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MANUFACTURE OF BIO LABNEH

Salem, M.M.E.
Dairy Science Dept., National Research Center, Dokki, Cairo, Egypt.

ABSTRACT

Labneh was manufactured from a mixture of buffaloes and soymilks by using different types of probiotic starter cultures. The resultant Labneh cheese were analyzed when fresh and periodically during storage period (4 weeks) for chemical, microbiological and organoleptic properties. The counts of probiotic bacteria in labneh containing soymilk were higher than those without soymilk, It recorded at zero time 8.5,8.7,8.6 and8.7 log10 cfu/g *for Lactobacillus acidophilus, Lactobacillus reuteri, Lactobacillus rhamnosus* and *Bifidobacterium .bifidum* respectively. While the counts of probiotic strains reached to 7.1 and 6.9 log 10 cfu/g for *L.reuteri* and *B.bifidum* a fter four weeks of refrigerator storage .The moisture content of all Labneh cheese samples were gradually decreased till the end of storage period . No marked differences could be observed among the different treatments. The pH values gave the same trend reaching the minimal values at the end of period . The Labneh made with *L.reuteri* had the highest organoleptic scores when fresh and throughout storage followed by that made with *B.bifidum* .Finally, addition of soymilk in manufactured of labneh improved the viability of probiotic strains.
Keywords: Labneh, probiotic, bacteria, soymilk.

INTRODUCTION

Labneh is a concentrated yoghurt, which organized in the Middle East area. It usually made by a curd coagulation of milk incubated with lactic acid bacteria and drainage of the curd in cloth bags (Hamad and Al sheikh, 1989). It should be soft, smooth; spread able with a consistency resembles cultured cream, acidic clean flavour and milky white colour. Also, it should not be dry or grainy and there should be no wheying off. A mixture of thermophilic strains of lactic acid bacteria, i.e. *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus salivarius* subsp.*thermophilus* is used to produce labneh (El-Samragy, 1997)

In recent years, the health benefits of soybean-based product have been widely recognized all over the world. Epidemiological studies and clinical trials have revealed that isoflavons and oligosaccharides in soybean are effective for the premeditation of various chronic diseases and harmone – associated health disorders (Setchel and Gassidy, 1999). Dominant oligosaccharides of soybean are sucrose, raffinose and stachuose, the latter two are x-galactosyl oligosaccharides, and called prebiotics which are defined as nondigestible food components that give beneficial effect to the health by selectively activating probiotics, beneficial enterobacteria such as bifdobacteria (Bordignon *et al..*, 2004).

Substitution of fresh milk by soymilk in dairy products has some advantages from commercial and nutritional points of view especially in the case of insufficient fresh milk supply. The health benefits of soybean products such as soymilk, tofu, soybean cheese and miso are traditional foods in Asia, they are rich in protein, unsaturated fatty acids, lecithin and isofavones, contain no cholesterol or lactose and may be consumed by those suffering from lactose intolerance (Fu-Mei Lin and Pan., 2004). Soymilk is also a

suitable medium for growing lactic acid bacteria, thus improving the quality of fermented products. Similar to yoghurt, fermented soymilk is called soyghurt (Mital *et al..*, 1975) and soymilk have been recently used to partially replace milk in the production of yoghurt-like products (Macedo *et al..*, 1999 and Omogbai *et al..*, 2005).

The aim of this study was to produce acceptable functional dairy product, effect of soymilk addition on the growth of probiotic bacteria and quality of resultant Labneh during storage period.

MATERIAL AND METHODS

Starter

Lactobacillus reuteri B-14171 and *Lactobacillus rhamnosus* B445, were provided by Northern Regional Research Laboratory, Illinois, USA (NRRL). *Lactobacillus acidophilus* and *Bifidobacterium bifidum* were obtained from Chr. Hansen's Lab., Denmark. All strains mentioned above had properties required of a probiotic microorganisms including bile salt tolerance, tolerance to low-pH values and antagonistic activity as reported by (Amin *et al.,* 2002). In addition, *Streptococcus thermophilus* and *Lactobacillus delbrueckii ssp. bulgaricus* were obtained from Dairy Microbiology Lab., National Research Centre, Dokki, Cairo, Egypt.

Soymilk preparation

Preparation of soymilk was performed according to the procedures described by Omogbai *et al..* (2005). Briefly, whole soybeans were first washed and soaked overnight in distilled water. The soaked soybeans, after decanting the water, were blanched at 98ºC in boiling distilled water for 20 min to remove their taste. They were then placed in a warring blender and boiled distilled water at 87-90 ^{0}C was added and then blended for 3 min. The boiled water inactivated the enzyme, lipoxygenase during blending (Wilkens *et al.*, 1967) . The resultant slurry was then filtered through double- layered cheesecloth to yield soymilk. Flasks containing soymilk were capped with cotton plugs, covered with aluminum foil and autoclaved at121 ^{0}C for 15 min and thereafter held at 5 ºC until used.

Preliminary experiments

Preliminary experiments were carried out to explore the ample conditions for preparation of acceptable product. These experiments included selection of suitable mixing ratio between buffaloes whole milk and soymilk. The buffalo's milks were mixed with 10,20,30,40and50% of sterilized soymilk. From these experiments good appearance, texture and quality product was obtained by using 30% of soymilk.

Manufacture of labneh:

Labneh was manufactured in triplicate according to El-Samargy (1997} from a heated mixture of buffalo's milk and soymilk (70:30) for 90ºC/10 min. Buffaloes-soymilk was immediately cooled to 45 ^{0}C and divided into five equal portions. Starters were added as

1-*S.thermophilus* and *L.delbrueckii ssp. bulgaricus,served as a control*
2- S.thermophilus and *L.delbrueckii ssp. bulgaricus and L.acidophilus*
3- S.thermophilus and *L.delbrueckii ssp. bulgaricus and L.rueteri*

4- S.thermophilus and *L.delbrueckii ssp. bulgaricus and L.rhamnosus*
5- S.thermophilus and *L.delbrueckii ssp. bulgaricus and B.bifidum*
all starters were added at the level of 3% and incubated at 42 ºC for 3h. When it

curded, yoghurt was cooled, mixed thoroughly and put into cheese clothe bags and then hung in the refrigerator overnight to allow whey drained .The resultant Labenh cheese was mixed with 1%(w/w) sodium chloride. The fresh Labneh cheese was filled into 250g. plastic containers and stored at refrigerator (5ºC+1) for 4 weeks. In each trial samples were analyzed when fresh and 1, 2, 3 and 4 weeks to follow chemical, microbiological and organoleptic properties.

Chemical and microbiological analysis

Labneh samples were drawn for chemical analysis when fresh and after 1,2,3and 4 weeks of storage to determine moisture and pH according to Kosikowski (1978) and examined microbiologically for total viable count and coliform bacteria (APHA, 1978), lactic acid bacteria (Elliker *et al.* 1956) proteolytic bacteria and lipolytic bacteria (Hammer and Bable, 1957), and yeasts and moulds (Harrigan and McCance, 1966). Enumeration of *B.bifidum* was done according to (Blanchette *et al.*, 1996) using modified MRS agar (Oxoid) supplemented with 0.05% L-cysteine Hcl (Merek Germany). Plates were incubated at 37ºC for 48 h. in anaerobic environment (BBL Gas Pak, Bacton Dickinson, Co. Keysville AM, USA). Viability of *Lreuteri and L.rhamnosus* was determined on MRS arabinose agar following 2-days incubation at 37ºC (Effat, 2000) and on LC agar following 3-4 days incubation at 27ºC (Ravula and Shah, 1998) under anaerobic conditions, respectively.

Organoleptic evaluation.

All labneh samples were sensory evaluated according to Amer *et al.*, (1997). when fresh and after 1,2,3and 4 weeks by panelists from staff members of Dairy Department at the National Research Centre

RESULTS AND DISSCUTION

Chemical properties

It is clear from the results presented in Fig.1 that Labneh made by addition of soymilk and probiotic bacteria had higher moisture content . Such effect may be due to the capability of soymilk to decrease the syneresis rate of whey during manufacture. The moisture content of all labneh samples were gradually decreased till the end of the storage period. No marked differences could be observed among the different treatments. On the other hand the pH values (Fig.2) gradually decreased in all treatments either when fresh or during storage period reaching their minimal values at the end of period. This results are in agreement with Amer *et al.*, 1997 and El- syed *et al.*, 1993.

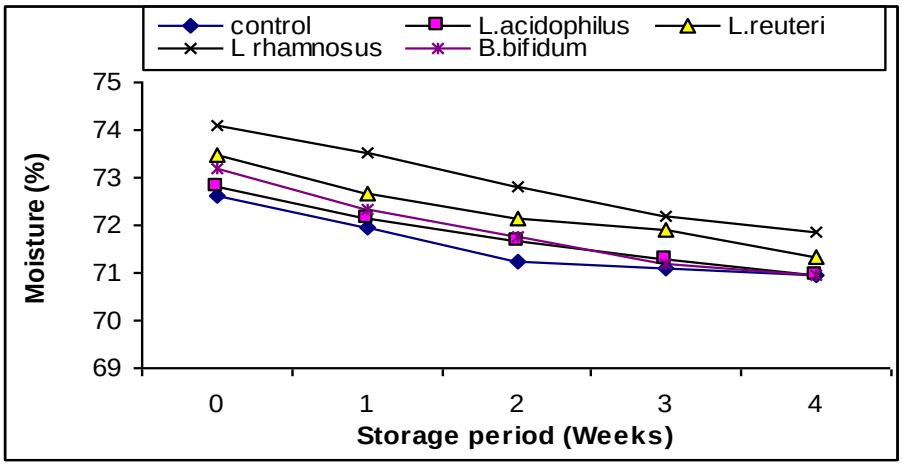

Fig (1). Changes in the moisture content of Labneh made with probiotic bacteria and soy milk during storage period.

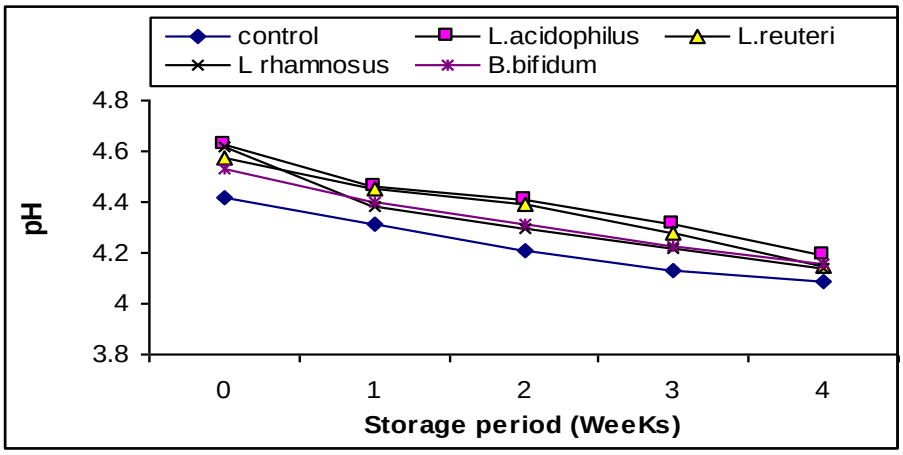

Fig (2): Changes in the pH values of Labneh with probiotic bacteria and soy milk during storage period

Microbiological properties

The viability of *L.acidophilus, L.reuteri,L.rhamnosus* and *B.bifidum* in labneh supplemented with and without soymilk stored at refrigerator 5-7ºC for 4 weeks are shown in figures (3a and 3b). The viability of all strains in labneh decreased during storage. However the counts of probiotic strains in labneh containing soymilk were higher than those without soymilk when fresh and throughout the storage period at 5-7ºC. At the zero time (Fig.3a), the highest counts of 8.5,8.7,8.6 and 8.7 log10 cfu/g was recorded for *L.acidophilus, L.reuteri, L.rhamnosus* and *B.bifidum* in labneh with soymilk respectively. That it is may be due to the suitability of soymilk as medium for growing lactic acid bacteria (Fu-Mei Lin and Ban, 2004). On the other hand the counts of

probiotic strains was increased gradually during the first two weeks of storage period at 5-7 ºC and then decreased till end of this period . This decrease may attributed to the sensitively of microorganisms to acid. Our results confirm the observation of Tawfik,1993; Robinson,1990; Gilliland and Speeck, 1977 and Sharaf *et al.*, 1996. Although viability of probiotic bacteria decreased during storage, it was observed that log of counts for the labneh supplemented with soymilk that 7.1 and 6.9 $\log_1$ cfu/g for *L.reuteri* and *B.bifiduhm*, in average, after four weeks of storage. These counts are close to 10^7 cfu /g, which is the level of suggested by some authors to have health promoting effect (Oliveira *et al.*, 2002 and Akin 2005). In gereral, our results have shown that probiotic strains persist in the soymilk product at moderately high numbers even after four weeks of storage. The viability of probiotic tested strains obtained in soymilk might be attributed to the presence of certain protective factors (glutamic acid, peptides and salts) present in the substrate (Garro *et al..*, 1999).

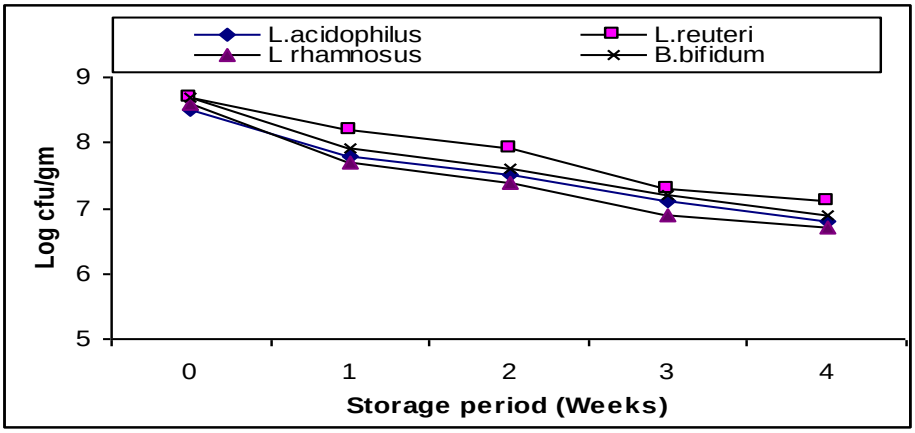

Fig (3 a): Viability of probiotic bacteria count of Labneh made with soy milk during storage period.

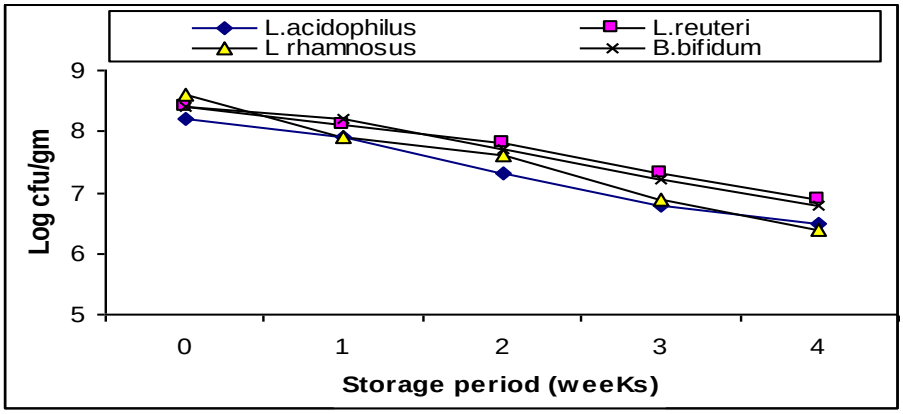

Fig (3 b): Viability of probiotic bacteria count of Labneh without soy milk during storage period.

Data given in Fig.(4) showed that the labneh containing soymilk and probiotic bacteria had similar total viable counts in all treatments ,when either fresh or during storage period and the total viable counts gradually decreased towards the end of storage period . These results agree with Ibrahim *et al..*, 1994. While Fig.(5) indicated that the lactic acid bacteria counts during storage period in all treatments exhibited similar trends. They reached their maximum counts after one week then decreased towards the end of storage period. Reduced viability of LAB could be due to the presence of lactic acid and hydrogen peroxide which inhibit the growth (El-sayd *et al.*,1993 and Gilliland &speck 1997).

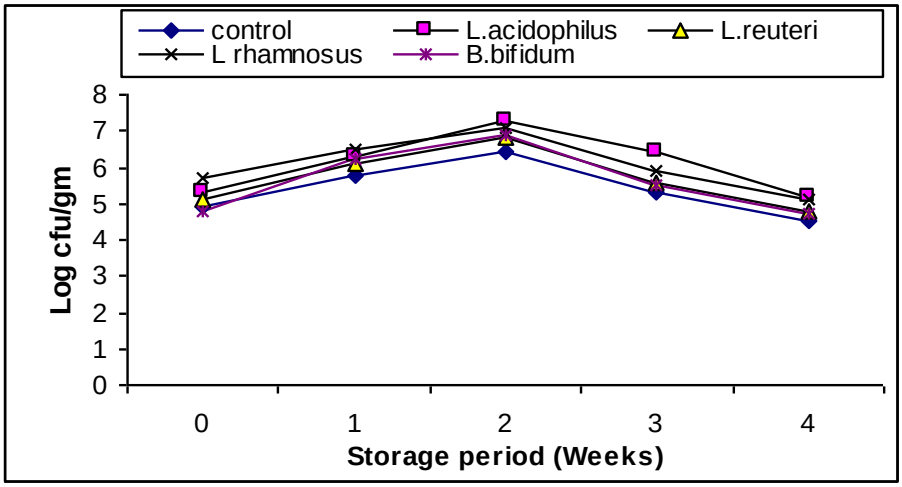

Fig (4). Changes in the total viable count of Labneh with probiotic bacteria and soy milk during storage period

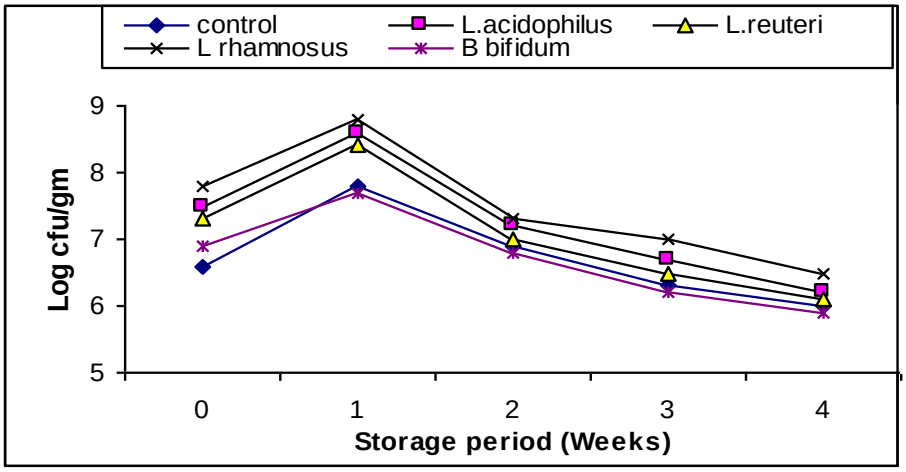

Fig (5). Changes in the lactic acid bacteria count of Labneh with probiotic bacteria and soy milk during storage period.

Lipolytic and proteolytic bacteria were gradually increased in all treatments with the progression of storage period (Fig.6 &Fig.7). They reached the maximum counts at the end of storage period. The forgoing results are consistent with previous findings with Ibrahim *et al.* (1994). On the other hand data in Fig (8) reveals that yeasts and molds counts were found to be low in fresh products it reached to 1.1 and 1.0 log cfu/g for Labneh cheese containing soymilk with *L.acidophilus* and *B. bifidum* respectively. These results are partly in agreement with that reported by Amer ,*et al.* (1997) who found that the production of some inhibitors as a result of the presence *L.acidophilus* and *B.bifidum*. Generally the yeasts and moulds count were low in fresh products in all treatments then increased gradually throughout storage period to record maximum values at the end of storage period . All examined labneh samples were free from coliforms and such results reflected the adequate hygienic conditions adapted during the manufacture of Labneh-soymilk product.

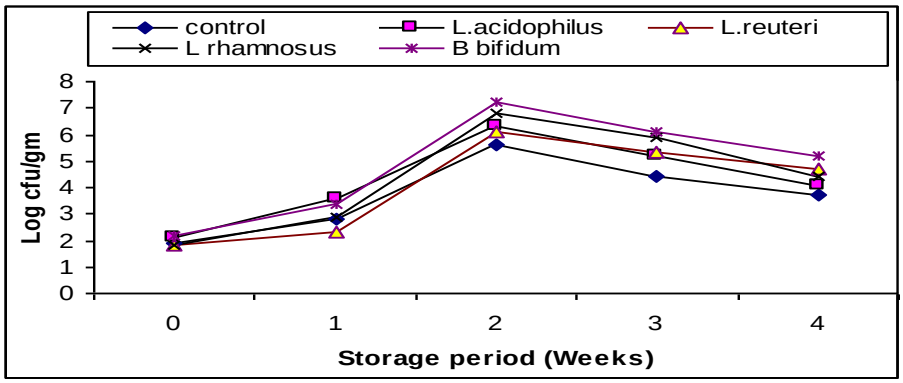

Fig (6). Changes in the proteolytic bacteria count of Labneh with probiotic bacteria and soy milk during storage period.

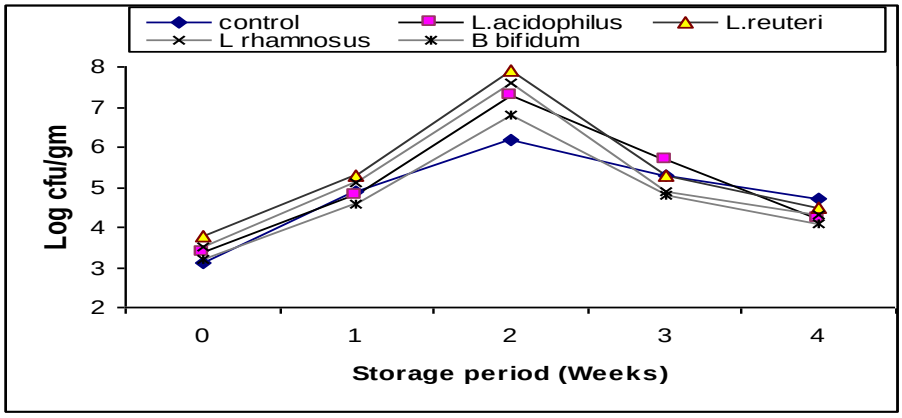

Fig (7). Changes in the lipolytic bacteria count of Labneh with probiotic bacteria and soy milk during storage period.

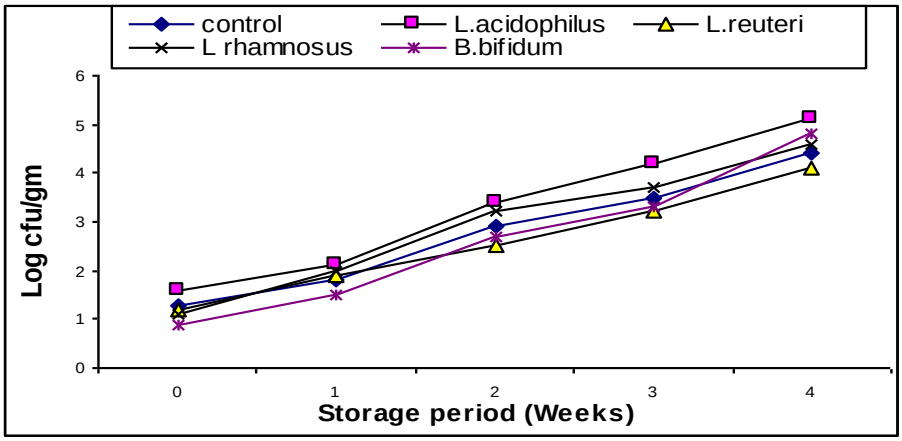

Fig (8). Changes in the molds and yeasts count of Labneh with probiotic bacteria and soy milk during storage period

Organolyptic properties

Data presented in Fig.(9) summarized the sensory evaluation of labneh as affected by storage. In fresh labneh with *L.reuteri* had the highest score for acceptability followed by that made with *B.bifidum* either when fresh or during storage. These products characterized by pleasant taste, good flavour, good appearance and favourable acidity. This could be due to that the main characteristics of *S.thermophilus* are its abilities to produce aroma (Mohamed *et al.*, (1999) Moreover, the presence of *L.reuteri* till the end of storage period which is acid tolerant (Toit *et al.*, 1998). On the other hand , labneh made with yoghurt culture and *L.rhamnosus* obtained the lowest scores. As storage progressed the organoleptic scores decreased in all treatments. This results are in agreement with Taha *et al.*, 1997 and El-Sayed *et al.*, 1993.

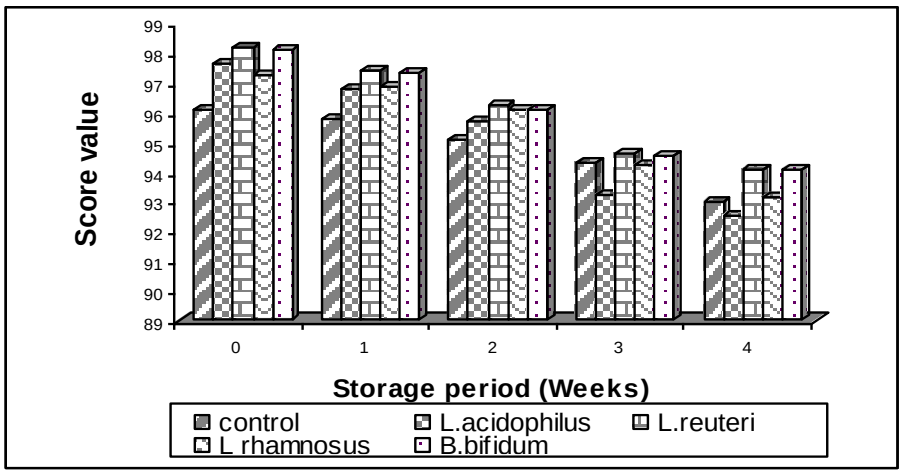

Fig (8). Sensory evaluation of Labneh with probiotic bacteria and soy milk during storage period

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تصنيع لبنة حيوية

موسي معالي عيد سالم

قسم الألبان – المركز القومي للبحوث

تم تصنيع لبنة باضافة لبن فول الصويا وذلك باستخدام ٤ سلالات من بكتريا المدعمات الحيوية، ثم تحليل المنتج كيماويا وميكربيولوجيا وحسيا خلال فترة التخزين.

وقد أوضحت النتائج أن أعداد بكتريا المدعمات الحيوية كانت في اللبنة المصنعة بإضافة لبن فول الوصيا حيث كان الأس اللوغاريتمي ٨,٥، ٨,٧، ٨,٦، ٨,٧ خلية /جم لكلا من *L.acidophilus, L.reuteri, L.rhamnosus* and *B.bifidum* علي التوالي. بينما كان الأس أللوغاريتمي ٧,٧ ، ٦,٩ خلية /جم لكلا من*L.reuteri* and *B.bifidum* بعد ٤ أسابيع من التخزين المبرد. هذا وقد لوحظ تناقص تدريجي في المحتوي من الرطوبة خلال فترة التخزين . وقد حصلت اللبنة المصنعة من لبن فول الصويا في وجود *L.reuteri,* كمدعم حيوي على أعلى درجات التحكيم الحسي ن يليها تلك المصنعة بإضافة *B.bifidum*

ومما سبق يتضح ان إضافة لبن فول ا لصويا إلي للبنة المصنعة باستخدام بكتريا المدعمات الحيوية قد أدي إلي زيادة حيويتها.