

## EVALUATION OF LABNEH MADE FROM COWS' MILK AND GOATS' MILK USING PROBIOTIC AND EXOPOLYSACCHARIDE - PRODUCING BACTERIA

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**ABSTRACT:** Many lactic acid bacteria strains can biosynthesis exopolysaccharides (EPS) that are added to many types of food to enhance their rheological properties, the aim of this work was to manufacture low fat Labneh using mixture of cows and goats' milk with EPS - producing probiotic starter. This starter consisted of *Streptococcus thermophilus* TA061, MR-1C+ *Lactobacillus delbrueckii* subsp. *bulgaricus* MR-1R+ *Bifidobacterium bifidum* and *Lactobacillus acidophilus*. The results indicated that the use of these bacteria increased the yield, total solids and fat in dry matter of Labneh prepared from mixture of cow and goat's milk containing 1% fat (CG1). The experimental Labneh was ranked higher scoring points compared with Labneh made from goat's milk 3% fat (G) and made with the use of traditional starter.

**Key words:** Mixtures of cow's milk and goat's milk, Labneh, probiotic, organoleptic properties, exopolysaccharides lactic acid bacteria.

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### INTRODUCTION

The world production of goat's milk is in a continuous increase. According to the Food and Agriculture Organization (FAO), goat's milk production reached approximately 12,8 MT in 2008 (FAO, 2011), There is a growing demand for goat's milk by consumers mainly due to the increasing number of children suffering from cow's milk allergy. Goat's milk and its products have been characterized by higher digestibility and certain nutritional and therapeutic effects in human nutrition (Haenlein, 2004). Although goat's milk has a higher fat content of strong smelling than cow and buffalo's milk (Park *et al.*, 2007) the good production and handling of goat's milk yield milk indistinguishable in taste and odor from cow's milk.

Labneh (concentrated yoghurt), is a semi solid fermented dairy food produced by removing part of the whey from yoghurt until the total solids level reaches between 23 and 25%, of which 8-11% is fat (Tamime and Robinson, 1999). The homogenization

of traditional and UF Labneh made from goat and sheep's milk produced a smoother texture, but markedly decreased the firmness compared with Labneh made from cow's milk. In addition, goat and sheep's Labneh had similar microstructure but less uniform than Labneh made from cow's milk (Tamime *et al.*, 1991). Recently, it could be avoid the goaty flavour, e.g. using adding aromatic oils (Abou ayana and Gamal El deen, 2011) and/ or might be mix two or three types of milk.

An alternative class of bio-thickeners is the microbial exopolysaccharides (EPS). Microbial exopolysaccharides are extracellular polysaccharides that are either associated with the cell surface in the form of capsules or secreted into the extracellular environment in the form of slime. They are referred to as capsular or slime exopolysaccharides, respectively. EPS occur widely among bacteria and microalgae and less among yeasts and fungi (Sutherland, 1972 and Sutherland, 1990).

EPS-producing LAB increased the yield and improved the rheological properties of low-fat yoghurt and texture profile of kareish cheese (Abou Ayana and Ibrahim, 2015).

The aim of this study was to improve the quality of Labneh made from mixtures of cow's and goat's milk by using EPS-producing bacteria strains (*S. thermophilus* TA061, MR-1C, and *L. delbrueckii* subsp. *bulgaricus* MR-1R) and probiotic strains of *B. bifidum* and *L. acidophilus*.

## MATERIALS AND METHODS

**Milk:** Fresh cow and goat's milks were obtained from the herds of Sakha Station, Animal Production Research Institute, Agriculture Research Center, Egypt. Milk was analyzed and had following composition : acidity 0.17 & 0.16, fat 3.1 & 3.8 specific gravity 27.7 & 28.3 /60°F, lactose 4.2 & 4.4 and pH 6.61 & 6.62 for goat and cow's milk, respectively. Low heat spray dried skim milk powder, of Dutch origin was used.

**Bacterial cultures:** The used bacterial mixtures in this study are listed in Table 1. All bacterial cultures were maintained by biweekly transfer, incubated at 37°C, and stored at 4°C. *St. thermophilus* TA061 MR-1C, were grown in M17 broth (Terzaghi and Sandine, 1975) containing 0.5% lactose, while *L. delbrueckii* subsp. *bulgaricus* MR-1R was cultured in MRS broth (De-Man *et al.*, 1960) and both were obtained from Chr. Hansen's Laboratory, Copenhagen, Denmark. *Lactobacillus acidophilus* and *Bifidobacterium bifidum* were obtained from Laboratorium Wiesby, Niebull, Germany. *B. bifidum* and *L. acidophilus* were separately transferred into sterile skim milk containing 10 g dextrose and 1g yeast extract /L. *L. acidophilus* was incubated at 37 °C until coagulation, while *Bifidobacterium bifidum* was incubated anaerobically at 37 °C until

coagulation. Further activation was achieved by three similar successive transfers in the same medium (Beena and Prasad, 1997).

**Making of Labneh:** Whole fresh cow's milk and goat's milk were standardized to 3% milk fat, mixtures of cow and goat' milk (1:1) contained 1, 2 and 3% milk fat, using fresh skim milk, and all portions of milk were standardized to 14% TS using skimmed milk powder. All milk samples were heated at 90°C for 15 min; homogenized at 250 kg cm<sup>-2</sup>/60°C cooled to 40°C then inoculated with 2 % of the starter cultures as in Table 1. Incubation was carried out at 40°C until pH 4.8. Fermented milks from all treatments were cooled to 10°C overnight, mixed and transferred into sterilized cloth bags, hanged in cooling room at 6-8°C to allow whey drainage for 12 h. NaCl (0.5%) was added to the bag contents, mixed thoroughly and the resultant Labneh was filled into plastic containers and stored at 6-8°C for 21 days. Three replicates of each treatment were made. The Labneh Samples were chemically, microbiologically and organoleptically analyzed as well as the yield of each treatment was determined when fresh and after 7, 14 and 21 days.

**Chemical analysis:** Titratable acidity (TA %), total solids (TS %) and fat contents were determined according to Ling (1963). Acetaldehyde was determined as given by Lees and Jago (1969). Diacetyl was determined as described by Westerfeld (1945) and lactose content was colorimetrically determined according to the method reported by Nickerson *et al.*, (1976). Tyrosine and tryptophan contents (mg/100) were determined according to Vakaleris and Price (1959). Total volatile free fatty acids (TVFA expressed as ml. 0.1N NaOH/10 gm) were estimated as described by Kosikowski (1982).

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**Table 1: The milk and bacterial cultures used in treatments of Labneh**

The milk	Type of cultures	Abbreviation
Cow's milk 3% fat (control)	4% <i>St. thermophilus</i> + <i>L. delbrueckii</i> subsp. <i>bulgaricus</i> (traditional) (1:1)	C
Goat's milk 3% fat	4% <i>St. thermophilus</i> + <i>L. delbrueckii</i> subsp. <i>bulgaricus</i> (traditional) (1:1)	G
Cow' milk + Goat's milk 1:1(3% fat, 1:1)	3% <i>St. thermophilus</i> TA061, MR-1C + <i>L. delbrueckii</i> subsp. <i>bulgaricus</i> MR-1R+ ( 1% <i>B. bifidum</i> and <i>L. acidophilus</i> ) (1:1)	CG3
Cow' milk + Goat's milk 1:1(2% fat, 1:1)	3% <i>St. thermophilus</i> TA061, MR-1C + <i>L. delbrueckii</i> subsp. <i>bulgaricus</i> MR-1R+(1 % <i>B. bifidum</i> and <i>L. acidophilus</i> (1:1)	CG2
Cow' milk + Goat's milk 1:1(1% fat, 1:1)	3% <i>St. thermophilus</i> TA061, MR-1C + <i>L. delbrueckii</i> subsp. <i>bulgaricus</i> MR-1R+( 1% <i>B. bifidum</i> and <i>L. acidophilus</i> (1:1)	CG1

**Microbiological tests:** *Bifidobacterium bifidum* was determined according to Dave and Shah (1996) using modified MRS agar supplemented with 0.05% L-cystein and 0.3% lithium chloride. *L. acidophilus* was enumerated according to Dave and Shah (1996) using MRS-sorbitol agar. Lactic acid bacteria count was determined according to the methods described by the American Public Health Association (1992). Total viable bacterial count was determined according to (APHA, 1978). The count of spore-forming bacteria was determined according to Chalmer (1962). Counts of coliform bacteria were enumerated using the method described by (APHA, 1960). MacConkey agar was prepared as described by Oxoid (1982) for determining Enterobacteriaceae. Counts of psychrotrophic bacteria were estimated by using PCS medium (Bridson, 1990). Potato dextrose agar recommended by the Oxoid (1962) was used for the enumeration of moulds and yeasts. *Staphylococcus* medium No.110 (Difco, 1974) was used to count and detect staphylococci.

**Organoleptic evaluations:** All Labneh samples were sensory evaluated by eleven of the staff of Sakha Station, Animal

Production Research Lab, according to Ahmed and Ismail (1978). The score points were 60 for flavour, 30 for body & texture and 10 for appearance which give a total score of 100 points.

**Statistical Analysis:** The obtained results were statistically analyzed using a software package (SAS, 1991) based on analysis of variance. When F-test was significant, least significant difference (LSD) was calculated according to Duncan (1955) for the comparison between means.

**RESULTS AND DISCUSSION**  
**Yield and chemical analysis of Labneh:**

Results presented in Table 2 illustrated that yield; TS and F/DM of fresh Labneh was affected by both the milk type and bacterial strain of starter used. Yield, TS and F/DM of fresh and stored Labneh were higher in control (C) than in goats milk Labneh (G). Using starter [3% *St. thermophilus* TA061, MR-1C + *L. delbrueckii* subsp. *bulgaricus* MR-1R+(1 % *B. bifidum* and *L. acidophilus* (1:1)] increased yield, TS and F/DM in CG3, CG2 and CG1 treatments despite the low fat in CG2 and CG1 (2 and 1% fat respectively). CG3 was exhibited the highest yield followed

by CG2, CG1, C then G treatments, TS and F/DM nearly took the same trend. There were statistical differences in yield, TS and F/DM after the seventh day of storage. These results might be due to the fact that these strains produce exopolysaccharide (EPS) that is tightly associated with the bacterial cell wall (capsular EPS) or liberated into the growth medium (Ropy EPS). Since EPS have viscosity enhancing and stabilizing properties. EPS-producing strains are commonly used to enhance water binding and viscosity in yoghurt and fermented milks and decrease the syneresis defect and improve yields in cheeses (Broadbent *et al.* 2001). These findings agreed with those reported by Low *et al.*, (1998) and Perry *et al.*, (1997). Genetic studies demonstrated that this effect was due to the MR-1C capsular EPS, and industrial cheese trials confirmed that MR-1C can result a 1.5% moisture increase in

part-skim Mozzarella (Broadbent *et al.*, 2011).

**Titrateable Acidity (TA):** Other chemical properties of fresh and stored Labneh were summarized in Table 3, TA all treatments increased rapidly till the fourteenth day (1.2-1.3%) but it increased slightly during the last week of storage (1.3 – 1.42%), These results correspond with Abou ayana and Ibrahim (2015). This was probably due to the adaptation of bacteria in the middle of storage period. Statistically, G treatment was the highest TA compared to other treatments, this perhaps because of increase the activity of traditional LAB in the goat's milk. Probiotic culture ABT (*B. bifidum*, *L. acidophilus* and *St. thermophilus*) has a mild acidic (Kurmann *et al.*, 1992) this might explain our findings. Amer *et al.*, (1998) mentioned that yoghurt (CH-N2) culture produced the greatest TA followed by cheese (CH-N1) culture then ABT culture.

**Table 2: The fat, yield and TS in dry matter (F/DM) of fresh and stored Labneh made from cow and goat's milk with EPS lactic acid bacteria (means ± SD)**

Items	Treatments	Storage period (day)				Means ± SD
		Fresh	7	14	21	
Yield (%)	C	29.0 <sup>a</sup>	28.4 <sup>a</sup>	26.5 <sup>c</sup>	24.2 <sup>c</sup>	27.02±2.01 <sup>D</sup>
	G	28.4 <sup>a</sup>	27.7 <sup>a</sup>	25.9 <sup>c</sup>	23.7 <sup>d</sup>	26.40±1.92 <sup>E</sup>
	CG3	30.5 <sup>a</sup>	30.0 <sup>a</sup>	29.1 <sup>b</sup>	27.5 <sup>d</sup>	29.28±1.48 <sup>A</sup>
	CG2	29.7 <sup>a</sup>	29.2 <sup>a</sup>	28.1 <sup>c</sup>	26.2 <sup>d</sup>	28.25±1.38 <sup>B</sup>
	CG1	28.6 <sup>a</sup>	28.2 <sup>a</sup>	27.0 <sup>c</sup>	25.1 <sup>f</sup>	27.20±1.43 <sup>C</sup>
TS (%)	C	26.3 <sup>a</sup>	26.2 <sup>a</sup>	26.1 <sup>ab</sup>	26.0 <sup>d</sup>	26.15±0.16 <sup>A</sup>
	G	26.2 <sup>a</sup>	26.1 <sup>b</sup>	26.0 <sup>c</sup>	26.0 <sup>c</sup>	26.07±0.14 <sup>A</sup>
	CG3	26.5 <sup>a</sup>	26.4 <sup>c</sup>	26.3 <sup>e</sup>	26.3 <sup>e</sup>	26.38 ±0.13 <sup>A</sup>
	CG2	26.3 <sup>a</sup>	26.3 <sup>a</sup>	26.1 <sup>e</sup>	26.1 <sup>e</sup>	26.20±0.15 <sup>A</sup>
	CG1	26.2 <sup>a</sup>	26.2 <sup>a</sup>	26.0 <sup>e</sup>	26.0 <sup>e</sup>	26.10±0.15 <sup>A</sup>
F/DM (%)	C	38.0 <sup>a</sup>	38.1 <sup>a</sup>	38.0 <sup>a</sup>	37.6 <sup>d</sup>	37.92±0.23 <sup>A</sup>
	G	37.7 <sup>a</sup>	37.7 <sup>a</sup>	37.5 <sup>c</sup>	37.2 <sup>d</sup>	37.52±0.24 <sup>B</sup>
	CG3	37.5 <sup>a</sup>	37.5 <sup>a</sup>	37.3 <sup>b</sup>	36.8 <sup>d</sup>	37.28 ±0.24 <sup>A</sup>
	CG2	37.0 <sup>a</sup>	37.0 <sup>a</sup>	36.8 <sup>b</sup>	36.2 <sup>d</sup>	36.75±0.37 <sup>C</sup>
	CG1	36.0 <sup>a</sup>	36.0 <sup>a</sup>	36.1 <sup>a</sup>	36.1 <sup>a</sup>	36.05±0.12 <sup>D</sup>

<sup>abcde</sup> Letters indicate significant differences between Labneh treatments, <sup>ABCDE</sup> Letters indicate the means of significant differences between Labneh treatments. C, G, CG3, CG2 and CG1: see table 1

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**Table 3: Chemical properties of experimental Labneh made from mixtures of cow and goat's milk using EPS lactic acid bacteria (means  $\pm$  SD)**

Properties	Treatments	Storage period (day)				Means $\pm$ SD
		Fresh	7	14	21	
Acidity (%)	C	0.79 <sup>d</sup>	0.99 <sup>c</sup>	1.20 <sup>a</sup>	1.30 <sup>a</sup>	1.07 $\pm$ 0.21 <sup>B</sup>
	G	0.81 <sup>d</sup>	0.98 <sup>d</sup>	1.30 <sup>b</sup>	1.42 <sup>a</sup>	1.12 $\pm$ 0.25 <sup>A</sup>
	CG3	0.80 <sup>e</sup>	0.97 <sup>d</sup>	1.25 <sup>b</sup>	1.40 <sup>a</sup>	1.11 $\pm$ 0.23 <sup>A</sup>
	CG2	0.78 <sup>d</sup>	0.96 <sup>c</sup>	1.20 <sup>b</sup>	1.30 <sup>a</sup>	1.06 $\pm$ 0.22 <sup>A</sup>
	CG1	0.78 <sup>d</sup>	0.97 <sup>c</sup>	1.20 <sup>b</sup>	1.30 <sup>a</sup>	1.06 $\pm$ 0.22 <sup>A</sup>
Acetaldehyde ( $\mu$ mol/100 g)	C	271 <sup>c</sup>	292 <sup>a</sup>	305 <sup>a</sup>	224 <sup>e</sup>	273.0 $\pm$ 32.9 <sup>E</sup>
	G	275 <sup>c</sup>	298 <sup>a</sup>	309 <sup>a</sup>	237 <sup>e</sup>	279.5 $\pm$ 29.5 <sup>D</sup>
	CG3	282 <sup>c</sup>	301 <sup>a</sup>	321 <sup>a</sup>	259 <sup>e</sup>	290.7 $\pm$ 67.5 <sup>A</sup>
	CG2	275 <sup>c</sup>	298 <sup>a</sup>	317 <sup>a</sup>	245 <sup>e</sup>	283.7 $\pm$ 28.7 <sup>B</sup>
	CG1	273 <sup>c</sup>	298 <sup>a</sup>	318 <sup>a</sup>	236 <sup>e</sup>	281.2 $\pm$ 32.7 <sup>C</sup>
Diacetyl ( $\mu$ mol/100 g)	C	117 <sup>c</sup>	128 <sup>b</sup>	136 <sup>a</sup>	130 <sup>b</sup>	127.7 $\pm$ 7.4 <sup>C</sup>
	G	122 <sup>d</sup>	138 <sup>b</sup>	145 <sup>a</sup>	138 <sup>b</sup>	135.7 $\pm$ 9.1 <sup>A</sup>
	CG3	124 <sup>d</sup>	139 <sup>b</sup>	145 <sup>a</sup>	138 <sup>b</sup>	136.5 $\pm$ 8.3 <sup>A</sup>
	CG2	122 <sup>d</sup>	137 <sup>b</sup>	141 <sup>a</sup>	136 <sup>b</sup>	134 $\pm$ 20.3 <sup>D</sup>
	CG1	122 <sup>d</sup>	133 <sup>b</sup>	140 <sup>a</sup>	134 <sup>b</sup>	132.2 $\pm$ 7.0 <sup>B</sup>
Lactose (%)	C	3.2 <sup>a</sup>	2.7 <sup>b</sup>	2.3 <sup>c</sup>	2.0 <sup>d</sup>	2.55 $\pm$ 0.49 <sup>A</sup>
	G	3.1 <sup>a</sup>	2.5 <sup>b</sup>	2.2 <sup>c</sup>	1.9 <sup>d</sup>	2.42 $\pm$ 0.49 <sup>A</sup>
	CG3	3.2 <sup>a</sup>	2.6 <sup>b</sup>	2.2 <sup>c</sup>	2.0 <sup>d</sup>	2.50 $\pm$ 0.50 <sup>A</sup>
	CG2	3.2 <sup>a</sup>	2.7 <sup>b</sup>	2.2 <sup>c</sup>	2.0 <sup>d</sup>	2.52 $\pm$ 0.50 <sup>A</sup>
	CG1	3.2 <sup>a</sup>	2.7 <sup>b</sup>	2.2 <sup>c</sup>	2.1 <sup>d</sup>	2.53 $\pm$ 0.49 <sup>A</sup>
Tyrosine (mg/100 g)	C	16.2 <sup>d</sup>	29.9 <sup>c</sup>	36.2 <sup>b</sup>	42.8 <sup>a</sup>	31.27 $\pm$ 10.5 <sup>B</sup>
	G	17.5 <sup>d</sup>	30.2 <sup>c</sup>	37.1 <sup>b</sup>	43.4 <sup>a</sup>	32.05 $\pm$ 10.3 <sup>A</sup>
	CG3	15.5 <sup>d</sup>	25.2 <sup>c</sup>	33.1 <sup>b</sup>	40.2 <sup>a</sup>	28.50 $\pm$ 9.8 <sup>C</sup>
	CG2	15.5 <sup>d</sup>	25.1 <sup>c</sup>	33.1 <sup>b</sup>	40.3 <sup>a</sup>	28.50 $\pm$ 9.9 <sup>C</sup>
	CG1	15.6 <sup>d</sup>	25.2 <sup>c</sup>	33.1 <sup>b</sup>	40.2 <sup>a</sup>	28.52 $\pm$ 9.8 <sup>C</sup>
Tryptophan (mg/100 g)	C	97.1 <sup>a</sup>	82.3 <sup>b</sup>	73.2 <sup>c</sup>	55.5 <sup>d</sup>	77.02 $\pm$ 16.11 <sup>B</sup>
	G	98.3 <sup>a</sup>	82.9 <sup>b</sup>	74.3 <sup>c</sup>	56.2 <sup>d</sup>	77.92 $\pm$ 16.26 <sup>A</sup>
	CG3	94.1 <sup>a</sup>	78.8 <sup>b</sup>	71.9 <sup>c</sup>	52.1 <sup>d</sup>	74.22 $\pm$ 16.13 <sup>C</sup>
	CG2	94.2 <sup>a</sup>	78.8 <sup>b</sup>	71.8 <sup>c</sup>	52.1 <sup>d</sup>	74.22 $\pm$ 16.17 <sup>C</sup>
	CG1	94.2 <sup>a</sup>	78.8 <sup>b</sup>	71.9 <sup>c</sup>	52.2 <sup>d</sup>	74.27 $\pm$ 16.13 <sup>C</sup>
TVFA*	C	0.33 <sup>f</sup>	0.32 <sup>f</sup>	1.30 <sup>a</sup>	1.40 <sup>a</sup>	0.849 $\pm$ 0.54 <sup>A</sup>
	CG	0.46 <sup>f</sup>	0.49 <sup>e</sup>	0.51 <sup>e</sup>	0.60 <sup>a</sup>	0.515 $\pm$ 0.06 <sup>B</sup>
	CG3	0.46 <sup>f</sup>	0.48 <sup>e</sup>	0.51 <sup>d</sup>	0.55 <sup>a</sup>	0.500 $\pm$ 0.04 <sup>CB</sup>
	CG2	0.45 <sup>f</sup>	0.46 <sup>d</sup>	0.49 <sup>c</sup>	0.52 <sup>a</sup>	0.480 $\pm$ 0.03 <sup>C</sup>
	CG1	0.41 <sup>f</sup>	0.43 <sup>e</sup>	0.46 <sup>c</sup>	0.49 <sup>a</sup>	0.447 $\pm$ 0.03 <sup>D</sup>

<sup>abcde</sup> Letters indicate significant differences between Labneh treatments, <sup>ABCDE</sup> Letters indicate the means of significant differences between Labneh treatments \* expressed as ml 0.1 NaOH 10 g<sup>-1</sup> Labneh. C, G, CG3, CG2 and CG1: see table 1

**Acetaldehyde content:** Production of flavor compounds (acetaldehyde and diacetyl) depends on the activity and type of starter used and condition of fermentation. Table 3 shows that the content samples of acetaldehyde, influenced by the type of bacterial strains used. Statistically, control Labneh contained the lowest content of acetaldehyde contrary to CG3 treatment that contained the highest acetaldehyde either in fresh or stored samples followed by CG2 then CG1 treatments. These findings might be due to a mixture of bacteria used and the type of milk, although acetaldehyde decreased slightly with the low level of fat. In all samples, acetaldehyde increased with progress the storage period till 14 d then decreased up to the end of storage period (21 day). Control and all treatments recorded 271 -282  $\mu\text{mol}/100\text{ g}$  of acetaldehyde at zero time. At the end of storing period, the levels of acetaldehyde were 224, 236, 237, 245, and 259  $\mu\text{mol}/100\text{ g}$  for C, CG1, G, CG2, and CG3, respectively. These results correspond with Amer *et al.*, (1998) Taha *et al.*, (1997).

**Diacetyl content:** Concerning diacetyl, data listed in the same table appeared that diacetyl production approximately followed similar trend of acetaldehyde production along the cold storage period. CG3 treatment exhibited the highest diacetyl in both fresh and cold stored Labneh samples but control sample contained the lowest diacetyl. These findings also correspond with Amer *et al.*, (1998) and Taha *et al.*, (1997).

**Lactose content:** As shown in Table 3, the lactose content clearly reflected the cultures activity; the changes in acidity depend on the changes in lactose which plays a great role. As expected, in G treatment had the lowest lactose content of Labneh compared with the control and other treatments; all fresh samples contained (3.1- 3.2%) lactose at zero time,

this quantity gradually decreased along with the storage period to reach between 1.9- 2.1% at the end of cold storage. Lactose content was 3.35% in fresh Labneh (Abu-Jdayil and Mohamed, 2002). These results somewhat differ from those reported by Taha *et al.*, (1997).

**Tyrosine and tryptophan content:** It is clear from the results in Table 3 that tyrosine content in all samples gradually increased as the age of the Labneh produced. Ammar (1995) reported similar results. The tyrosine contents of fresh Labneh ranged from 15.5 to 17.5 mg/100 g and increased at a variable rate to reach 40.2 to 43.4 mg/100 g after 21 days in refrigerator. Noticeable, tyrosine clearly increased in the presence of traditional culture, G treatment had the highest level of tyrosine which might be due to increase the activity of traditional culture in goat's milk more than in cow's milk. These findings confirmed with those of chervaux *et al.*, (2000).

With regard to tryptophan content, it appears that it followed similar trend of tyrosine production, G treatment contained the highest level of tryptophan and it affected by bacterial strain. Traditional bacteria produced the greatest quantities of tryptophan in G and C (98.3 -97.1 mg/100 g at zero time, respectively) and decreased as storage period to reach 56.2-55.5 mg/100 g at the end of storage period. Increases which associated G and C treatments might be due to effect of traditional culture activity. It is well known that therapeutic bacteria have little proteolytic ability on protein compared with yoghurt culture. These results are similar to those of Amer *et al.*, (1998).

**TVFA content:** The total volatile fatty acids content were affected by the type of milk, the percent of fat and bacterial of culture (Table 3) the control sample had the lowest level of TVFA, contrary G treatment which had the

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highest level. These results might be due to the goat's milk, contains rather higher ratio of volatile fatty acids namely caproic, caprylic and capric acid. This is clearly evident control sample (made of cow's milk) containing the lower level of TVFA. Noteworthy observation, that with decrease the percentage of fat, TVFA clearly decreased *S. thermophilus* and *L. delbrueckii subsp. bulgaricus*, produced higher amount of TVFA than [*S. thermophilus* TA061, MR-1C, *L. delbrueckii subsp. bulgaricus* MR-1R and (*B. bifidum* & *L. acidophilus* 1:1)]. This may be due to the ability of traditional bacteria to hydrolyze of protein more than the other bacteria used. On the other hand, TVFA in all control and treatments gradually increased along the storage period to range between 0.52 and 0.1.4 ml 0.1 NaOH/10 g at the end of the storage period (21 days).

### **Microbial properties:**

Experimental Labneh samples were microbiologically analyzed during storage; confirmed that counts of total viable count, *B. bifidum*, *L. acidophilus* and lactic acid bacteria increased up to the fourteenth day then decreased (Table 4). CG3, CG2 and CG1 treatments exhibited the highest total viable count unlike C that recorded the lowest total viable count. This was the opposite of what was expected, these may be due to the type of milk used, goat's milk is rich of elements and compounds more than cows' milk, bacteria need that elements and compounds to grow well. After 14 day, CG1 treatment had the highest lactic acid bacteria counts ( $168 \text{ cfu} \times 10^6/\text{g}$ ) followed by CG3, CG2, G then C, meaning that the presence of polysaccharide producing strains encouraged clearly lactic bacteria growth. In connection with *B. bifidum* and *L. acidophilus* counts, they were similar in all treatments. There were no significant differences.

It is necessary to detect the presence of psychrotrophic bacteria that may release heat-resistant proteases and lipases, these enzymes will not be totally inactivated and may give rise to off-flavours (Tamime, 2009), Sahan *et al.*, (2004) reported that the number of psychrotrophic bacteria significantly decreased after 60 days in the samples stored at refrigeration temperature and to a smaller degree decreased in the samples stored at room temperature. Through Table 4 shows that Psychrotrophic bacteria did not detect until 21 day of cold storage. C had the highest count of Psychrotrophic ( $12 \text{ cfu} \times 10^2/\text{g}$ ) but CG1 contained the lowest number ( $6 \text{ cfu} \times 10^2/\text{g}$ ).

With regard to molds and yeasts, it is clear from Table 4 that molds and yeasts did not appear up to 7 d and appear after 14 d then slightly increased to reach between 8-11  $\text{cfu} \times 10^2/\text{g}$  at the end of cold storage period. C had the highest count of molds and yeasts ( $11 \text{ cfu} \times 10^2/\text{g}$ ). Our results agree with those of Al-Kadamany *et al.*, (2002) who stated that psychrotrophic yeasts increased in stored Labneh at 5 and 15°C. Results of this work are important to avoid chemical preservatives, Mihyar *et al.*, (1997) confirmed that more than 400 mg  $\text{kg}^{-1}$  of sodium benzoate were needed to limit the counts of *S. cerevisiae*, *Cryptococcus curvatus*, *Pichia farinosa*, *Candida blankii*, *Debaryomyces hansenii* and *Trichosporon brassicae* to  $10^5 \text{ cfu/g}$  after 14 days at 5°C; 150 and 300 mg  $\text{kg}^{-1}$  were needed for *Geotrichum candidum* and *Trichosporon cutaneum*, respectively. However, 100 - > 400 mg of potassium sorbate is needed to inhibit the same yeast in Labneh.

Both of *Staph. aureus* and coliform group were not detected in Labneh samples (data not shown), this might be due to the efficient heat treatment of milk (95°C for 15 min) and high sanitation conditions during manufacture and storage, the obtained results agree with those of Ammara (2000).

**Table 4: Microbiological properties of Labneh manufactured from mixtures of cow and goat's milk with EPS lactic acid bacteria (means ± SD)**

Properties	Treatments	Storage period (day)				Means ± SD
		Fresh	7	14	21	
Total viable count (cfux10 <sup>6</sup> /g)	C	159 <sup>d</sup>	175 <sup>a</sup>	182 <sup>a</sup>	155 <sup>d</sup>	167±11.94 <sup>C</sup>
	G	165 <sup>d</sup>	181 <sup>a</sup>	187 <sup>a</sup>	159 <sup>d</sup>	173±12.23 <sup>B</sup>
	CG3	171 <sup>d</sup>	183 <sup>a</sup>	189 <sup>a</sup>	162 <sup>e</sup>	176±11.25 <sup>A</sup>
	CG2	170 <sup>d</sup>	182 <sup>b</sup>	188 <sup>a</sup>	162 <sup>f</sup>	175±10.89 <sup>A</sup>
	CG1	171 <sup>d</sup>	182 <sup>a</sup>	187 <sup>a</sup>	160 <sup>f</sup>	175±11.19 <sup>A</sup>
<i>B. bifidum</i> (cfux10 <sup>6</sup> /g)	C	-	-	-	-	-
	G	-	-	-	-	-
	CG3	22 <sup>d</sup>	26 <sup>a</sup>	27 <sup>a</sup>	21 <sup>d</sup>	24±2.93 <sup>A</sup>
	CG2	23 <sup>d</sup>	25 <sup>b</sup>	27 <sup>a</sup>	21 <sup>f</sup>	24±2.62 <sup>A</sup>
	CG1	22 <sup>d</sup>	25 <sup>a</sup>	25 <sup>a</sup>	22 <sup>d</sup>	23±1.93 <sup>A</sup>
<i>L. acidophilus</i> (cfux10 <sup>6</sup> /g)	C	-	-	-	-	-
	G	-	-	-	-	-
	CG3	24 <sup>d</sup>	27 <sup>a</sup>	28 <sup>a</sup>	23 <sup>d</sup>	25±2.44 <sup>A</sup>
	CG2	25 <sup>c</sup>	28 <sup>a</sup>	28 <sup>a</sup>	22 <sup>d</sup>	25±2.87 <sup>A</sup>
	CG1	25 <sup>b</sup>	26 <sup>a</sup>	26 <sup>a</sup>	23 <sup>d</sup>	25±1.69 <sup>A</sup>
Lactic acid bacteria (cfux10 <sup>6</sup> /g)	C	135 <sup>f</sup>	157 <sup>a</sup>	161 <sup>a</sup>	149 <sup>d</sup>	150±10.68 <sup>C</sup>
	G	141 <sup>e</sup>	163 <sup>a</sup>	164 <sup>a</sup>	153 <sup>c</sup>	155±10.24 <sup>B</sup>
	CG3	143 <sup>e</sup>	165 <sup>a</sup>	166 <sup>a</sup>	154 <sup>c</sup>	157±10.06 <sup>AB</sup>
	CG2	143 <sup>e</sup>	165 <sup>a</sup>	165 <sup>a</sup>	154 <sup>c</sup>	156±9.80 <sup>AB</sup>
	CG1	144 <sup>e</sup>	167 <sup>a</sup>	168 <sup>a</sup>	154 <sup>c</sup>	157±10.15 <sup>A</sup>
Mold & Yeast (cfux10 <sup>2</sup> /g)	C	ND	ND	7 <sup>c</sup>	11 <sup>a</sup>	4.50±5.10 <sup>A</sup>
	G	ND	ND	6 <sup>d</sup>	9 <sup>a</sup>	3.75±4.23 <sup>AB</sup>
	CG3	ND	ND	5 <sup>e</sup>	8 <sup>b</sup>	3.25±3.73 <sup>B</sup>
	CG2	ND	ND	6 <sup>d</sup>	8 <sup>b</sup>	3.50±3.89 <sup>AB</sup>
	CG1	ND	ND	5 <sup>e</sup>	9 <sup>a</sup>	3.50±4.10 <sup>AB</sup>
Psychrotrophic bacteria (cfux10 <sup>2</sup> /g)	C	ND	ND	ND	12 <sup>a</sup>	3.00±5.58 <sup>A</sup>
	G	ND	ND	ND	9 <sup>a</sup>	2.37±4.47 <sup>AB</sup>
	CG3	ND	ND	ND	7 <sup>a</sup>	1.75±3.28 <sup>B</sup>
	CG2	ND	ND	ND	7 <sup>a</sup>	1.75±3.28 <sup>B</sup>
	CG1	ND	ND	ND	6 <sup>a</sup>	1.50±2.83 <sup>B</sup>

<sup>abcde</sup> Letters indicate significant differences between Labneh treatments, <sup>ABCDE</sup> Letters indicate the means of significant differences between Labneh treatments ND: not detected, -: the culture starter is free of this strain. C, G, CG3, CG2 and CG1: see table1

Aerobic spore forming bacteria were not detected in all of examined treatments whether fresh or after storage.

**Organoleptic properties:**

Data listed in Table 5. indicated that CG1 gained the highest scoring points followed

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by CG2, CG3, C then G whether in fresh or after 7, 14 and 21 days of the cold storage. These results explain the effect of bacterial strains producing EPS on properties of Labneh. EPS-producing (EPS<sup>+</sup>) LAB are commonly used as starter cultures for yogurt manufacture because EPS improves the viscosity and texture of yogurt and decreases its susceptibility to syneresis (loss of whey from the curd) (Kang and Cottrell, 1979 and Van den Berg *et al.*, 1995). Because EPS have viscosity enhancing and stabilizing properties, exopolysaccharide-

producing (EPS) starter cultures are commonly used to enhance water binding and viscosity in yogurt and fermented milks as well as thickness. Low fat Mozzarella cheese made with an EPS starter pair, *Streptococcus thermophilus* MR-1C and *Lactobacillus delbrueckii* subsp. *bulgaricus* MR-1R, contained significantly more moisture and had better melt properties than cheese made with a control starter pair (Broadbent *et al.* 2011). These results are in agreement with those reported by Abou ayana and Ibrahim (2015).

**Table 5: organoleptic evaluation of Labneh made from mixtures of cow and goat's milk with exopolysaccharide bacteria cultures (means ± SD)**

Properties	Treatments	Storage period (day)				Means ± SD
		Fresh	7	14	21	
Flavor (60)	C	55 <sup>a</sup>	55 <sup>a</sup>	54 <sup>b</sup>	53 <sup>c</sup>	54.50±1.60 <sup>A</sup>
	G	53 <sup>a</sup>	51 <sup>b</sup>	50 <sup>c</sup>	48 <sup>d</sup>	50.50±2.20 <sup>C</sup>
	CG3	56 <sup>a</sup>	54 <sup>b</sup>	52 <sup>d</sup>	50 <sup>e</sup>	53.00±2.62 <sup>B</sup>
	CG2	58 <sup>a</sup>	56 <sup>a</sup>	54 <sup>c</sup>	52 <sup>d</sup>	55.00±2.62 <sup>A</sup>
	CG1	58 <sup>a</sup>	57 <sup>a</sup>	55 <sup>c</sup>	54 <sup>c</sup>	56.00±2.00 <sup>A</sup>
Body & Texture (30)	C	26 <sup>a</sup>	24 <sup>c</sup>	21 <sup>e</sup>	20 <sup>e</sup>	22.75±2.76 <sup>B</sup>
	G	26 <sup>a</sup>	24 <sup>c</sup>	23 <sup>d</sup>	22 <sup>e</sup>	23.75±1.91 <sup>B</sup>
	CG3	27 <sup>a</sup>	27 <sup>a</sup>	25 <sup>c</sup>	25 <sup>c</sup>	26.00±1.51 <sup>A</sup>
	CG2	27 <sup>a</sup>	26 <sup>a</sup>	24 <sup>c</sup>	24 <sup>c</sup>	25.25±1.75 <sup>A</sup>
	CG1	28 <sup>a</sup>	26 <sup>c</sup>	25 <sup>d</sup>	25 <sup>d</sup>	26.00±1.69 <sup>A</sup>
Appearance (10)	C	8 <sup>a</sup>	8 <sup>a</sup>	7 <sup>b</sup>	6 <sup>c</sup>	7.25±1.39 <sup>A</sup>
	G	8 <sup>a</sup>	8 <sup>a</sup>	7 <sup>b</sup>	6 <sup>c</sup>	7.25±1.39 <sup>A</sup>
	CG3	8 <sup>a</sup>	8 <sup>a</sup>	7 <sup>b</sup>	7 <sup>b</sup>	7.50±1.19 <sup>A</sup>
	CG2	9 <sup>a</sup>	9 <sup>a</sup>	8 <sup>b</sup>	8 <sup>b</sup>	8.50±1.19 <sup>A</sup>
	CG1	9 <sup>a</sup>	9 <sup>a</sup>	8 <sup>b</sup>	8 <sup>b</sup>	8.50±1.19 <sup>A</sup>
Total (100)	C	90 <sup>a</sup>	88 <sup>a</sup>	82 <sup>c</sup>	79 <sup>d</sup>	84.75±2.23 <sup>C</sup>
	G	87 <sup>a</sup>	84 <sup>a</sup>	80 <sup>c</sup>	76 <sup>d</sup>	81.75±1.99 <sup>D</sup>
	CG3	91 <sup>a</sup>	89 <sup>a</sup>	84 <sup>c</sup>	82 <sup>e</sup>	86.50±2.11 <sup>B</sup>
	CG2	94 <sup>a</sup>	91 <sup>a</sup>	86 <sup>c</sup>	84 <sup>d</sup>	88.75±2.34 <sup>A</sup>
	CG1	94 <sup>a</sup>	91 <sup>a</sup>	87 <sup>c</sup>	86 <sup>c</sup>	89.50±2.25 <sup>A</sup>

<sup>abcde</sup> Letters indicate significant differences between Labneh treatments, <sup>ABCDE</sup> Letters indicate the means of significant differences between Labneh treatments. C, G, CG3, CG2 and CG1: see table1

On the other hand, an negative correlation between the percentage of goats' milk fat and sensory evaluation of flavor of Labneh was observed. Although goats' milk fat improved the body, texture and appearance of Labneh but the flavor of G3 samples had lowest points 53 out of 60 in fresh and decreased to reach 48 after 21 days of storage and was not accepted and rejected by most of the panelists. This could be attributed to free fatty acids especially Caproic, caprylic and capric (totaling of 15% of goat milk fat). These findings agree with those reported by Abou Ayana and Gamal El Deen (2011)

## **CONCLUSIONS**

Using bacterial strains producing EPS is recommended to enhance flavor and rheological properties of Labneh made from mixtures of cow's and goat's milk as well as increase of yield.

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تقييم اللبن المصنعة من خليط من لبن الماعز واللبن البقري باستخدام البكتريا  
الداعمة للحوية والبكتريا المنتجة للسكريات العديدة

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**الملخص العربي :**

السكريات الميكروبية العديدة هي مثبتات حيوية يمكن ان تُضاف إلى العديد من المنتجات الغذائية، حيث تزيد لزوجة واستحلاب وثبات المنتج كما أنها بمثابة عوامل للحالة الجيلية المرغوبة. والعديد من هذه السكريات العديدة المنتجة خارج الخلية لها تركيب كيميائي وأحجام جزيئية مختلفة، وهي تُنتج بواسطة بعض سلالات بكتريا حامض اللاكتيك.

الهدف من هذا البحث هو تقييم اللبن المصنعه من خليط من لبن الماعز واللبن البقري باستخدام بكتريا البروبيوتك و بكتريا حامض اللاكتيك المنتجة للسكريات العديدة. أشارت النتائج الى : أن استخدام السلالات [ *S. thermophilus* TA061, MR-1C + *L. delbrueckii subsp. bulgaricus* MR-1R+ (*B. bifidum* and *L. acidophilus*) ] زاد من تصافي اللبن و أيضا من محتواها من المواد الصلبة الكلية و الدهن/ المادة الجافة (F/DM). كما أن اللبن المصنعة من خليط من لبن الماعز واللبن البقري والمحتوى على 1% دهن باستخدام البادئ [ *S. thermophilus* TA061, MR-1C + *L. delbrueckii subsp. bulgaricus* MR-] (*B. bifidum* and *L. acidophilus* 1:1) حصلت على أعلى تقييم حسي على عكس اللبن المصنعة من لبن الماعز 3% دهن و المصنعة باستخدام البادئ التقليدي.

***Gabr***

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