Antibacterial Activity of Yogurt Cheese Made from Barki Sheep Milk Supplemented with Olive Oil

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ABSTRACT

The purpose of this study was investigating the antibacterial effects of olive oil supplemented to cheese yoghurt made from Barki sheep milk, on the growth of some probiotic bacterial strains (*Bifidobacterium bifidum* (ATCC15708), *Lactobacillus acidophilus* (ATCC4356), *Lactobacillus delbrueckii* spp. *bulgaricus* (ATCC7995) and *Streptococcus thermophilus* (DSM20259) as well as some pathogenic bacterial strains. Results showed that olive oil (1%) had no effect on the growth of all probiotic bacterial strains used in Barki cheese yoghurt making. No yeasts, moulds, Enterobacteria spp and Staphylococcus spp were detected in cheese yoghurt containing olive oil (1%) through the entire storage period (21days). However, control treatment had Yeast and Moulds at the end of storage period. When four strains of pathogens were added to cheese yoghurt containing different probiotic bacteria and supplemented with olive oil (1%) then stored at 5C for 72hours, results revealed that *E. coli* (ACCT8739) was the most sensitive microorganism while, *Staphylococcus aureus* (ATCC6538) was the most resistant one. The antibacterial activity of cheese yoghurt supplemented with olive oil (1%) was higher than control treatment because olive oil supported the growth of Lactic acid bacteria. From these results, it is recommended that olive oil (1%) can be used as a natural and safe antimicrobial substance in Barki cheese yoghurt and other dairy products, and olive oil may well have the beneficial role in promoting probiotic bacteria and inhibiting harmful bacteria.

Keywords: antibacterial activity, Barki sheep milk, Barki cheese yoghurt, olive oil, pathogenic bacteria, probiotics.

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I. INTRODUCTION

Barki sheep belongs to Barbary breed which inhabits the hot area of North Africa [1]. Smaller fat globule diameter and greater percentage of short chain fatty acids contribute to easier and more rapid digestion of sheep milk. Short-chain fatty acids such as caproic, caprylic and capric give sheep milk the special taste and aroma and have health benefits [2]. Sheep milk is rich in vitamins A, B and E, calcium, phosphorous, potassium and magnesium than cow's milk [3]. Meanwhile, it is a unique product with high nutritional qualities containing shorter – chain fatty acid, more protein, more calcium, and more vitamins than cow's milk. Olive oil is classified as virgin olive oil if it has been extracted exclusively by mechanical or physical procedures such as milling, beating, centrifugation and decantation [4]. The importance of virgin olive oil is related to its high levels of mono-unsaturated fatty acids (mainly oleic acid) and to the presence of minor components including aliphatic and triterpenic alcohols, sterols, hydrocarbons, volatile compounds, and several antioxidants [5]. Limited studies have been carried out on the utilization of olive oil in dairy products. Concentrated yoghurt or cheese yoghurt is popularly known as Labneh in the Middle East, as strained yoghurt in Greece, the rest of Europe and as Suzme yoghurt in Turkey [6], [7].

In the last decades, consumer in the field of food production have considerably changed; consumers believe that foods contribute directly to their health [8] such as olive oil is needed. On the other hand, several researches were done to improve the quality and shelf-life of Labneh by different attitudes such as using olive oil [9], [10] and by using essential oil [11]-[15]. Probiotic bacteria are recognized for their fermentative ability and thus enhancing food safety, improving organoleptic attributes, enriching nutrients, and increasing health benefits [16]-[19]. It is well known that milk fermentation using lactic acid bacteria results in increase its shelf-life as interest to develop a variety of fermented milk products for other beneficial purpose, particularly for health purpose and preventing of toxins produced by food borne pathogens and spoilage bacteria enter human body [16], [18], [20]-[26]. Therefore, the objective of this study was investigating the antibacterial effects of some probiotic bacterial strains as well as olive oil on the activity of some food borne pathogens and spoilage bacteria.

II. MATERIAL AND METHODS

A. Milk Samples

Barki sheep milk samples were obtained from Maryoot Research Station, Al-Amariya, Alexandria, Egypt.

B. Bacterial Strains

All bacterial strains include *Bifidobacterium bifidum* (ATCC15708), *Lactobacillus acidophilus* (ATCC4356), *Lactobacillus delbrueckii ssp bulgaricus* (ATCC7995), *Streptococcus thermophilus* (A) as well as *Escherichia coli* (ACCT8739), *Staphylococcus aureus* (ATCC6538), *Salmonella typhimurim* (ACCT25566) and *Bacillus cereus* (ATCC9639) were obtained from MIRCEN Center, Faculty of Agriculture, Ain Shams University, Egypt.

Virgen olive oil obtained from El-Amryia local market, Alexandria, Egypt.

C. Preparation of Probiotic Barki Cheese Yoghurt (Labneh)

Barki sheep milk (1.5% fat) was divided into two portions. The first portion had no olive oil, while the second one supplemented with 1% olive oil. The two portions were heated at 90 °C/10 min, cooled to 42 °C and then subdivided into four equal portions which inoculated with *Bifidobacterium bifidum* (ATCC15708), *L. acidophilus* (ATCC4356), *Lactobacillus delbrueckii ssp bulgaricus* (ATCC35009) and *Streptococcus thermophiles* (DSM20259), respectively. All portions with olive oil treatments were inoculated with four strains of pathogenic bacterial cultures as *Escherichia coli* (ACCT8739) (portion 1), *Staphylococcus aureus* (ATCC6538) (portion 2), *Bacillus cereus* (ATCC9639) (portion 3) and *Salmonella typhimurium* (ACCT25566) (portion 4). Then all portions were incubated at 42 °C for 24 hours. After fermentation. The coagulum was placed in a hanging cloth bag to drain the whey [9]. Labneh samples of all treatments were analysed at 1, 7, 14 and 21 days of storage for PH, viable cell counts of probiotic bacteria, pathogenic bacteria, and antibacterial activity.

D. Microbial Growth and Enumeration of Microorganisms

Samples (10 g) of contaminated Barki cheese yoghurt were taken after 0, 12, 24, 36, 48, 60 and 72 hours while, control samples after 1, 7, 14, and 21 days. Then mixed with 90 ml of 0.1% sterilized peptone water and then homogenized for 5 min with lab blender (MX32). Homogenized samples were serially diluted in peptone solution and plated for bacterial enumeration according to pour plate method. 1 ml of the serial diluted samples were inoculated into molten MRS media, M17 media, MacConkey media, Mannitol salt agar media, Nutrient agar media, Sabouraud dextrose agar media for *Bifidobacterium bifidum*, *L. acidophilus*, *L. bulgaricus*, *Enterobacteria spp*, *Staphylococcus spp* count, *Bacillus spp* count and Yeast and Moulds count, respectively. Plates were incubated at 37 °C for 48 hours. Then CFUs of the microbes were counted on plates. The experiments were performed in quadruplicates and then the average of four parallel measurements of count in CFU/ml was reported.

E. Antibacterial Activity Measurement

Antibacterial activity of supernatant of yoghurt cheese supplemented with olive oil against *Escherichia coli* (ACCT8739), *Staphylococcus aureus* (ATCC6538), *Bacillus cereus* (ATCC9639) and *Salmonella typhimurium* (ACCT25566) was carried out at 1, 7, 14 and 21 days of storage at 5 °C. The experiments were performed in quadruplicates and the average of the four parallel and

measurements of inhibition zone in cm was reported.

III. RESULTS AND DISCUSSION

Table I shows the PH values of different probiotic Barki cheese yoghurt supplemented with olive oil (1%) at 1, 7, 14 and 21 days of storage at 5 °C. The PH values of both control and olive oil cheese yoghurt samples were decreased with time progress, however, no significant difference between both treatments.

TABLE I: CHANGES IN PH VALUES OF BARKI SHEEP CHEESE YOGHURT MADE USING DIFFERENT PROBIOTIC STRAINS AND SUPPLEMENTED WITH OLIVE OIL AND STORED FOR 21 DAYS AT 5 °C.

Probiotic Strain	Olive oil %	Initial PH	Storage Dayes			
			1	7	14	21
<i>Bifidobacterium bifidum</i>						
(ATCC15708)	0.0	6.62	5.42	5.35	5.30	5.19
	1.0	6.62	5.45	5.38	5.32	5.30
<i>L. acidophilus</i>						
(ATCC4356)	0.0	6.61	5.46	5.34	5.30	5.30
	1.0	6.61	5.48	5.43	5.44	5.26
<i>Strep thermophilus</i>						
(DSM20259)	0.0	6.60	5.54	5.47	5.30	5.12
	1.0	6.60	5.57	5.45	5.33	5.18
<i>L. bulgaricus</i>						
(ATCC7995)	0.0	6.62	5.55	5.44	5.35	5.12
	1.0	6.62	5.58	5.48	5.40	5.18

Table II shows the growth rates of probiotic bacteria in Barki cheese yoghurt supplemented with olive oil (1%) at 1, 7, 14 and 21 days of storage at 5 °C. In all control samples the viable cell count of *Bifidobacterium bifidum* (ATCC15708), *Lactobacillus acidophilus* (ATCC4356), *Lactobacillus delbrueckii ssp bulgaricus* (ATCC35009) and *Streptococcus thermophiles* (DSM20259) were increased gradually up to 14th day of storage and then decreased. However, in olive oil (1%) treatment, it was noticed that the activity of all probiotic strains was at highest level at 7th day of storage then decreased. Generally, no significant difference between control and olive oil treatment in viable count of all probiotic. This mean that olive oil had no effect of the activity of all probiotic bacterial strains. Thus, it can be proposed that the addition of olive oil (1%) to all studied probiotic bacterial strains is a suitable combination, and sufficient to exert the health benefits of the host. These results were agreement to [27]. The minimum required level of probiotic bacteria to be useful for the consumer's body is 10⁷ cfu/ml of living bacteria and the level in the present study was found to be 10¹⁰, thus, it could be beneficial for the consumers [27].

It was reported that combined either aqueous or alcoholic olive leaf extracts with skim milk containing *Bifidobacterium* infants and *Lactobacillus acidophilus* increased viability of both probiotic strains were observed after 16 h incubation period for olive extract concentration below 3.0 mg Catechin Equivalents /ml, whereas adding 5.0 mg Catechin Equivalents /ml resulted in significantly decreased probiotic viability [28]. The concentration at which olive oil inhibited microbial growth was greater than a previous study where 2 ml of olive oil exerted bactericidal activity against probiotic microorganisms such as *L. acidophilus* and *Bifidobacterium bifidum* when placed in bactericidal activity assay [29]. In other study, mixed cinnamon or licorice with probiotic yoghurt containing *Lactobacillus acidophilus* LA-5 and NCMF, *B. lactis* BB-12, and *Lactobacillus casi* LC-10 were

evaluated [30]. It was found that both the addition of cinnamon or licorice increased antioxidant activity in the probiotic yoghurt and correspondingly, viability of *Lactobacillus* species was reduced during fermentation and storage. A small amount of red ginseng extract promoted the growth of Lactic acid bacteria; however, a large amount inhibited the growth of them [31]. In a study on the effects of

soya powder on the growth of *Lactobacillus acidophilus* and *Bifidobacterium bifidum*, in probiotic products, it was demonstrated that the shelf life for the acidity reaching the desired level during incubation decreased for the milk with both bacteria and combined soya and malt, compared to the milk with only soya [27], [32].

TABLE II: CFU/ML OF PROBIOTIC BACTERIAL STRAINS IN BARKI SHEEP CHEESE YOGHURT MADE USING DIFFERENT PROBIOTIC STRAINS AND SUPPLEMENTED WITH OLIVE OIL AND STORED FOR 21 DAYS AT 5 °C

Probiotic Strain	Olive oil %	Storage Days			
		1	7	14	21
<i>Bifidobacterium</i>	0.0	32.0×10 ¹⁰	43.2×10 ¹⁰	45.1×10 ¹⁰	29.5×10 ¹⁰
<i>bifidum</i> (ATCC15708)	1.0	12.55×10 ¹⁰	22.32×10 ¹⁰	20.0×10 ¹⁰	9.25×10 ¹⁰
<i>L. acidophilus</i>	0.0	30.0×10 ¹⁰	40.28×10 ¹⁰	42.32×10 ¹⁰	38.21×10 ¹⁰
(ATCC4356)	1.0	13.22×10 ¹⁰	15.30×10 ¹⁰	8.32×10 ¹⁰	5.32×10 ¹⁰
<i>Strep thermophilus</i>	0.0	40.3×10 ¹⁰	49.2×10 ¹⁰	58.1×10 ¹⁰	53.17×10 ¹⁰
(DSM20259)	1.0	15.20×10 ¹⁰	26.4×10 ¹⁰	22.0×10 ¹⁰	18.1×10 ¹⁰
<i>L. bulgaricus</i>	0.0	35.0×10 ¹⁰	40.20×10 ¹⁰	50.33×10 ¹⁰	46.11×10 ¹⁰
(ATCC7995)	1.0	14.53×10 ¹⁰	23.11×10 ¹⁰	20.0×10 ¹⁰	16.22×10 ¹⁰

It was demonstrated that Vanillin promoted the growth of the bacteria in probiotic milk and yoghurt [33]. Taking into account the results of the above- mentioned studies investigating the effects of malt, soya, ginseng and dill on the growth of *L. acidophilus* and *B. bifidum*, we can conclude that they all enhance the bacterial growth in dairy products. Results revealed also that no *Enterobacteria spp* or *Staphylococcus spp* were present in control or olive oil treatment. Yeast and molds were also not detected through storage in olive oil treatment, however control treatment had Yeast and Moulds at the end of storage period 21st day. These results are in accordance with that reported by EL-Sayed *et al.* [9] and Thabet *et al.* [13].

Tables (III-VI) show zone inhibition assay of supernatant of uncontaminated Barki cheese yoghurt supplemented with olive oil 1% containing different probiotic bacteria against pathogenic bacteria stored for 1, 7, 14 and 21 days at 5 °C. Results showed that the most sensitive microorganism was *E. coli* (ACCT8739), followed by *Salmonella typhimurium* (ACCT25566) then *Bacillus cereus* (ATCC9639) and the most resistant one was *Staphylococcus aureus* (ATCC6538). *Streptococcus thermophilus* (DSM20259) supernatant had the highest inhibition zone against *E. coli* (CCT8739), *Salmonella typhimurium* (ATCC25566) and *Bacillus cereus* (ATCC9639). In olive oil 1% treatment, *Bifidobacterium bifidum* (ATCC15708) have the highest inhibition zone against *Staphylococcus aureus* (ATCC6538). Our results are similar to that reported by Eduardo *et al.* [34], they stated that olive oil had a strong bactericidal action against broad spectrum of microorganisms, this effect being higher in general against Gram-positive than Gram -negative bacteria. Cornu *et al.* [35] studied inhibiting effect of phenolic compounds on probiotic bacteria, according to their constitution and concentration and the bacterial strain. The growth rate stimulation by phenolic compounds and the increase in cell incubation could be related to their ability to metabolize these phenolic compounds [36]-[38]. Olive oil had fatty acids that are responsible for the bactericidal action. Although it has been reported that fatty acids possess antimicrobial activity against bacteria and Yeasts [39] and Alfa-Beta –unsaturated aldehydes from olives and olive oil flavor have also been demonstrated to possess antibacterial

activity against pathogens of human intestinal and respiratory tracts [40], [41].

TABLE III: ANTIBACTERIAL ACTIVITY OF PROBIOTIC SUPERNATANT OF YOGHURT CHEESE SUPPLEMENTED WITH OLIVE OIL AGAINST *ESCHERICHIA COLI* (ACCT8739)

COLI(ATCC18759)								
Storage (days)	<i>Bifidobacterium bifidum</i> (ATCC15708)		<i>L. acidophilus</i> (ATCC4356)		<i>L. bulgaricus</i> (ATCC7995)		<i>Strep thermophilus</i> (DSM20259)	
	Olive oil %							
	0.0	1.0	0.0	1.0	0.0	1.0	0.0	1.0
	Zone inhibition assay *(cm)							
1	0.6	1.0	0.7	0.9	1.0	0.9	1.0	1.0
7	1.3	2.0	1.3	1.9	1.7	2.0	1.9	2.4
14	1.2	1.9	1.3	1.6	1.6	2.0	1.9	2.4
21	1.0	1.8	1.0	1.6	1.6	1.9	1.7	2.2

TABLE IV: ANTIBACTERIAL ACTIVITY OF PROBIOTIC SUPERNATANT OF YOGHURT CHEESE SUPPLEMENTED WITH OLIVE OIL AGAINST *BACILLUS CEREUS* (ATCC6538)

Storage (days)	LAKES (ATCC6356)							
	<i>Bifidobacterium</i> <i>bifidum</i> (ATCC15708)		<i>L.</i> <i>acidophilus</i> (ATCC4356)		<i>L.</i> <i>bulgaricus</i> (ATCC7995)		<i>Strep</i> <i>thermophilus</i> (DSM20259)	
	Olive oil %							
	0.0	1.0	0.0	1.0	0.0	1.0	0.0	1.0
Zone inhibition assay * (cm)								
1	1.0	1.9	0.8	0.9	0.8	1.0	0.9	1.2
7	2.1	2.6	1.2	2.0	1.3	2.1	1.4	2.1
14	2.0	2.5	1.2	2.0	1.2	2.1	1.3	2.1
21	2.0	2.4	1.0	1.9	1.0	2.0	1.1	2.0

TABLE V: ANTIBACTERIAL ACTIVITY OF PROBIOTIC SUPERNATANT OF YOGHURT CHEESE SUPPLEMENTED WITH OLIVE OIL AGAINST *STAPHYLOCOCCUS AUREUS* (ATCC6538)

Storage (days)	Lactobacillus acidophilus (ATCC4356)							
	<i>Bifidobacterium</i>		<i>L.</i>		<i>L.</i>		<i>Strep</i>	
	<i>bifidum</i>		<i>acidophilus</i>		<i>bulgaricus</i>		<i>thermophilus</i>	
	(ATCC15708)		(ATCC4356)		(ATCC7995)		(DSM20259)	
	Olive oil %							
	0.0	1.0	0.0	1.0	0.0	1.0	0.0	1.0
	Zone inhibition assay * (cm)							
1	0.8	1.0	0.7	1.0	0.8	1.0	0.9	1.2
7	1.1	2.2	1.2	2.1	1.2	2.2	1.3	2.3
14	1.0	2.2	1.1	2.0	1.2	2.2	1.2	2.2
21	1.0	2.0	1.0	2.0	1.0	2.2	1.1	2.1

TABLE VI: ANTIBACTERIAL ACTIVITY OF PROBIOTIC SUPERNATANT OF YOGHURT CHEESE SUPPLEMENTED WITH OLIVE OIL AGAINST *SALMONELLA* *TYPHIMURIUM* (ATCC25566)

Storage (days)	<i>Bifidobacterium</i>		<i>L.</i>		<i>L.</i>		<i>Strep</i>	
	<i>bifidum</i>		<i>acidophilus</i>		<i>bulgaricus</i>		<i>thermophilus</i>	
	(ATCC15708)		(ATCC4356)		(ATCC7995)		(DSM20259)	
	Olive oil %							
	0.0	1.0	0.0	1.0	0.0	1.0	0.0	1.0
Zone inhibition assay * (cm)								
1	0.9	1.0	0.8	1.0	1.0	1.4	1.0	1.5
7	1.2	2.2	1.2	1.4	1.4	2.4	1.6	2.6
14	1.2	2.1	1.1	2.1	1.3	2.3	1.6	2.5
21	1.1	2.1	1.0	2.1	1.2	2.3	1.5	2.4

*The experiments were performed in quadruplicates and then the average of the four parallel measurements of the inhibition zone in cm were reported.

Tables (VII-X) show the antibacterial activity of contaminated Barki cheese yoghurt supplemented with olive oil 1% at storage time 0, 12, 24, 36, 48, 60 and 72 hours at 5 °C against *E. coli* (ACCT8739), *Salmonella typhimurium* (ACCT25566), *Bacillus cereus* (ATCC9639) and *Staphylococcus aureus* (ATCC6538). The results showed various degrees of inhibition against the four bacterial strains. In olive oil 1% treatment the growth of *E. coli* (ACCT8739) and *Salmonella typhimurium* (ATCC25566) was stopped in the presence of *Streptococcus thermophiles* (DSM20259), and *L. bulgaricus* (ATCC7995) after 24 h. However, the growth of *Bacillus cereus* (ATCC9639) was stopped with all different probiotic bacteria after 36 h. *Bifidobacterium bifidum* (ATCC15708) was stopped the growth of

Staphylococcus aureus (ATCC6538) after 24 h.

Markrin *et al.* [42] reported that olive leaf extract was able to inhibit the growth of *C. albicans* after 1 day of contact. It must be assumed that active compounds of the olive leaf extract, mainly phenolic compounds, which are different from olive oil. Olive leaf extract was more inhibitory against *E. coli* than against *C. albicans*. It has been reported that virgin olive oil was effective in preserving yoghurt cheese inoculated with the *Kluyveromyces marxianus* [43]. Phenolic compounds from berry extracts inhibited the growth of Gram –negative [44]. Olive oil exhibited bactericidal activity against *E. coli* and *C. perfringens*, which grow in the intestine. Park *et al.* [45] reported that many plant extracts and foods such as tea may improve the intestinal microbial by inhibiting harmful microorganisms but also promoting or maintaining the beneficial ones such as lactic acid bacteria [46]. The bio active compounds such as poly phenols can be absorbed before they reach the colon [47] and they are even transformed by intestinal bacteria [48] olive oil. Pretty *et al.* [49] reported that olive oil can decrease both bacterial growth and adhesion. Eduardo *et al.* [34] found that olive oil has a very strong bactericidal effect against *S. mutans*. Raford *et al.* [50] observed a faster death rate of *S. enterica* in mayonnaise made with olive oil. Alwaili [51] found that honey, olive oil and bee wax have been used to treat the disease diaper dermatitis.

TABLE VII: CFU/ML OF *ESCHERICHIA COLI* (ATCC8739) INOCULATED IN BARKI SHEEP CHEESE YOGHURT MADE USING DIFFERENT PROBIOTIC STRAINS AND SUPPLEMENTED WITH OLIVE OIL AND STORED FOR 72 H AT 5°C

PROBIOTIC STRAINS AND SUPPLEMENTED WITH OLIVE OIL AND STORED FOR 72 H AT 5 °C								
Storage (h)	<i>Bifidobacterium bifidum</i> (ATCC15708)		<i>L. acidophilus</i> (ATCC4356)		<i>L. bulgaricus</i> (ATCC7995)		<i>Strep thermophilus</i> (DSM20259)	
	Olive oil %							
	0.0	1.0	0.0	1.0	0.0	1.0	0.0	1.0
0	8.22×10 ⁷	9.2×10 ⁷	7.7×10 ⁷	8.8×10 ⁸	8.4×10 ⁷	8.9×10 ⁷	8.44×10 ⁷	10.0×10 ⁸
12	3.0×10 ⁵	2.0×10 ⁸	6.0×10 ⁶	5.3×10 ⁶	3.3×10 ⁶	8.0×10 ⁵	10.0×10 ⁷	5.3×10 ⁴
24	5.0×10 ³	3.3×10 ⁴	5.0×10 ⁵	3.5×10 ⁸	5.2×10 ⁴	—	7.0×10 ⁶	—
36	6.0×10 ²	—	9.0×10 ³	—	6.8×10 ²	—	5.0×10 ²	—
48	—	—	—	—	—	—	—	—
60	—	—	—	—	—	—	—	—
72	—	—	—	—	—	—	—	—

TABLE VIII: CFU/ML OF *STAPHYLOCOCCUS AUREUS* (ATCC6538) INOCULATED IN BARKI SHEEP CHEESE YOGHURT MADE USING DIFFERENT PROBIOTIC STRAINS AND SUPPLEMENTED WITH OLIVE OIL AND STORED FOR 72 H AT 5°C

STRAINS AND SUPPLEMENTED WITH OLIVE OIL AND STORED FOR 12 H AT 5 °C								
Storage (hrs)	<i>Bifidobacterium</i> (ATCC15708)		<i>L. acidophilus</i> (ATCC4356)		<i>L. bulgaricus</i> (ATCC7995)		<i>Streptococcus thermophilus</i> (DSM20259)	
	Olive oil %							
	0.0	1.0	0.0	1.0	0.0	1.0	0.0	1.0
0	8.1×10 ⁷	8.9×10 ⁷	6.6×10 ⁷	7.3×10 ⁷	8.7×10 ⁷	6.0×10 ⁷	8.3×10 ⁷	7.4×10 ⁷
12	3.0×10 ⁸	5.2×10 ⁷	7.0×10 ⁶	2.0×10 ⁶	6.4×10 ⁹	7.7×10 ⁵	5.7×10 ⁹	1.3×10 ⁶
24	4.0×10 ⁵	—	9.0×10 ⁷	6.2×10 ³	4.3×10 ⁸	2.2×10 ²	4.2×10 ⁶	4.4×10 ²
36	4.0×10 ²	—	4.0×10 ⁵	—	5.0×10 ⁴	—	5.6×10 ⁴	—
48	—	—	8.0×10 ³	—	2.0×10 ²	—	7.2×10	—
60	—	—	—	—	—	—	—	—
72	—	—	—	—	—	—	—	—

TABLE IX: CFU/ML OF *BACILLUS CEREUS* (ATCC9639) INOCULATED IN BARKI SHEEP CHEESE YOGHURT MADE USING DIFFERENT PROBIOTIC STRAINS AND SUPPLEMENTED WITH OLIVE OIL AND STORED FOR 72 HOURS AT 5 °C

AND SUPPLEMENTED WITH OLIVE OIL AND STORED FOR 72 HOURS AT 5 °C								
Storage (hrs)	<i>Bifidobacterium</i> (ATCC15708)		<i>L. acidophilus</i> (ATCC4356)		<i>L. bulgaricus</i> (ATCC7995)		<i>Streptococcus thermophilus</i> (DSM20259)	
	0.0	1.0	0.0	1.0	0.0	1.0	0.0	1.0
0	7.8×10 ⁷	8.14×10 ⁷	8.29×10 ⁷	9.18×10 ⁷	9.29×10 ⁷	8.5×10 ⁷	6.19×10 ¹⁰	9.8×10 ⁷
12	3.3×10 ¹⁰	6.8×10 ⁶	6.7×10 ⁹	6.2×10 ⁶	6.12×10 ⁹	6.6×10 ⁵	5.2×10 ⁸	9.0×10 ⁵
24	7.7×10 ⁷	4.4×10 ³	4.5×10 ⁷	8.8×10 ⁴	5.4×10 ⁸	8.2×10 ²	4.9×10 ⁶	2.0×10 ²
36	9.9×10 ⁴	_____	6.8×10 ⁵	_____	6.0×10 ⁷	_____	5.7×10	_____
48	5.2×10 ²	_____	3.9×10 ⁴	_____	3.3×10 ³	_____	_____	_____
60	_____	_____	2.3×10 ²	_____	_____	_____	_____	_____
72	_____	_____	_____	_____	_____	_____	_____	_____

TABLE X: CFU/ML OF SALMONELLA TYPHIMURIUM (ATCC25566) INOCULATED IN BARKI SHEEP CHEESE YOGHURT MADE USING DIFFERENT PROBIOTIC STRAINS AND SUPPLEMENTED WITH OLIVE OIL AND STORED FOR 72 HOURS AT 5 °C

STRAINS AND SUPPLEMENTED WITH OLIVE OIL AND STORED FOR 72 HOURS AT 5 °C								
Storage (hrs)	<i>Bifidobacterium</i> (ATCC15708)		<i>L. acidophilus</i> (ATCC4356)		<i>L. bulgaricus</i> (ATCC7995)		<i>Streptococcus thermophilus</i> (DSM20259)	
	Olive oil %							
	0.0	1.0	0.0	1.0	0.0	1.0	0.0	1.0
0	8.16×10 ⁷	7.7×10 ⁷	8.19×10 ⁷	9.9×10 ⁷	8.8×10 ⁷	7.8×10 ⁷	6.8×10 ⁷	8.8×10 ⁷
12	9.0×10 ¹⁰	5.0×10 ⁶	3.0×10 ⁶	6.0×10 ⁶	7.4×10 ⁹	3.0×10 ⁵	9×10 ¹⁰	6.8×10 ³
24	5.0×10 ⁹	4.4×10 ⁵	7.0×10 ⁷	7.2×10 ⁴	6.6×10 ⁷	_____	8.3×10 ⁷	_____
36	7.0×10 ⁶	_____	5.2×10 ⁵	_____	8.7×10 ⁵	_____	6.8×10	_____
48	2.0×10 ²	_____	6.5×10 ²	_____	4.4×10 ²	_____	_____	_____
60	_____	_____	_____	_____	_____	_____	_____	_____
72	_____	_____	_____	_____	_____	_____	_____	_____

IV. CONCLUSION

Consumers are more willing to accept natural extracts as preservatives than synthetic ones due to the nature and relative safety of natural products. From the results obtained, it could be concluded that olive oil can be added to fermented milk to increase both the quality and the nutritional value of the final dairy product, without inducing any negative effects on the viability of the lactic acid bacteria. Using olive oil (1%) had high antibacterial effect against the four tested pathogens. *E. coli* (ACCT8739) was more sensitive, followed by *Salmonella typhimurium* (ATCC25566) and the most resistant microorganism was *Staphylococcus aureus* (ATCC6538). Olive oil (1%) had no effect against the growth of different probiotic bacterial strains. The antibacterial activity of cheese yoghurt supplemented with olive oil (1%) was higher than control treatment because olive oil supported the growth of Lactic acid bacteria. It is recommended that olive oil (1%) can be used as a natural and safe anti-microbial substance in Barki cheese yoghurt and other dairy products, and olive oil may well have the beneficial role in promoting probiotic bacteria and inhibiting harmful bacteria.

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