Journal of Food and Dairy Sciences

Journal homepage: www.jfds.mans.edu.eg
Available online at: www.jfds.journals.ekb.eg

Preparation of Functional Yoghrt Drink Fortified with *Moringa olifera* Leaves

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ABSTRACT

Fermented dairy products are considered functional foods, because they have many beneficial effects on human health. Yoghurt is a popular fermented product worldwide. Can yoghurt be innovated by adding ingredients that can improve the quality of the diet, Moringa oleifera (moringa). Leaves have high percentage of nutrient and bioactive compounds. These compounds are proteins, carbohydrates, calcium, phosphorus, potassium, iron, vitamins, beta carotene and other bioactive compounds which are essential for preventer anticancer, antiulcer, antimicrobial, antioxidant and some of therapeutic properties. Various studies have concluded that moringa can be used in food products as a functional ingredient. Different ratios of Moringa oleifera leaves powder extract with ethanol 80 % or water (1, 3 and 5%) were used in preparation of drinking yoghurt and coded respectively as MAE1, MAE2 and MAE3 for alcohol extract and MWE1, MWE2 and MWE3 for water extract. Result showed that MAE2, MAE3 and MWE1 They were the best choice for all sensory attributes and received the highest mean ranking and had the greatest performance for general acceptability in contrast with other treatments. Addition of moringa leaves extract can increase the nutritional and therapeutic properties of the prepared drinking yoghrt. Control has lower total lactic acid bacteria but can increase in addition 5% moringa leaves alcohol extract (MAE3).

Keywords: drinking Yoghrt, *Moringa oliefera,* water extract ,alcohol extract

INTRODUCTION

One of the most common foodstuffs worldwide is fermented milk .With the consumption rate rising over time, yoghurt is regarded as one of the world's most popular fermented dairy products. It is obtained by the action of lactic acid bacteria on milk (Nguyen *etal*., 2018). Owing to its nutritious value and health benefits, yoghurt's popularity, such as increased resistance to lactose intolerance and its physiological benefits, such as antimicrobials and anticancer activity against gastrointestinal infections, immune system stimulation and reduction in serum cholesterol (El-Abbadi *etal*.,2014) .yoghurt is considered a good source of bioactive peptides which are produced during fermentation . For this reason, there is a considerable interest in several attempts to manufacture yogurt with natural antioxidants fortified and present a novel product. (Gahruie *etal*., 2015, .Caleja *etal*., 2016)]. Drinking yoghurt has low viscosity and it is consumed as a cooling drink in many countries.

Drinking yoghrt is one of the world's fastest growing fermented milkproduct, often referred to as drinkable yogurt (Douaud, 2007, Ho *etal*.,2000). A common fermented drink, typically made from milk, yoghurt or drinking yoghurt. It is useful for treating people with lactose intolerance, for preventing diarrhea, minimizing the risk of digestive and other organ cancer or tumors, and also reducing blood cholesterol [Legowo 2009].Yoghurt sold on the market is made from animal sources such as cow's milk and vegetable sources extract. Extracts of many plants, herbs, fruits and mushrooms rich in bioactive can be added to drinking yoghurt to increase its functional propertiesare increasingly used as an better nutrition . (Cossu, *etal*.,2009). Addition may play a role in reducing or mitigating the risk of certain diseases and other health conditions [I.F.I.C.F 2017] . In food industry, the functional food market is the fastest growing segment worldwide. *Moringa oleifera* is one such natural food sources which can be used to add functionality to other foods and fermented foods like yoghrt . New yoghrt can be achieved by using a highly nutritious component , one of which is moringa leaves extract. moringa is easy to be found in the environment. As almost any part of the moringa tree can be used for food, medicine and industrial purposes, it is considered one of the world's most useful plants (Khalafalla *etal*., 2010). Leaves, flowers and fresh pods are used by people as vegetables, while others use them as animal feed (Anjorin *etal*., 2010).

Moringa oleifera is a complete food it has an extraordinary range of medicinal benefits with high nutritional value., it is also well-known as extremely nutritious, being a high-quality basis of protein, vitamins, minerals, amino acids , a variety of phenol compounds and contain all of the essential amino acids in good proportion (Mishra *etal*., 2012,Anwar *eta.,l* 2007 and Salama *etal*., 2017). Leaf extracts are known to have biological properties and often differ depending on the form of solvent used to extract vital components (Anwar *etal*., 2007, Lockett 2000) .These compounds consist of phenols, flavonoids, vita C, vita E, carotene, flavones, zinc and selenium which have been recognized to have strong antioxidant potential (Aqil *etal*., 2006, Okwu,2004). Thus, the addition of the extract of leaves in various fermented dairy products will increase the growth of lactobacilli and bifidobacteria. (Amer *etal*., 2014). Studies have shown that extracts of moringa leaves are good for controlling hyperthyroidism (Tahiliani and Kar, 2000)

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DOI: 10.21608/jfds.2021.201813

Various extracts from the leaves of Moringa oleifera are active against bacteria such as:. *E coli*, *S. areues*, *P. aeruginosa and B. cereus* as these organisms range from pathogenic and toxigenic organism liable to cause food borne illnesses and food spoilage (Abalaka *etal*., 2012). Moringa leaves are commonly used by integrating its extract into food products such as sauces, juices, species, milk and bread, to enhance nutritional and sensory assessment (Mukunzi *etal*., 2011). The objective of this study was to evaluate the effect of the *Moringa oleifera* leaves extract on the sensory and chemical composition of drinking yogurt as fresh and cold storage

MATERIALS AND METHODS

Milk samples: All fresh cow's milk samples were collected from the herd at the Department of Animal production, Faculty of Agriculture, Minia University.

Starter: Yoghurt starter consisted of *Loctobacillus delbrueckii* subsp *bulgaricus* (EMCC 11102) and *Streptococcus salivarius* subsp *Thermophilus* (EMCC 11044) was obtained from Cairo Microbiological Resource's Center (MIRCEN) Faculty of Agriculture, Ain Shams University.

Plant sample : *Moringa olifera* was collected from Minia University's Department of Plant Protection, Faculty of Agriculture.

Preparation of moringa extract solutions:

Water leaves extract: preparation according to .(Eman *etal*., 2019)

Alcohol leaves extract: preparation according to .(Eman *etal*., 2019) and respectively El-Shemy *etal*., 2007; Khalafalla *etal*.,2009.

Preparation of drinking yoghurt: Cow's milk was analyzed for physicochemical properties. Sugar (8.5%) was dissolved in milk at 50 ^{o} C and then heated at 90 ^{o}C for 15 min. Then milk was cooled to 45 ^{o} C and it was inoculated with starter culture. Milk was divided into two portions. The first portion was added to moringa alcohol leaves extract(MAE) and the second portion was added moringa water leaves extract(MWE). Six treatments were carried out as following C1; yoghurt culture with 10% of boiled water as a control, MA1;yoghut with 1% moringa alcohol leaves extract and 9% boiling water,MA2; yoghurt with 3% moringa alcohol leaves extract and 7% boiling water,MA3; yoghurt culture with 5% maringa alcohol leaves extract solution and 5% of boiled water, , MW1;yoghrt with 1% maringa water leaves extract and 9% boiling water,MW2; yoghurt with 3% moringa water leaves extract and 7% boiling water,MW3; yoghurt culture with adding 5% moringa water leaves extract solution and 5% of boiled water. Treatments were incubated at 42^{o}C until pH drops to 4.7 and then kept at 5^{o} C for overnight. Coagulated yoghurt was stirred by mechanical mixer, filled in plastic bottles and stored at 5^{o} C for 21 days as modification described by Eman *etal* (2019) Physiochemical parameters total solids, Moisture, ash, fat, pH and protein contents of drinking yoghurt with Moringa alcohol and water extract were determined by AOAC (2007).

Physicochemical and microbiological analysis:

Calcium ,Phosphor, potassium and iron contents were determined according to Perkin Elmer (1994) and AOAC (2009).

Color characteristics were measured by a color difference meter (model color Tec-PCM, USA) (Francis 1983). Where: L: Value represents darkness from black (0) to white (100) a: Value represents color ranging from red (+) to green(-) b: Value represents yellow (+) to blue.

Ascorbic acid was estimated by (AOAC,2000) titrimetric method of analysis using 2,6-dichlorophenol – indophenol.

Brookfield (BROOKFIELD ENGINEERING LABS.INC.STOUUGHON, MA02072U.S.A)with a NO.1spindle used to measure the viscosity of the beverages as described by Shahnawaz and sheikh (2011).

The total flavonoid content was determined using catechin as a reference using the aluminium chloride method (Chang *etal*., 2002). was determined according to (Kanika *etal.*, 2015) was measured using a Folin- Ciocalteu assay according to . (Stojan *eta* ., 2019).

Acetoin and diacetyl were determined according to Westerfeleld (1945). Standard plate count agar was used for total bacterial count (ISO., 2013).

MRS agar was used for total lactic acid bacterial count (Dave, and Shah, 1996). The enumeration of yeasts and molds were carried out according to (ISO 2008). and *E. coli* was detected using Macconocky agar (APHA, 1994).

Sensory analysis: Sensorial properties of the drinking yoghurt samples were tested by 10 trained panelists using a sensorial rating scale scale of 10 points for texture, 10 points for color, 20 points for flavor and 40 points for acceptability.(Eman *etal* .,2019)

Statistical analysis Experimental results were means ± standard deviation (SD) of three measurements. Analysis of variance was performed by ANOVA procedures. GraphPad Prism® was used for statistical calculations Graph Pad Software, San Diego, CA, USA (Motulsky, 1999).

RESULTS AND DISCUSSION

The chemical compostion of drinking yoghurt using different ratios of ethanol extract(1,3 and5%) and water extract (1,3 and 5%) is presented in Table (1) .it is clear that MAE3 had the highest total solids (21.43%),total proteins (6.400%), fat (4.325%),Ash (0.69%), Fe(84.50ug/dl), Ca (23.21mg/dl) K (315.4mmol/l) and P(73.10mg/dl) whereas it had lowest moister (78.82 %)in comparison with the control and other treatments. Total solids, total proteins , ash, fat, Fe,Ca,K,P are increased with increase the ratios of ethanol extract and water extract Anwar *etal* 2007, Lockett 2000, Bukar *et al* 2010 reported that extracts of the leaves were recognized to possess biological properties and these were frequently become vary according to the kind of solvent used to extract the vital components but moisture took an opposite trend it decreased with increaseing the ratio of ethanol and water extract .This is related to natural evaporation(AL-Assar *etal* 2005).statistical analysis showed that there was a significant different between control and treatments for total solids, total proteins, moisture, Fe ,P,K .This may be due to the composition of *moringa olifera* dry leave extract but Ash,fat,Ca were not significant. Table (2) shows the development of PH in drinking yoghurt supplemented with *moringa olifera* extract. pH gradually decreased during storage until 21 days and the adition of moringa leaves extract significantly of affect the PH of drinking yoghurt compared with control due to composition of moringa leave extract and it decreased with increasing the moringa leave extract.. Moringa contains organic acids, phenolic acids, and flavonoids (Rodríguez-Pérez *etal*.,2016). These phytochemical components may promote the growth of LAB and cause an accelerated drop in pH.

vitamin C content of drinking yoghurt supplemented with moringa leaves alcohol or water extract presented in table (3) it was found that MWE3(drinking water with moringa leaves water extract 5%) had higher vitamin C than control and other treatments . Vit C had a steady decrease during storage until 21 days in all treatments and control .This phenomenon is mainly du to oxidation by light which causes reduction in ascorbic acid content (Surbhi *etal*., 2007).Statistical analysis showed that there is a significant difference between control and treatments due to moringa, it had 7 times more vitamin C (Rockwood, *etal*., 2013).

Table 1. Chemical composition of supplemental drinking yoghourt with moringa alcohol or water extract.

	control	MAE1	MAE2	MAE3	MWE1	MWE2	MWE3
Ash	0.60±0.05	0.64 ±0.05	0.68 ±0.05	0.69 ±0.05	0.62±0.02	0.64 ±0.03	0.65±0.05
Fat %	4.250 ±0.1	4.315 ±0.02	4.320±0.02	4.327±0.03	4.305±0.01	4.313±0.02	4.316 ±0.02
Total protein%	3.6 ±0.1	4.633 ^{a}±0.1	5.633 ^{a}±0.1	6.400^{a}±0.1	3.800 ^{a}±0.1	4.233 ^{a}±0.1	4.300 ^{a}±0.1
Moisture%	80.22 ±0.02	79.59 ^{a}±0.1	78.82 ^{a}±0.02	78.57^{a}±0.1	79.92 ^{a}±0.1	79.85 ^{a}±0.1	79.77 ^{a}±0.04
T.S%	19.68 ±0.1	20.26 ^{a}±0.1	21.09 ^{a}±0.1	21.43^{a}±0.1	20.08 ^{a}±0.1	20.15 ^{a}±0.1	20.23 ^{a}±0.1
fe(µg/100ml)	21.50 ±0.01	31.50a±0.01	77.50 a±0.01	84.50 a±0.01	31.30 a±0.03	67.50 a±0.01	72.50 a±0.01
Ca(mg/100ml)	21.35±0.01	22.08±0.02	22.5±0.01	23.21±0.01	21.38±0.04	21.44±0.01	22.15±0.03
K(mmol/100ml)	284.5±0.02	302.2a±0.01	310.4a±0.03	315.4a±0.01	291.3 a±0.01	292.3 a±0.04	300.1 a±0.01
P (mg/100ml)	69.5±0.02	70.25 a±0.01	73.07 a±0.01	73.10 a±0.01	69.10 ±0.01	70.20 a±0.01	72.35 a±0.01

Values are expressed as mean± standard deviation (*N*=3). a (significantly different from control group at P < 0.05 .MAE1(drinking yoghurt supplemented with 1% *moringa olifera* alcohol extract).MAE2(drinking yoghurt supplemented with 3% *moringa olifera* alcohol extract)MAE3(drinking yoghurt supplemented with 5% *moringa olifera* alcohol extract)MWE1(drinking yoghurt supplemented with 1% *moringa olifera* water extract)MWE2(drinking yoghurt supplemented with 3% moringa olifera water extract)MWE3(drinking yoghurt supplemented with 5% *moringa olifera* water extract)

Table 2. Development of pH in drinking yoghurt supplemented with moringa leave powder extract during storage.

Treatment	Fresh	7 days	15	21
Control	4.78±0.01	4.665±0.01	4.555±0.03	4.515±0.02
MAE1	4.690±0.02	4.635 ^{a}±0.04	4.425 a±0.02	4.410 ^{a} ±0.02
MAE2	4.683±0.01	4.620 ^{a}±0.01	4.404 ^{a}±0.02	4.368 ^{a} ±0.01
MAE3	4.662±0.02	4.611 ^{a}±0.02	4.390 ^{a}±0.01	4.350 ^{a} ±0.01
MWE1	4.693±0.02	4.650 ^{a}±0.01	4.455 ^{a}±0.02	4.440 ^{a} ±0.01
MWE2	4.690±0.02	4.634 ^{a}±0.04	4.435 ^{a}±0.01	4.396 ^{a} ±0.01
MWE3	4.681±0.02	4.629 ^{a}±0.03	4.422 ^{a}±0.02	4.360 ^{a} ±0.02

Values are expressed as mean± standard deviation (*N*=3). a (significantly different from control group at P < 0.05 .MAE1(drinking yoghurt supplemented with 1% *moringa olifera* alcohol extract).MAE2(drinking yoghurt supplemented with 3% *moringa olifera* alcohol extract)MAE3(drinking yoghurt supplemented with 5% *moringa olifera* alcohol extract)MWE1(drinking yoghurt supplemented with 1% *moringa olifera* water extract)MWE2(drinking yoghurt supplemented with 3% moringa olifera water extract) MWE3(drinking yoghurt supplemented with 5% *moringa olifera* water extract)

Table 4 show the *L** value which designates whiteness of the product, decreased significantly (*p<0.05*) as the level of moringa leaves extract increased. Goraya and Bajwa (2015), reported that the *L** value decreased, which reflected the addition of product and their oxidation that made product darker and in turn increased the absorption of light.

Table 3. vitamin C (mg /100g) content of drinking yoghurt supplemented with moringa leave powder extract:

Treatment	Fresh VIT C	7 day	15	21
Control	7.050 ±0.01	7.017±0.01	6.90±0.1	6.483±0.02
MAE1	29.43 ^{a}±0.04	28.17 ^{a}±0.02	27.66^{a}±0.01	24.21 ^{a}±0.02
MAE2	31.04 ^{a}±0.01	30.25 ^{a}±0.04	28.28 ^{a}±0.1	28.24 ^{a}±0.03
MAE3	31.11 ^{a}±0.01	30.54 ^{a}±0.04	30.29 ^{a}±0.02	29.54 ^{a}±0.1
MWE1	35.7 ^{a}±0.03	30.56 ^{a}±0.04	33.07 ^{a}±0.01	32.64 ^{a}±0.02
MWE2	35.78 ^{a}±0.03	34.44 ^{a}±0.03	33.05 ^{a}±0.1	32.47 ^{a}±0.1
MWE3	35.84 ^{a}±0.01	34.83 ^{a}±0.04	33.18 ^{a}±0.1	32.79 ^{a}±0.1

Values are expressed as mean± standard deviation (*N*=3). a (significantly different from control group at P < 0.05 .MAE1(drinking yoghurt supplemented with 1% *moringa olifera* alcohol extract).MAE2(drinking yoghurt supplemented with 3% *moringa olifera* alcohol extract)MAE3(drinking yoghurt supplemented with 5% *moringa olifera* alcohol extract)MWE1(drinking yoghurt supplemented with 1% *moringa olifera* water extract)MWE2(drinking yoghurt supplemented with 3% moringa olifera water extract)MWE3(drinking yoghurt supplemented with 5% *moringa olifera* water extract)

Table 4. Color properties of drinking yoghurt supplemented with moringa leave powder extract:

	Period store	control	MAE1	MAE2	MAE3	MWE1	MWE2	MWE3
L*	Fresh	90.40±0.1	85.21 ^{a}±0.12	82.33 ^{a}±0.1	80.43 ^{a}±0.1	88.31 ^{a}±0.1	86.2 ^{a}±0.1	83.5 ^{a}±0.1
	7 days	90.20±0.04	85 ^{a}±0.1	82.01 ^{a}±0.1	80.15 ^{a}±0.1	88.11 ^{a}±0.01	86 ^{a}±0.2	83 ^{a}±0.2
	15days	90±0.1	84.88 ^{a}±0.1	82 ^{a}±0.05	80.0 ^{a}±0.1	88 ^{a}±0.1	86.77 ^{a}±0.1	82.55 ^{a}±0.1
	21 day	89.98±0.1	84.50 ^{a}±0.03	81.99 ^{a}±0.06	70.89 ^{a}±0.1	87.88 ^{a}±0.1	86.3 ^{a}±0.12	82.33 ^{a}±0.01
a*	Fresh	-6.41±0.1	-10.27 ^{a}±0.1	-14.02 ^{a}±0.03	-16.13^{a}±0.1	-9.14 ^{a}±0.1	-11.20 ^{a}±0.1	-13.50 ^{a}±0.1
	7 days	-6.02±0.1	-10.05 ^{a}±0.01	-14 ^{a}±0.07	-16.01 ^{a}±0.1	-9.01 ^{a}±0.1	-11.04 ^{a}±0.1	-13.11 ^{a}±0.14
	15days	-5.99±0.1	-10 ^{a}±0.03	-13.66 ^{a}±0.03	-15.99 ^{a}±0.04	-8.99 ^{a}±0.05	-10.99 ^{a}±0.1	-13 ^{a}±0.13
	21 day	-5.98±0.1	-9.87 ^{a}±0.03	-13.64 ^{a}±0.1	-15.65 ^{a}±0.02	-8.87 ^{a}±0.3	-10.89 ^{a}±0.1	-12.67 ^{a}±0.01
b*	Fresh	8.80±0.1	9.70 ^{a}±0.06	10.03 ^{a}±0.02	13.40 ^{a}±0.1	9.30 ^{a}±0.2	-9.90 ^{a}±0.1	11.60 ^{a}±0.1
	7 days	8.75±0.1	9.65 ^{a}±0.03	10 ^{a}±0.03	13.22 ^{a}±0.1	9.09 ^{a}±0.1	-9.60 ^{a}±0.17	11.32 ^{a}±0.1
	15days	8.50±0.1	9.31 ^{a}±0.03	9.98 ^{a}±0.04	13.02 ^{a}±0.02	8.97 ^{a}±0.1	-9.22 ^{a}±0.1	11.04 ^{a}±0.4
	21 day	8.34±0.1	9 ^{a}±0.03	9.76 ^{a}±0.01	12.99 ^{a}±0.1	8.70 ^{a}±0.1	-9 ^{a}±0.2	10.99 ^{a}±0.03

Values are expressed as mean± standard deviation (*N*=3). a (significantly different from control group at P < 0.05 .MAE1(drinking yoghurt supplemented with 1% *moringa olifera* alcohol extract).MAE2(drinking yoghurt supplemented with 3% *moringa olifera* alcohol extract)MAE3(drinking yoghurt supplemented with 5% *moringa olifera* alcohol extract)MWE1(drinking yoghurt supplemented with 1% *moringa olifera* water extract)MWE2(drinking yoghurt supplemented with 3% moringa olifera water extract)MWE3(drinking yoghurt supplemented with 5% *moringa olifera* water extract).

The greenness of drinking yoghurt were increased as the level of moringa increased, which reflected the negative *a** value. Drinking yoghurt with moringa leaves alcohol extract showed greener color than water extract ,compared to the control. This may be related to β- carotene (Chakraborty *etal.,* 2019*).* In agreement with what was found by Najgebauer-Lejko *etal*., 2013 who reported that the addition of material to yoghrt had a significantly and effect a concentration dependent manner lowered the lightness while a* shifted to positive range with higher values of b* color parameter.

The viscosity of fresh drinking yoghurt was in range 200.95-288.5 at 10 oC (Table 5). It was increased when the moringa leave extract was increased both to alcohol and water extract as well as the viscosity was higher in drinking yoghurt with moringa leave alcohol extract when compared with control and drinking yoghurt moringa leaves extract. Viscosity increased during storage to 21 days. Statistical analysis showed that there was a significant difference between control and treatments. The viscosity of drinking yoghurt was affected by supplementation with moringa leaves extract and its level. According to Lee and Lucey 2004 the rheological properties of stirred yoghurts were greatly influenced by the physical properties of the original intact (set) yoghurt gels. Increasing of viscosity during the cold storage is related to the structural recovery after stirring.

Table 5. Viscosity (cp) of drinking yoghurt supplemented with moringa leave powder extract

Treatment	Fresh	7 day	15	21
Control	200.95±0.07	260.8±0.35	295±0.35	304±0.2
MAE1	240.50^{a}±0.07	296^{a}±0.4	330^{a}±0.4	370^{a}±0.5
MAE2	269.5^{a}±0.7	300.4^{a}±0.5	380.4^{a}±0.5	430.1^{a}±0.4
MAE3	288.5^{a}±0.01	350.5^{a}±0.5	410.2^{a}±0.4	490.4^{a}±0.5
MWE1	210.50^{a}±0.3	277.1^{a}±0.07	290.2 ^{a}±0.3	322.2 ^{a}±0.3
MWE2	223.25^{a}±0.2	289.1^{a}±0.07	320.5 ^{a}±0.7	360.4 ^{a}±0.5
MWE3	250.15^{a}±0.21	300.3^{a}±0.42	360.5 ^{a}±0.7	393.3 ^{a}±0.3

Values are expressed as mean± standard deviation (*N*=3). a (significantly different from control group at P < 0.05 .MAE1(drinking yoghurt supplemented with 1% *moringa olifera* alcohol extract).MAE2(drinking yoghurt supplemented with 3% *moringa olifera* alcohol extract)MAE3(drinking yoghurt supplemented with 5% *moringa olifera* alcohol extract)MWE1(drinking yoghurt supplemented with 1% *moringa olifera* water extract)MWE2(drinking yoghurt supplemented with 3% moringa olifera water extract)MWE3(drinking yoghurt supplemented with 5% *moringa olifera* water extract).

Antioxidant capacity , TP and TF of drinking yoghurt enhanced by moringa leaves extract with alcohol and water shown in table (6). It is clear that MAE3 had highest contant of antioxidant (160.4%) followed by MAE2 and MAE1(128.4,105)respectively compared with control . Total phenolic took same trend of antioxidant MAE3 recorded the highest total phenolic (28.81%)whereas (MAE2,MAE1) (23.12, 21.88)respectively. On the other hand water extract had the lowest total phenolic (MWE1,MWE2,MWE3) (19.72, 20.10, 21.40). Total flavonoid also had the same trend of antioxidant and total phenolic MAE3 had highest total flavonoid (431.9mg/100) followed by MAE2 and MAE1 (420.1 ,385.2) respectively compared with control and other treatments . These results may be due to the higher content of total phenolic and total flavonoid in ethanol extract (Youssef and Mokhtar 2014). Total phenolic have a strong antioxidant and ability to scavenge free radicals (Li etal., 2006: Abo El-Maati etal., 2015).Antioxidant , total phenolic and total flavonoid gradually increased during storage until 21 days either control and all treatments statically analysis showed that there was a significant different between control and treatments

Table 7 illustrates diacetyl and acetoin content in drinking yoghurt supplemented with moringa olifera extracts **Diacetyl**: Control samples had the lowest diacetyl content, either fresh or in storage with the increased ethanol extract ratio, diacetyl frequently increased. During the time of cold storage, control and all treatments increased periodically. Statistical analysis show that controls and treatments vary greatly. The same trend was adopted by Acetoin. . These results are in the same trend with Law 1981 and El-Sayed *etal*., 2017 ,

Table 6. Phytochemical and antioxidant activity of supplemental drinking yoghourt with moringa alcohol or weter extract

	Period store	control	MAE1	MAE2	MAE3	MWE1	MWE2	MWE3
Antioxidant (Mg/100)	Fresh	74.50±0.2	105^{a}±0.1	128.4^{a}±0.1	160.4^{a}±0.1	99.21^{a}±0.1	102.3^{a}±0.1	103.0^{a}±0.01
	7 days	73.03^{a}±0.04	99.1^{a}±0.1	120.8^{a}±0.1	140.4^{a}±0.1	91.9^{a}±0.1	95.28^{a}±0.1	102.3^{a}±0.1
	15days	74.6^{a}±0.1	104.7^{a}±0.1	132.8^{a}±0.1	157.7^{a}±0.1	99.7^{a}±0.1	111.6^{a}±0.1	118.4^{a}±0.1
	21 day	110.8^{a}±0.1	119.5^{a}±0.1	140.8^{a}±0.1	170.9^{a}±0.1	102.9^{a}±0.1	115^{a}±0.1	121.6^{a}±0.1
Total phenolic (Mg/100)	Fresh	3.73^{a}±0.1	21.88^{a}±0.1	23.12^{a}±0.02	28.81^{a}±0.01	19.72^{a}±0.02	20.10^{a}±0.001	21.40^{a}±0.001
	7 days	4.5^{a}±0.02	22.4^{a}±0.05	23.2^{a}±0.03	29.46^{a}±0.04	20.06^{a}±0.006	20.42^{a}±0.02	21.80^{a}±0.002
	15days	4.6^{a}±0.09	22.93^{a}±0.1	23.87^{a}±0.10	29.77^{a}±0.05	20.21^{a}±0.01	20.54^{a}±0.01	22.22^{a}±0.03
	21 day	4.9^{a}±0.05	23.08a^{a}±0.11	24.11^{a}±0.15	30.07^{a}±0.1	20.83^{a}±0.04	21.24^{a}±0.1	22.78^{a}±0.1
Total flavonoid (Mg/100)	Fresh	108.4±0.14	385.2^{a}±0.02	420.1^{a}±0.04	431.9^{a}±0.12	248.5^{a}±0.1	250^{a}±0.1	265.1^{a}±0.1
	7 days	108.5±0.07	438.5^{a}±0.07	451.8^{a}±0.02	463.5^{a}±0.07	305^{a}±0.05	320.1^{a}±0.05	338.5^{a}±0.07
	15days	171.7±0.01	443.5 ^{a}±0.1	468.5 ^{a}±0.1	478.5 ^{a}±0.1	333.5 ^{a}±0.1	435.1 ^{a}±0.01	476.8^{a}±0.1
	21 day	181.7±0.1	490.1 ^{a}±0.1	500.1 ^{a}±0.1	543.1 ^{a}±0.1	358.4 ^{a}±0.1	466.7 ^{a}±0.1	486.9 ^{a}±0.1

Values are expressed as mean± standard deviation (*N*=3). a (significantly different from control group at P < 0.05 .MAE1(drinking yoghurt supplemented with 1% *moringa olifera* alcohol extract).MAE2(drinking yoghurt supplemented with 3% *moringa olifera* alcohol extract)MAE3(drinking yoghurt supplemented with 5% *moringa olifera* alcohol extract)MWE1(drinking yoghurt supplemented with 1% *moringa olifera* water extract)MWE2(drinking yoghurt supplemented with 3% moringa olifera water extract)MWE3(drinking yoghurt supplemented with 5% *moringa olifera* water extract).

Table 7. Diacetyl and Acetoin contant of supplemental drinking yoghourt with moringa alcohol or weter extract.

	Period store	Control	MAE1	MAE2	MAE3	MWE1	MWE2	MWE3
Diacetyl	Fresh	1.133±0.1	1.271^{a}±0.1	1.275^{a}±0.1	1.335 ^{a} ±0.05	1.23 ^{a} ±0.04	1.244 ^{a} ±0.1	1.250 ^{a} ±0.03
	7 days	1.238±0.1	1.531 ^{a} ±0.04	1.538 ^{a} ±0.1	1.546 ^{a} ±0.1	1.338 ^{a} ±0.1	1.415 ^{a} ±0.02	1.44 ^{a} ± 0.1
	15days	1.562±0.1	1.627^{a}±0.1	1.734^{a}±0.04	1.834 ^{a} ±0.04	1.631 ^{a} ±0.04	1.635 ^{a} ±0.04	1.645 ^{a} ±0.1
	21 day	1.631±0.04	1.825 ^{a} ±0.04	1.830 a ±0.04	1.834 ^{a} ±0.05	1.712 ^{a} ±0.02	1.733 ^{a} ±0.05	1.746 ^{a} ±0.1
Acetoin	Fresh	1.710±0.01	1.949 ^{a} ±0.06	1.950 ^{a} ±0.070	2.01 ^{a} ±0.01	1.903 ^{a} ±0.01	1.910 ^{a} ±0.01	1.925 ^{a} ±0.04
	7 days	1.730±0.04	1.950 ^{a} ±0.07	2.023 ^{a} ±0.04	2.040 ^{a} ±0.05	1.904 ^{a} ± 0.01	1.925 ^{a} ±0.03	1.945 ^{a} ±0.06
	15days	1.750±0.07	2.015 ^{a} ±0.02	2.111 ^{a} ±0.1	2.150 ^{a} ±0.21	1.942 ^{a} ±0.1	1.950 ^{a} ±0.1	1.950 ^{a} ±0.1
	21 day	1.750±0.07	2.050 ^{a} ±0.07	2.201 ^{a} ±0.14	2.335 ^{a} ±0.05	1.943 ^{a} ±0.06	2.015 ^{a} ±0.2	2.005 ^{a} ±0.01

Values are expressed as mean± standard deviation (*N*=3). a (significantly different from control group at P < 0.05 .MAE1(drinking yoghurt supplemented with 1% *moringa olifera* alcohol extract).MAE2(drinking yoghurt supplemented with 3% *moringa olifera* alcohol extract)MAE3(drinking yoghurt supplemented with 5% *moringa olifera* alcohol extract)MWE1(drinking yoghurt supplemented with 1% *moringa olifera* water extract)MWE2(drinking yoghurt supplemented with 3% moringa olifera water extract)MWE3(drinking yoghurt supplemented with 5% *moringa olifera* water extract).

Microbiological analysis:

There were no coliform bacteria in all drinking yoghrt samples either fresh or during refrigerated storage, because of the efficient heat treatment of the different yoghurt milks (85ºC for 15 min) and de contamination conditions during making and storage of drinking yoghrt and the effect of acidity in different yoghurt which prevent pathogenic bacteria growth. The results are in harmony with the results of (Gould, 1991, and El Batawy, 2012).

All drinking yoghurt samples were free from yeast and mold till 7th day of storage period. At 14th day from storage period, yeast and mould has been appeared but were less than 10 cfu/ml and this may be due to some looking in the bottles lead. The obtained results in agreement with (El Batawy, 2012).

Results in table 8 show that the all samples of drinking yoghrt were free from coliform bacteria, yeasts and molds. this may be due to the different rates of extracts from moringa leaves which may be active against bacteria such as: *E. coli*, as these organisms range from pathogenic and toxigenic organism (Abalaka et al., 2012).These findings showed that the moringa extract had a high phenolic component level that is a good source of antioxidant , as well as boosting the health benefits of drinking yogurt.Phenolic componentshave significant antimicrobial effect against pathogenic bacteria, .(FU etal 2016). The data in Table 8 also show that the level of colony forming units of total lactic acid bacteria on the MRS agar with various addition extract of moringa leaves in dilutions10^{7}. Control has lower total lactic acid bacteria but can increase in addition 5% moringa leaves alcohol extract (MAE3) followed by MAE2 and MAE1 respectively this agreement with Amer et al 2014).Total lactic acid bacteria decrease during storage may be due to the decrease of pH.

Table 8. Microbiological analysis drinking yoghurt supplemented with moringa leave powder extract

	Period store	control	MAE1	MAE2	MAE3	MWE1	MWE2	MWE3
L.A.B	Fresh	50×10^{7}	60×10^{7}	66×10^{7}	70×10^{7}	53×10^{7}	55×10^{7}	57×10^{7}
	7 days	40×10^{7}	55×10^{7}	60×10^{7}	67×10^{7}	49×10^{7}	50×10^{7}	51×10^{7}
	15days	35×10^{7}	40×10^{7}	43×10^{7}	50×10^{7}	38×10^{7}	40×10^{7}	43×10^{7}
	21 day	33×10^{7}	39×10^{7}	40×10^{7}	44×10^{7}	35×10^{7}	36×10^{7}	37×10^{7}

Values are expressed as mean± standard deviation (*N*=3). a (significantly different from control group at P < 0.05 .MAE1(drinking yoghurt supplemented with 1% *moringa olifera* alcohol extract).MAE2(drinking yoghurt supplemented with 3% *moringa olifera* alcohol extract)MAE3(drinking yoghurt supplemented with 5% *moringa olifera* alcohol extract)MWE1(drinking yoghurt supplemented with 1% *moringa olifera* water extract)MWE2(drinking yoghurt supplemented with 3% moringa olifera water extract)MWE3(drinking yoghurt supplemented with 5% *moringa olifera* water extract).

The values of texture, color, flavor and overall acceptability are shown in (Table 9). It is clear that addition of the aqueous extract solution of moringa leaves extract(alcohol or water) at all levels have a significant effect (p>0.05) on all parameters, texture, color, flavor and overall acceptability scores at fresh and at the end of storage period between control and all treatments.

For all prametars the addition of *Moringa oleifera* extract with water at 3 and 5 % percent had unfavorable effect .The drinking yogurt was fortified with *Moringa oleifera alcohol* extract at a concentration of 3,5 %gained the highest score, followed by the drinking yogurt with *Moringa oleifera* extract alcohol 1%,and water extract 1% whereas the control had the lowest value followed 2 and 3% water extract . The scores of sensory properties of all treatments and control gradually decreased over storage until 21 days.

Table 9. effect of *moringa olifera* extract on sensory evaluation of drinking yoghurt.

	Period store	control	MAE1	MAE2	MAE3	MWE1	MWE2	MWE3
Flavor(20)	Fresh	16.90 ±0.1	17.90 ^{a}±0.1	18.90 ^{a}±0.1	19.94 a±0.1	17.85 ^{a}±0.1	14.90 ^{a}±0.1	13.85^{a}±0.1
	7 days	17.95 ±0.1	17.90 ^{a}±0.1	18.95 ^{a}±0.1	18.95 a±0.1	17.90 ^{a}±0.1	14.30 ^{a}±0.1	13.70 ^{a}±0.1
	15days	17.95 ±0.1	17.90 ^{a}±0.1	18.95 ^{a}±0.1	18.95 a±0.1	17.90 ^{a}±0.1	14 ^{a}±0.1	13.50 ^{a}±0.1
	21 day	17 ^{a}±0.1	17 ^{a}±0.1	18.60 ^{a}±0.1	18.60 a±0.1	17.80 ^{a}±0.1	13.80 ^{a}±0.1	13.1 ^{a}±0.1
Texture(10)	Fresh	7.5±0.1	9 ^{a}±0.1	10 ^{a}±0.1	10 a±0.1	9 ^{a}±0.1	6.50 ^{a}±0.1	5.50 ^{a}±0.1
	7 days	7.5±0.1	9 ^{a}±0.1	10 ^{a}±0.1	10 a±0.1	9 ^{a}±0.1	6.50 ^{a}±0.1	5.50 ^{a}±0.1
	15days	6.5±0.1	7.5 ^{a}±0.1	8.5 ^{a}±0.1	8.5 a±0.1	8 ^{a}±0.1	6 ^{a}±0.1	5 ^{a}±0.1
	21 day	6.5±0.1	7.5 ^{a}±0.1	8 ^{a}±0.1	8 a±0.1	7 ^{a}±0.1	5.5 ^{a}±0.1	3.5 ^{a}±0.1
Color(10)	Fresh	10 ±0.1	9.1 ^{a}±0.1	9.1 ^{a}±0.1	8.5 a±0.1	8.5 ^{a}±0.1	7.75 ^{a}±0.1	7.75 ^{a}±0.1
	7 days	9.5±0.1	8.5 ^{a}±0.1	8.5 ^{a}±0.1	8.5 a±0.1	8.5 ^{a}±0.1	7.5 ^{a}±0.1	7.5 ^{a}±0.1
	15days	8.5±0.1	8.5^{a}±0.1	8.5 ^{a}±0.1	8.5 a±0.1	8.5 ^{a}±0.1	7.5 ^{a}±0.1	7.5 ^{a}±0.1
	21 day	8.5 ±0.1	8.5^{a}±0.1	8.5 ^{a}±0.1	8.5 a±0.1	8.5 ^{a}±0.1	7.5 ^{a}±0.1	7.5 ^{a}±0.1
acceptability (40)	Fresh	39 ±0.1	38 ^{a}±0.1	38 ^{a}±0.1	38 a±0.1	36.50 ^{a}±0.1	36 ^{a}±0.1	32.50 ^{a}±0.1
	7 days	39 ^{a}±0.1	38 ^{a}±0.1	38 ^{a}±0.1	38 a±0.1	35 ^{a}±0.1	35.5 ^{a}±0.1	32 ^{a}±0.1
	15days	38 ^{a}±0.1	36 ^{a}±0.1	37 ^{a}±0.1	37 a±0.1	35.5 ^{a}±0.1	34 ^{a}±0.1	31 ^{a}±0.1
	21 day	38 ^{a}±0.1	36 ^{a}±0.1	37 ^{a}±0.1	37 a±0.1	33 ^{a}±0.1	32 ^{a}±0.1	30 ^{a}±0.1

Values are expressed as mean± standard deviation (*N*=3). a (significantly different from control group at P < 0.05 .MAE1(drinking yoghurt supplemented with 1% *moringa olifera* alcohol extract).MAE2(drinking yoghurt supplemented with 3% *moringa olifera* alcohol extract)MAE3(drinking yoghurt supplemented with 5% *moringa olifera* alcohol extract)MWE1(drinking yoghurt supplemented with 1% *moringa olifera* water extract)MWE2(drinking yoghurt supplemented with 3% moringa olifera water extract)MWE3(drinking yoghurt supplemented with 5% *moringa olifera* water extract).

CONCLUSION

As a fresh drink and a good source of nutrient components, drinking yoghurt is recommended to be consumed. Results of this study concluded that moringa extract stimulated the growth of starter culture as well as drinking yoghurt made with and moringa alcohol extract was the best to be used in the manufacture of drinking yogurt. This extract enhances the nutritional value and sensory assessment of the resulting drinkin yoghurt and its chemical properties.

ACKNOWLEDGEMENT

The authors would like to express our profound gratitude and heartfelt gratitude to Professor Dr. Ahmed Shawky Zahran, Professor of Dairy Science, Minia University, for his support in reviewing and delivering the study in the best form.

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تحضير مشروب زبادي وظيفي باستخدام اوراق نبات المورنجه

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تعتبر منتجات الالبان المتخمره من الاغذيه الوظيفيه حيث لها تاثير ايجابي علي صحة الانسان و تعتبر الزبادي من اكثر المنتجات اللبنيه المتخمره شيوعا ويمكن اضافه بعض المواد لزيادة قيمتها ومن بين هذه الاضافات نبات المورنجه حيث تحتوي اوراقه علي مكونات ذات قيمه حيويه وغذائيه عاليه مثل الكربوهيدرات و البروتين والكالسيوم والفسفور والبوتاسيوم والحديد و الفيتامينات و البيتاكاروتين ومكونات اخري لها قيمه حيويه . ولهذه المواد دور في الوقايه من العديد من الامراض السرطانيه و القرح وتعتبر ايضا كمضادات بكتيريه و مضادات للاكسده وتؤثر كذلك علي بعض المسببات المرضيه الاخري . لذا في هذه الدراسه تم اضافة مستخلصات اوراق المورنجه الي مشروب الزبادي (مستخلص كحولي ومستخلص مائي) وتم اضافتها بنسب (1,3,5%) ووجد ان اضافة المستخلص المائي بنسبه 1% و المستخلص الكحولي بنسبه 3 , 5 % من افضل النسب وتميزت بقابليه عاليه وكانت العينات المضاف اليها المستخلص الكحولي افضل من العينات المضاف اليها المستخلص المائي من حيث ارتفاع نسبة مضادات الاكسده و الاسكوربيك اسيد ووالفلافونويد والفينوليك وعموما تم الوصول من خلال البحث الي ان اضافه مستخلصات المورنجه الي مشروب الزبادي ادي الي زيادة القيمه الغذائيه والحيويه له مقارنة بعينة المقارنه