	0.11	-
Lauric	12:0	2.98	0.94	1.41	1.06	1.03	1.10	1.12
Tridecanoic	13:0	0.46	-	-	0.10	-	-	0.13
Myristic	14:0	9.98	1.02	4.09	4.21	3.62	4.20	4.46
Pentadecanoic	15:0	3.50	0.67	1.38	0.94	0.65	1.04	1.11
Palmitic	16:0	27.92	13.54	19.29	18.16	17.84	18.23	18.62
Heptadecanoic	17:0	2.74	0.78	1.33	1.11	0.92	1.20	1.31
Stearic	18:0	10.62	8.12	8.97	7.68	7.44	7.81	7.91
Arachidic	20:0	0.13	0.62	0.52	0.41	0.33	0.49	0.42
Behenic acid	22:0	0.25	-	-	0.10	-	-	-
Total		61.50	27.29	38.79	35.48	33.07	35.89	36.92
Unsaturated fatty acids (USFA) %								
	12:1 ω5	0.50	-	0.65	0.54	0.55	0.51	0.49
5-Tetradecenoic (phytosteric)	14:1 ω5	0.40	0.71	0.69	0.86	0.95	0.81	0.80
	14:1 ω7	0.32	0.44	0.30	0.39	0.45	0.37	0.38
Myristioleic acid	14:1 ω9	0.34	0.42	0.38	0.43	0.47	0.33	0.40
	16:1 ω5	0.27	0.30	0.24	0.34	0.40	0.35	0.35
Palmitoleic	16:1 ω7	2.40	3.15	2.63	2.88	2.91	2.85	2.38
	16:2 ω4	0.30	0.51	0.42	0.59	0.56	0.51	0.50
Hexagonic	16:3 ω4	0.55	0.78	0.69	0.64	0.63	0.60	0.56
	18:1 ω4	0.17	0.25	0.30	0.29	0.33	0.30	0.31
Octadecosaenoic	18:1 ω5	0.47	0.57	0.50	0.53	0.65	0.55	0.54
Vaccienic	18:1 ω7	1.06	1.29	1.21	1.35	1.50	1.27	1.25
Oleic	18:1 ω9	26.71	43.12	36.84	37.63	38.90	37.58	37.45
	18:2 ω4	0.58	0.50	0.51	0.66	0.37	0.65	0.55
	18:2 ω5	0.38	0.42	0.33	0.45	0.55	0.40	0.42
Linoleic	18:2 ω6	1.90	17.09	12.32	13.34	13.74	13.37	13.24
	18:2 ω7	0.25	0.20	0.20	0.31	0.30	0.32	0.27
α-Linolenic	18:3 ω3	0.61	1.83	1.50	1.78	1.98	1.76	1.67
	18:3 ω4	-	-	0.10	0.10	-	0.15	0.10
Gamma linolenic	18:3 ω6	-	-	-	0.15	0.10	-	-
Octadecatetraenoic	18:4 ω3	0.26	0.24	0.27	0.52	0.54	0.50	0.53
Gadoleic acid	20:1 ω9	-	-	0.17	0.27	0.29	0.31	0.32
Eicosaenoic	20:1 ω11		0.17	-	-	0.15	-	-
Eicosatrienoic	20:3 ω6	-	-	0.11	-	0.10	-	0.10
Total		37.47	71.99	60.36	64.05	66.42	63.49	62.61
Non identified fatty acid		1.03	0.72	0.85	0.47	0.51	0.62	0.47

Table 4: Effect of adding different types of sweeteners on free fatty acid indices ratios of fresh Rayeb milk.

Treatments	SFA	USFA	MUSFA	PUSFA	SCFA	MCFA	LCFA
A	61.50	37.47	32.64	4.83	6.40	45.98	46.59
B	27.29	71.99	50.42	21.57	2.54	21.54	75.20
C	38.79	60.36	43.91	16.45	3.86	30.11	65.18
D	35.48	64.05	45.51	18.54	3.31	29.54	66.68
E	33.07	66.42	47.55	18.87	2.82	28.48	68.19
F	35.89	63.49	45.23	18.26	3.43	29.29	66.66
G	36.92	62.61	44.67	17.94	3.45	29.69	66.39

SFA: saturated fatty acids; USFA: unsaturated fatty acids; MUFA: monounsaturated fatty acids (C:1); PUSFA: polyunsaturated fatty acids (C:2+ C:3); SCFA: short chain fatty acids (C8 to C12); MCFA: medium chain fatty acids (C13 to C16); LCFA: long chain fatty acids (> C16).

III Saturated and Unsaturated Fatty Acids

The levels of saturated fatty acids (SFA) were higher than unsaturated fatty acids (USFA) in sample A (cow milk Rayeb). In treatments which contained sesame milk, the trend was quite the opposite where the values of USFA were considerably greater than SFA. The addition of 50% sesame milk to cow milk markedly lowered the concentration of SFA and inversely increased the values of USFA. Values of SFA for samples A, B and C were 61.50, 27.29 and 38.79% respectively.

It could be viewed form (Tables 3 & 4) that fortification of Rayeb milk with various sweeteners especially honey reduced the amounts of SFA and increased the USFA contents of fresh Rayeb. Values of SFA were 35.48 and 33.07% for samples D and E respectively. Based on these results, combination of sesame milk, ABT culture and honey in one fermented dairy product like Rayeb milk greatly lowered SFA level whereas highly increased USFA content. Decreasing SFA and increasing USFA levels in bio-Rayeb contained sesame milk raise the healthy benefit of this product because it is well known that unsaturated fatty acids are more important in human nutrition.

As a general, the most predominant SFA detected in different Rayeb milk treatments was palmitic acid (C_{16}) followed by stearic acids (C_{18}). The highest acid ratio of USFA was oleic acid (18:1 ω9) followed by palmitoleic acid (16:1 ω7) for treatment A. For treatments B, C, D, E, F and G, the major acid of USFA was also oleic acid but followed by linoleic acid (18:2 ω6).

IV Monounsaturated (MUSFA) and Polyunsaturated Fatty Acids (PUSFA) Fatty Acids

In all Rayeb milk treatments, the levels of MUSFA were considerably higher than PUSFA. Rayeb contained sesame milk possessed higher levels of MUSFA and PUSFA than that made from cow milk. Blending 50% sesame milk with cow milk greatly increased the amounts of MUSFA and PUSFA of Rayeb. The values of PUSFA for treatments A, B and C were 4.83, 21.57 and 16.45% respectively.

Mixing sweeteners with milk used in Rayeb making increased the levels of MUSFA and PUSFA. The contents of MUSFA of treatments A, B, C, D, E, F and G were 32.64, 50.42, 43.91, 45.51, 47.55, 45.23 and 44.67% respectively. Oleic acid was found to have the greatest concentration of MUSFA in different Rayeb milk treatments. The dominant fatty acid of PUSFA was linoleic acid. As it is well known, omega fatty acids are a group of essential fatty acids very important for human health. Rayeb manufactured from sesame milk or cow and sesame milk mixture had very high levels of linoleic acid (omega-6), oleic acid (omega-9) and α-linolenic acid (omega-3) as compared with Rayeb made from cow milk.

V Short Chain Fatty Acids (C8 - C12)

Using sesame milk in Rayeb preparation decreased the contents of SCFA. Also, supplementation of Rayeb milk with sweeteners reduced the SCFA contents. The levels of SCFA in samples A, B, C, D, E, F and G were 6.40, 2.54, 3.86, 3.31, 2.82, 3.43 and 3.45% respectively. In treatment A, the fatty acid lauric (C:12) was the predominant SCFA. In other samples capric acid (C10:0) was the predominant.

VI Medium Chain Fatty Acids (C13 – C16)

Medium chain fatty acids (MCFA) of Rayeb milk toke the same trend of SCFA. Using sesame milk in Rayeb production led to lowering of the content of these fatty acids. The addition of sweeteners slightly lowered the values of MCFA in Rayeb milk samples. Values of MCFA for treatments A, B, C, D, E, F and G were 45.98, 21.54, 30.11, 29.54, 28.48, 29.29 and 29.69% respectively. In various Rayeb milk samples, the concentration of palmitic acid (C16) was the highest of medium chain fatty.

VII Long Chain Fatty Acids (> C16)

The values of long chain fatty acids (LCFA) were considerably higher in sesame milk Rayeb as compared with those of Rayeb made from cow milk. Mixing 50% sesame milk with cow milk increased the concentration of LCFA in Rayeb milk. The levels of LCFA increased in rayeb milk supplemented with sweeteners. Among all the long chain fatty acids tested, the level of oleic acid was the highest in different Rayeb milk treatments. On a general note, the values of LCFA were higher than SCFA and MCFA in all Rayeb milk samples.

VIII Antioxidants Activity of Rayeb Milk

The effect of utilization sesame milk and adding sweeteners on the levels of antioxidant activity of Rayeb milk was showed in (Table 5). Rayeb manufactured from sesame milk contained higher antioxidant activity values than those found in Rayeb made from cow milk. Moreover, Rayeb made from cow and sesame mixture (50:50) had higher levels of antioxidant activity than Rayeb prepared from cow milk or sesame milk.

Table 5: Effect of adding different types of sweeteners on antioxidant activity of fresh Rayeb.

Treatments	Antioxidant activity (DPPH inhibition %)
A	51.11
B	59.42
C	61.02
D	63.74
E	67.98
F	62.13
G	61.67

The levels of antioxidant activity were higher in sweeteners Rayeb than control. Rayeb milk contained honey had the highest values of antioxidant activity followed by sucrose Rayeb. Values of antioxidants activity of fresh samples A, B, C, D, E, F and G were 51.11, 59.42, 61.02, 63.74, 67.98, 62.13 and 61.67% respectively. Gheldof *et al.* cleared that honey and other bee products, such as royal jelly and propolis may be used as functional foods because of their naturally high antioxidant potential [21]. Apart from sugars, honey contains many minor components with antioxidant activity, among them amino acids and proteins, carotenes, phenolic compounds and flavonoids, ascorbic acid, organic acids, and Maillard reaction products [22, 23].

In previous study (Abou-Dobara *et al.*), we found that the populations of *S. thermophilus, L. acidophilus* and *Bifidobacterium* (starter bacteria) were higher in Rayeb milk made from sesame milk than those detected in Rayeb made from cow milk [24]. Also, the numbers of starter bacteria increased in Rayeb prepared using mixtures of cow milk with sesame milk. Adding the sweeteners especially fructose and honey to mixed milk led to pronounced increasing in probiotic bacteria populations of Rayeb milk. Based on these results plus data of FFA and antioxidants activity, it appears that the incorporation of probiotics into the mixture of cow milk and sesame milk and sweeteners seems to offer additional health-promoting features.

IX Changes in Sensory Evaluation of Rayeb Milk During Storage

Data in (Table 6) show the organoleptic properties of Rayeb milk manufactured from cow milk or sesame milk or their mixture and supplemented with various sweeteners. Scores of color and appearance properties of Rayeb made from sesame milk (sample B) were slightly lower than those of Rayeb manufactured from cow milk. The grades of these attributes for Rayeb made from cow and sesame milk mixture (sample C) were similar to those of control Rayeb (sample A). On the other hand, the effect of fortification Rayeb milk with sweeteners was not so much pronounced in color and appearance.

Table 6: Effect of adding different types of sweeteners on sensory evaluation of Rayeb milk

Properties	Treatments	Storage period (days)			Means
		Fresh	7	14	
Color	A	9	9	9	9.0^{a}
	B	8	8	8	8.0^{ab}
	C	9	9	9	9.0^{a}
	D	9	9	9	9.0^{a}
	E	9	9	9	9.0^{a}
	F	9	9	9	9.0^{a}
	G	9	9	9	9.0^{a}
	Means	8.9^{A}	8.9^{A}	8.9^{A}	
Appearance	A	9	9	9	9.0^{a}
	B	9	8	8	8.3^{b}
	C	9	9	9	9.0^{a}
	D	9	9	9	9.0^{a}
	E	9	9	9	9.0^{a}
	F	9	9	9	9.0^{a}
	G	9	9	9	9.0^{a}
	Means	9^{A}	8.9^{A}	8.9^{A}	
Smell	A	9	9	8	8.7^{b}
	B	9	9	9	9.0^{a}
	C	9	9	9	9.0^{a}
	D	10	9	9	9.3^{a}
	E	10	9	9	9.3^{a}
	F	10	9	9	9.3^{a}
	G	10	9	9	9.3^{a}
	Means	9.6^{A}	9.0^{A}	8.9^{AB}	
Taste	A	9	9	9	9.0^{a}
	B	9	9	9	9.0^{a}
	C	9	9	9	9.0^{a}
	D	10	9	9	9.3^{a}

	E	10	9	9	9.3[a]
	F	10	9	9	9.3[a]
	G	10	9	9	9.3[a]
	Means	9.6[A]	9[A]	9[A]	
Mouth feel	A	9	9	9	9.0[a]
	B	9	9	9	9.0[a]
	C	9	9	9	9.0[a]
	D	10	9	9	9.3[a]
	E	10	9	9	9.3[a]
	F	10	9	9	9.3[a]
	G	10	9	9	9.3[a]
	Means	9.6[A]	9[A]	9[A]	
Texture & Body	A	9	9	9	9.0[a]
	B	9	9	9	9.0[a]
	C	9	9	9	9.0[a]
	D	10	10	9	9.7[a]
	E	10	10	9	9.7[a]
	F	10	10	9	9.7[a]
	G	10	10	9	9.7[a]
	Means	9.6[A]	9.6[A]	9[A]	

[abcde] Letters indicate significant differences between Rayeb milk treatments.
[ABCD] Letters indicate significant differences between storage times.

Using sesame milk in Rayeb manufacture had no clear effect on the scores of smell, taste and mouth feel. The same trend was observed for Rayeb made from cow and sesame milk mixture. Fortification of Rayeb milk with sweeteners highly improved the smell, taste and mouth feel evaluation scores. Treatments contained sucrose, honey, fructose and sorbitol gained the highest scores. When compared with plain Rayeb samples, the sweetened Rayeb samples were preferred by the panelists who attributed that to the lovely sweet taste. In similar report to our present work, Amiri *et al.* found that the incorporation of honey led to development of sweetened synbiotic acidophilus milk [25]. The addition of honey (7%) to acidophilus milk made by Lactobacillus acidophilus + Bifidobacterium bifidum + Lactobacillus casei increased the sensory score for colour, flavor, texture and overall acceptability of the product developed. They also mentioned that incorporation of B. bifidum increased the flavour of synbiotic acidophilus milk when compared to L. acidophilus as control, whereas L. casei culture showed thinner consistency in the product. The addition of prebiotic affected only the sensory scores, whereas the probiotics addition resulted in a marginal variation of pH and titratable acidity.

Confirming the enhanced effect of the sweet taste on vegetarian milk, Giyarto *et al.* found that supplementation with sugar give some contribution to the sweetness of the fermented peanut milk drink [26]. Fermented peanut milk drink with no addition of sugar had the lowest scores in aroma, sweetness, and sourness. Addition of sugar increased the acceptability of panelists to aroma, sweetness, and sourness. The panelists preferred the aroma, sweetness and sourness of fermented peanut milk with addition of 6-10% of sugar.

Texture and body scores were similar in Rayeb milk made from cow milk, sesame milk or mixture of cow and sesame milk. Adding different sweeteners improved these properties in Rayeb milk which may be due to the increasing of TS content.

Fresh Rayeb milk samples gained the highest scores of sensory evaluation. During storage period, the sensory evaluation degrees of different samples slightly lowered. Our results are in agreement with Osman and Ismail who cleared that significant ($p< 0.001$) decreases in the total organoleptic scores of bio-yoghurt were noticed when storage period progressed [27].

Conclusion

It can be concluded that blending 50% sesame milk with 50% cow milk and adding 5% sucrose, 5% honey, 2.5% fructose and 1.5% sorbitol and using of ABT-5 culture produced bio-Rayeb with highly nutritional and healthy values. This fermented dairy product contained high levels of linoleic acid (omega-6), α-linolenic acid (omega-3) and oleic acid (omega-9) and had the recommended numbers of probiotic bacteria to show healthy effect. The results of this study suggest that consumption of this sweetened Rayeb milk is suitable for ordinary people or diabetics or obesity.

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