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Quality of Probiotic Gouda Cheese as a Functional Food

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GOUDA cheese was mad from cow's milk inoculated with cheese starter culture (CSC) without (the control C) and with partial replacement of cheese milk with buffalo's milk (25%). The mixed milk with CSC was applied in T1, while *Lactobacillu shelveticus* CH5 and *Lactobacillus plantarum* ATCC 14917 were used separately with CSC in T2 and T3, respectively.Microbiological analysis of fresh cheese and during ripening revealed that Gouda cheese treated with *Lactobacillus helveticus* (T2), followed by cheese treated with *Lactobacillus plantarum* (T3) had the highest values in total bacterial count and count of proteolytic andlipolytic bacteria when fresh and during ripening period, while no colonies of yeasts & moulds appeared in all cheese treatments when fresh, but after one month of ripening few colonies were observed. *Lactobacillus helveticus* CH5 significantly increased during ripening and reached 9.2 log cfu. g^{-1} at day 60, while the number of the *Lactobacillus plantarum* ATCC14917 bacteria at the beginning of the maturation period was about 8.2 log cfu g^{-1} then increased at 60 days of ripening (8.8 log cfu g^{-1}). The counts of the prementioned bacteria at the end of ripening were more than 7 log cfu/g.Organoleptically, appearance and colour of control cheese of any age had the maximum scoring points. The control cheese and cheese from T3 ranked the maximum scores for body, texture and flavour and the differences between them were statistically significant. This was noticed in fresh cheese and during ripening period.

Keywords: Gouda Cheese, Probiotic bacteria, *Lactobacillus helveticus*, *Lactobacillus plantarum*, Buffalo's milk, Cheese quality.

Introduction

It is well-known that the concept of functional foods (FF) was first originated in Japan in 1984 while FF has no universally accepted definition (Hasler, 2002). The new term used for FF was the food which possesses naturally occurring or enriched with natural substances with a specific health promoting effect beyond. It is basic nutritive value (Vukasovic, 2017). The dairy industry is in an excellent position to develop and exploit the FF market (Aadinath et al. 2017).

Probiotics as source for FF were given in details by Heller (2001), Granato et al. (2010)and Tripathi and Giri (2014) since there is no doubt that dairy products are the main vehicle for probiotic supplementation. Indeed, consumption of probiotic dairy products including yoghurt and cheese to promote health benefits were comprehensively reported in the literature (Granato, et al. 2010, Boylston et al. 2004; Ong et al., 2007; Plessaset al. 2012).

Although yoghurt and fermented milks have received the most attention as carriers of probiotic bacteria, some cheese varieties such as Gouda, white and Cheddar cheeses (Gomes et al. 1995) have a number of advantages over fermented milks as a delivery system for viable probiotic microorganisms, because they generally have higher pH and buffering capacity, more solid consistency, and relatively higher fat content (Joutsjoki, 2009). These features give protection to probiotic bacteria during storage and passage through the gastrointestinal tract.

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Several *Lb. plantarum* isolates proved the ability to survive gastric transit and to colonize the intestinal tract of humans and other mammals (Georgieva et al. 2008). Also, Zago et al. (2011) reported that *Lb. plantarum* strains from cheeses displayed good resistance to bile salts. List of probiotic strains used in commercial applications and the starter organisms for probiotic dairy products as well as their viability were reviewed by Heller (2001), Mohammadi and Mortazavian (2011) and Tripathi and Giri (2014).

The objective of the present study was to the use of *Lb. helveticus* CH5 and *Lb. plantarum* ATCC 14917 in making probiotic Gouda cheese. Survival and growth of the prementioned bacteria during cheese repining and the quality of the resultant cheese were taken into consideration.

Materials and Methods

- Fresh cow's and buffalo's milk were obtained from the herds of Faculty of Agriculture, Cairo University, Egypt.

- Commercial starter of mesophilic culture FD-DVS R-704 (consisting of *Lactococcus lactis subsp. lactis* and *Lactococcus lactis sub sp. cremoris)* was obtained from Chr. Hansen's Lab., Denmark and used as the control starter.

- Probiotic bacteria of *Lactobacillus helveticus* CH5 and *Lactobacillus plantarum* (ATCC14917) were obtained from the Egyptian Microbial Culture Collection (EMCC) aging to Cairo Microbial Resources Center (MIRCEN), Faculty of Agriculture, Ain Shams University, Egypt.

- Rennet powder and Annatto were obtained from Chr. Hansen's Lab., Denmark.

- Yellow wax coating material imported from Germany was obtained from AWA for food additives company, Alexandria, Egypt.

Manufacture of Gouda Cheese

It was carried out as described by Scott (1998). Standardized cow's milk (3.0% fat) was used for making the control cheese (C), whereas in the three treatments applied, cow's milk was partially replaced with buffalo's milk (6.0% fat) to give ratio of 3:1. All milk samples were pasteurized at 73ºC/20 sec., cooled to 32ºC, while 0.02% (w/w) $CaCl_2$ was added before carrying out the following treatments:

- Control cheese was made from cow's milk inoculated with commercial cheese mesophilic starter culture (CMSC).

- T1was made form mixture of cow's milk and buffalo's milk (3:1) with CMSC.

-T2 was made as given in T1 but with using CMSC + *Lb. helveticus* (1:1).

- T3 was made as given in T1 but with using CMSC+ *Lb. plantarum* (1:1).

All cheese samples were coated using yellow wax under certain conditions and kept for ripening at 10 – 12ºC and RH of 85-95 % for 3 months. The samples were analyzed when fresh and after 1,2 and 3 months of ripening period.Three replicates were done whereas all samples were analyzed twice.

Microbiological examination

Total bacterial counts (TBC), yeasts & moulds and coliforms were counted according to Marshall (1992). Proteolytic bacterial count was determined as described by Chalmer (1962). Lipolytic bacterial count was determined as given by Sharf (1970)**.**Counts of *Lb.helveticus* CH5 and *Lb.plantarum* ATCC14917 were determined using MRS-agar (Merck, Germany) at pH 5.4 according to IDF (1988).

Sensory evaluation

The organoleptic properties of cheese were evaluated by 10 judges of staff members of Food Technol. Res.Inst., Agric. Res. Cent.,Giza.

In the light of the information given by Nelson and Trout (1981) for hard cheese – in general and by El- Nimr et al. (2010) and El-Nagar et al. (2010) for Gouda cheese, all cheese samples were judged for general appearance and colour, body, texture and flavour while, the maximum attainable scoring point was 10 for each of the prementioned attributes except 20 points were given for flavour.

Statistical analysis

Statistical analysis of the obtained data was performed according to SAS Institute (1990) using liner Model (GLM). Duncan's multiple rang was used to separate among means of three replicates of the data.

Results and Discussion

Microbiological examination of Gouda cheese

The changes of total bacterial count (TBC) and counts of proteolytic (PBC) and lipolytic bacteria (LBC) of Gouda cheese manufactured using cow's and buffalo's milk and treated with cheese starter culture (CSC) either alone (control and T1) and in combination with *Lb. helveticus* CH5 (T2), or with *Lb. plantarum* ATCC14917 (T3) during the ripening period are shown in Table 1. The TBC was relatively higher in fresh cheese made from T2 and T3 than the counts in the control cheese (C) or cheese from T1. However, statistical analysis showed that the differences in this respect were insignificant. This was also noticed in cheese of 30 days old,but the differences in this case were significant ($p \leq 0.05$). The control cheese of 60 and 90 days old had the lowest counts compared to all treated samples. However, the TBC at the end of ripening period were 5.7, 6.03, 6.12 and 6.08 logcfu/g of C, T1, T2 and T3 respectively. TBC of all cheese samples significantly decreased after 60 days of ripening period. The decrease in TBC could be attributed to the decrease of water activity and the increase of salt content and acidity in cheese. These results are in agreement with those obtained by EL-Nagar et al. (2010) who found a gradual decrease in TBC during ripening of Gouda cheese**.**

As expected impact of adding *Lb. helveticus* as in T2 or *Lb. plantarum* as in T3 should be taken into consideration as a main factor responsible for increasing the TBC in cheese.This is quite important and reveals that the prementioned probiotic bacteria grew well in cheese during the ripening period. Table 1 shows also the PBC were not affected significantly by the applied treatments during the first 60 days of cheese ripening but at the end of ripening period the PBC were 5.19, 5.18, 5.83 and 5.47 logcfu/g suggesting the use of *Lb.helveticus* (T2) or *Lb. plantarum* (T3) significantly increased PBC of the ripened Gouda cheese.

TABLE 1. Total bacterial count (TBC) and counts of proteolytic bacteria (PBC) and lipolytic bacteria (LBC) as log cfu /g of Gouda cheese as affected by the applied treatments (Average ± SD of 3 replicates)*

Ripening period (days)	Treatments				LSD (0.05)
	C	T1	T2	T3	
	TBC				
Fresh	6.03 ± 0.055Ac	6.17 ± 0.305Ac	6.330.193 ± Ac	6.340.204 ±Ac	-------
30	6.41 ± 0.107Bb	6.80 ± 0.223Ab	6.93 ± 0.176Ab	6.82 ± 0.237Ab	0.3635
60	6.84 ± 0.112Ca	7.28 ± 0.207Ba	7.65 ± 0.232Aa	7.32 ± 0.204ABa	0.3667
90	5.70± 0.176Bd	6.03 ± 0.160ABc	6.12 ± 0.211Ac	6.08 ± 0.228Ac	0.3689
LSD	0.2271	0.4336	0.3843	0.4144	
	PBC				
Fresh	4.35 ± 0.325Ac	4.17 ± 0.304Ac	4.33 ± 0.221Ac	4.27 ± 0.216Ac	--------
30	4.62 ± 0.150Abc	4.67 ± 0.405Abc	4.71 ± 0.305Ac	4.74 ± 0.240Abc	--------
60	5.01 ± 0.201Aab	5.04 ± 0.146Aab	5.24 ± 0.242Ab	4.94 ± 0.317Ab	---------
90	5.19 ± 0.101Ba	5.18 ± 0.076Ba	5.83 ± 0.273Aa	5.47 ±0.311ABa	0.4082
LSD	0.3989	0.5019	0.4940	0.5179	
	LBC				
Fresh	4.23 ± 0.310Ac	4.100.328 ± Ab	4.200.337 ±Ab	4.300.242 ±Ac	--------
30	4.66 ± 0. 231Abc	4.60 ± 0.291Ab	4.48 ± 0.324Ab	4.62 ± 0.276Abc	--------
60	4.92 ± 0.236Aab	5.19 ± 0.235Aa	5.04 ± 0.201Aa	4.86 ± 0.289Aab	--------
90	5.35 ± 0.288Aa	5.39 ± 0.276Aa	5.45 ± 0.292Aa	5.54 ± 0.293Aa	--------
LSD	0.5059	0.5362	0.5532	0.5110	

C: control-100% Cow's milk + Cheese starter

T1: 75% Cow's + 25% Buffalo's milk + Cheese starter

T2: 75% Cow's + 25% Buffalo's milk + {Cheese starter + Lb. helveticus (1:1)}

T3: 75% Cow's + 25% Buffalo's milk + {Cheese starter + Lb. plantarum (1:1)}

A, B, C & D and a, b, c & d: means with the same letter among treatments and the storage period respectively are not significantly different ($p<0.05$).

The rate of proteolysis in cheese with probiotic bacteria (T2 and T3) was probably as a consequence of their different proteolytic activity. Proteolysis is the most complex and important biochemical event that occurs during cheese ripening period, and it plays a direct role on cheese texture and flavour development in most cheese varieties (Juan et al. 2007). However, a gradual increase in PBC was given by El- Nagar et al. (2010) during ripening of Gouda cheese made using Lb. helveticuswith commercial starter.

The differences in LBC at any ripening time of Gouda cheese due to the applied treatments were statistically insignificant. At the end of ripening period relatively higher LBC were recorded in T2 and T3 when compared with those of C or T1.The recorded LBC as given in Table (1) were 5.35, 5.39, 5.45 and 5.54 log cfu/g of C, T1, T2 and T3 samples respectively. The significant increase in LBC withadvancing ripening agrees with the finding of El -Nagar et al. (2010) who showed the same for Gouda cheese.

Variations noticed among the counts of all treatments were probably due to the primary environmental factors controlling growth of microorganisms in cheese include water and salt contents, pH value, presence of organic acids and ripening temperature (Beresford et al. 2001)

Counts of probiotic bacteria

Counts of *Lb. helveticus*and *Lb. planatrum* during Gouda cheese repining are presented in Fig. 1 *Lb. plantarum* was found to be survive and grew well during the ripening period. In fresh cheese the counts were 8.35 log cfu/g. This was followed by gradual increase during the first 60 days of ripening to reach 8.71 and 9.14 log cfu/g at 30 and 60 days of ripening respectively.This increase was followed by gradual decrease,but the counts still more than 8 log cuf/g as showenin Fig. 1. This agrees with the results given by El- Nagar et al. (2010) who found that counts of *Lb. helveticus* gradually increased during Gouda cheese ripening reaching the maximum counts after 60 days of ripening and then slightly decreased at the end of ripening period. In all cases, the counts were more than 30 x10^5cfu/g.This probiotic bacteria showed positive impact on flavour development of Cheddar cheese and Gouda cheese as mentioned by Hannon et al. (2007) and El- Nagar et al. (2010) respectively. However, presence of such bacteria or their lysis are quite important for Cheddar cheese flavour (Kenny et al. 2006; Hannon et al. 2007).

Nearly similar trend of results was observed with respect to counts of *Lb. plantrum.* The number in fresh cheese was 8.22 log cuf/g and gradually increased to reach the maximum of 8.92 log cuf/g when the old of cheese was 60 days and then decreased to be 7.93 log cuf/g at the end of ripening period .The probiotic activity and the role of *Lb. plantrum* in Cheddar cheese were recently given by Duan et al. (2019).

Finally, survivability and growth of the used probiotic bacteria during manufacture and ripening of Gouda cheese are quite important since their counts were always higher than minimum required counts given in the literature (10^6cfu/g as a minimum) to cause the probiotic effect (Granato et al. 2010).

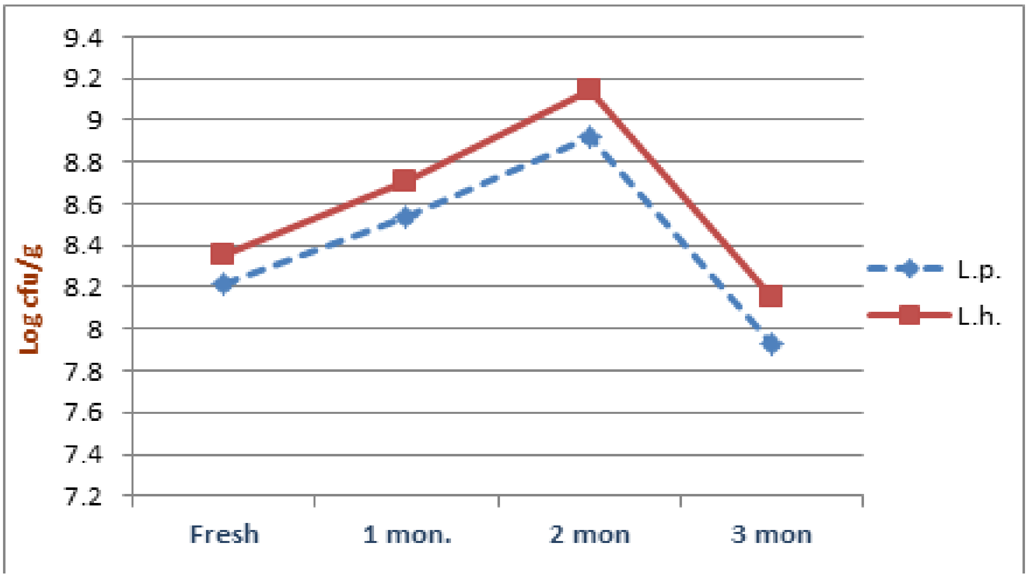

Fig. 1. Counts (log cfu/g) of *Lb. helveticus and Lb. plantarum* of fresh Gouda cheese and during ripening period of 3 months.

Detection of yeasts and moulds

Presence of yeasts and moulds gives unpleasant impression about the food, but the visual examination of our all cheese wells even those of ripened cheese revealed absence of such microorganisms in the control and the treated Gouda cheese. Table 2 shows that all fresh samples were free of yeasts and moulds but the lab. examination using specific medium revealed presence numbers of them.Relativelyhigher counts were detected in cheese from T1, while lower counts were observed in T3. In all cases a gradual increase ($p \leq 0.05$) in the counts was recorded with advancing ripening period .

The present results are in agreement with those given by El- Nagar et al. (2010) who mentioned that no colonies of yeasts and moulds were detected, in all fresh and 30 days Gouda cheese samples, but after the second month of ripening few colonies were observed. Such finding may be attributed to developing of more lactic acid and other acids responsible for decreasing the pH that creating suitabe optimum conditions for growth of yeasts and moulds. However, such finding had no adverse impact on the general appearance and flavour of all cheese samples even at the end of ripening period as will be discussed in the following section.

Concerning coliform bacteria, no colonies were detected in the control and all cheese treatments either when fresh or during the ripening period. The same finding was given by El- Nagar et al. (2010) who tried to accelerate ripening of Gouda cheese by means of using *Lb. delbrueckii* sub sp. *helveticus*. This reflects the good hygienic standards and sanitary conditions during the cheese making and ripening period. The role of lactic acid bacteria in preservation of the product which associated with their ability to produce a range of antimicrobial compounds should be taken into consideration in this respect (Gould, 1991).

Organoleptic properties of Gouda cheese

The scoring points given for thesensorial attributes of Gouda cheese as affected by the applied treatments are shown in Table 3. The first procedure in the scoring of cheese is the examination of the finish or make-up and noticing if the general appearance (GA) is neat and attractive or not (Nelson and Trout, 1981). At thebeginning of ripening the control cheese had the highest score ($P \leq 0.05$) comparing to cheese from T1, T2 and T3 which were not differed significantly in their scoring points. A significant increase in the scores of all samples was noticed with advancing ripening period but still showing that the control cheese had the maximum score of 9.28 out of 10 points whereas the score of T1, T2 and T3 were 8.3, 8.44 and 8.58 out of 10 respectively. The colour of all samples was bright, uniform and clear. The gradual increase in the scores during ripening agrees with the results of El-Nimr et al. (2010).

Type of milk seems to be an important factor affecting body and texture of Gouda cheese since when cow's milk was partially replaced with buffalo's milk, the body and texture of the resultant cheese (T1) had always lower scoring points when compared with the control cheese made using cow's milk. This was true in fresh and ripened cheese of any age. Table 3 shows that the differences in scoring points of C and T1 with respect to body and texture were statistically significant. Nature and composition of caseinate from buffalo's milk are responsible for the slightly hard body and crumbly texture of the resultant semi-hard and hard cheese in general –as previously reported in many earlier studies. However, the use of probiotic bacteria greatly improved the body and texture of the resultant Gouda cheese.

TABLE 2. Count of yeasts and moulds (log cfu/g) detected during ripening of Gouda cheese as affected by presence of buffalo's milk, Lb.helveticus CH5 and Lb. plantarum ATCC14917 (Average ± SD of 3 replicates)*

Ripening period (days)	Treatments				LSD (0.05)
	C	T1	T2	T3	
Fresh	ND	ND	ND	ND	------
30	2.16 ± 0.121Bc	2.64 ± 0.310Ac	2.29 ± 0.207ABc	2.05 ± 0.161Bc	0.3997
60	3.50 ± 0.347Bb	4.03 ± 0.155Ab	3.18 ± 0.166Bb	3.16 ± 0.243Bb	0.4534
90	4.84 ± 0.337Aa	4.85 ± 0.276Aa	4.34 ± 0.192ABa	4.30 ± 0.298Ba	0.5294
LSD	0.4700	0.4180	0.3091	0.3934	

*See Legend to Table (1) for details.

- ND = not detected.

TABLE 3. Sensory evaluation of Gouda cheese during ripening period of 90 days asaffected by the applied treatments (Average ± SD scores given by 10 judges)*.

Property	Ripening period (days)	Treatments				LSD (0.05)
		C	T1	T2	T3	
General appearance (10)	Fresh	7.46 ± 0.215Ad	6.900.170 ± Bc	6.780.120± Bd	6.860.133 ±Bd	0.3436
	30	7.92 ± 0.190Ac	7.47 ± 0.255BCb	7.15 ± 0.146Cc	7.54 ± 0.182Bc	0.3723
	60	8.53 ± 0.250Ab	7.67 ± 0.183Bb	7.80 ± 0.180Bb	8.36 ± 0.110Ab	0.3536
	90	9.28 ± 0.194Aa	8.30 ± 0.190Ba	8.44 ± 0.207Ba	8.58 ± 0.071Ba	0.3293
	LSD	0.2461	0.3816	0.3487	0.4031	
Body (10)	Fresh	7.53 ± 0.212Ac	6.84 ± 0.143Bc	6.94 ± 0.105Bd7.45	7.51 ± 0.322Ac	0.4002
	30	7.94 ± 0.182Ab	6.33 ± 0.172Cd	± 0.243Bc	7.83 ± 0.179Abc	0.3695
	60	8.22 ± 0.196Ab	7.65 ± 0.186Bb	8.03 ± 0.091Ab	8.25 ± 0.231Ab	0.3455
	90	8.92 ± 0.046Aa	8.37 ± 0.233Ba	8.73 ± 0.155Aa	9.06 ± 0.242Aa	0.3518
	LSD	0.3244	0.3518	0.3016	0.4689	
Texture (10)	Fresh	7.24 0.160 ±Ad	6.940.120 ±Bc	6.82 ±0.150 Bd	7.34 ± 0.200Ad	0.3023
	30	7.72 ± 0.140Ac	7.36 ± 0.187Bb	7.18 ± 0.135Bc	7.90 ± 0. 195Ac	0.3138
	60	8.54 ± 0.179Ab	7.95 ± 0.186Ba	8.01 ± 0.111Bb	8.48 ± 0.216Ab	0.0293
	90	9.26 ± 0.172Aa	8.16 ± 0.092Ba	8.47 ± 0.245Ba	9.43 ± 0.240Aa	0.0293
	LSD	0.3088	0.2863	0.3174	0.4023	
Flavour (20)	Fresh	14.70 ± 0.260Ad	13.56 ± 0.427Bc	13.87 ± 0.332Bd	14.59 ± 0.241Ad	0.5286
	30	15.64 ± 0.310Ac	14.69 ± 0.240Bb	14.96 ± 0.282Bc	15.50 ± 0.203Ac	0.4283
	60	17.07 ± 0.483Ab	15.08 ± 0.330Bb	15.69 ± 0.293Bb	16.70 ± 0.223Ab	0.5648
	90	18.59 ± 0.644Aa	16.37 ± 0.320Ba	16.25 ± 0.180Ba	18.07 ± 0.257Aa	0.6408
	LSD	0.7360	0.5490	0.4536	0.3788	

* - See legend to Table (1) for details.

- The values given in parentheses represent the maximum attainable scores.

Table 3 shows that the use of *Lb. helveticus* (T2) and Lb. plantarumATCC14917 (T3) greatly increased the scores given for body and texture of Gouda cheese samples as compared with those of T1 while the differencesin the scores given for C and T3 were insignificant (P ≥0.05) at any ripening time. However, at the end of ripening period the scores given for the body of cheese were 8.92 and 9.06 out of 10 points for C and T3 samples respectively, whereas those for texture were 9.26 and 9.43 out of 10 in order. Corresponding scores for T2 boy and texture were 8.73 out of 10 (P >0.05) and 8.47 out of 10 (P≤ 0.05) respectively when compared statistically with the scores of C and T3. Exterkate et al. (1987) and David et al. (1988) attributed such finding to role the proteolytic enzymes from the used starter bacteria and the probiotic bacteria. The role of *Lb. helveticus* in this respect was given during ripening of Gouda cheese made by El-Nagar et al. (2010).

Concerning flavour of Gouda cheese, Table 3 shows that the use ofbuffalo's milk in a mixture with cow's milk (T1) significantly decreased the scores given for flavour than those of C cheese made fromcow's milk only.This was noticed at any ripening time suggesting the adverse impact of buffalo's milk in developing flavour components during ripening period. This agrees with the results of David ***et al***. (1988) who mentioned that Gouda cheese made from buffalo's milk required longer time for ripening to be like the corresponding cheese made from cow's milk. In Egypt, many trials and studies were done since 1952 to manufacture hard cheese from buffalo's milk but the resultant cheese barely resembled the corresponding cow's milk cheese (Mehanna et al. (1985). However, in the last two decades an attention was directed towards the use of selected bacteria with the traditional cheese starter to improve quality of different cheeses and toaccelerate the ripening process (El-Sodaa et al. 2000; Boylston et al. 2004; Hannon et al. 2007; Plessaset al.2012; Ehsani et al. 2018; Duan et al. 2019). The present study contributed in this respect since the use of *Lb. helveticus* CH5 (T2) and *Lb. plantarum* ATCC14917(T3) greatly improved the sensorial properties of Gouda cheese made from mixed cow'sand buffalo's milk (3:1).

Conclusion

In conclusion, a good quality probiotic Gouda cheese can be manufactured successfully using *Lactobacillus helveticus* CH5 or *Lactobacillus plantarum* ATCC14917 with the traditional Gouda cheese starter culture.

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جودة جبن الجودا ذات التأثير الحيوي كغذاء وظيفي

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جبن الجودا هولندية المنشأ تصنع عادة من اللبن البقري.. ولوفرة اللبن الجاموسي في مصر ووفقاً لتوصية دستور الأغذية (كودكس ٢٠١٩) بامكانية استخدام اللبن الجاموسي منفرداً أو مع اللبن البقري فقد اهتمت هذه الدراسة بتصنيع جبن الجودا من اللبن البقري (جبن المقارنة) ومن خليط اللبن البقري مع اللبن الجاموسي (١:٣) واستخدام البادئ التقليدي.. وتم ذلك في المعاملة (١) أما في المعاملة رقم (٢) ورقم (٣) فتم استخدام اللبن الخليط والبادئ التقليدي مع اضافة *Lb. helveticus* CH٥ (معاملة ٢) و *Lb. plantarum* ATCC١٤٩١٧ (معاملة ٣) كبكتريا ذات تأثير حيوي (Probiotic).

يمكن اجمال النتائج المتحصل عليها من تحليل الجبن الطازج وخلال فترة التسوية التي استمرت٩٠ يوماً فيما يلي :-

- كان العدد الكلي للبكتريا وأعداد البكتريا المحللة للبروتين والمحللة للدهن أعلي في جبن المعاملات رقم (٢) و(٣) مقارنة بجبن المقارنة وجبن المعاملة رقم(١) في الجبن الطازج وخلال فترة التسوية.. ولم تتأثر الاعداد معنويا باستخدام اللبن الجاموسي مع اللبن البقري عند نهاية فترة التسوية عند مقارنة الاعداد في جبن المعاملة (١) وجبن المقارنة.
- زادت أعدادالبكتريا ذات التأثير الحيوي بتقدم فترة التسوية وكانت أكبر الاعداد عند عمر ٦٠ يوماً لجبن المعاملة رقم (٢) ورقم (٣) وتناقصت الاعداد تدريجياً بتقدم فترة التسوية بعد ذلك ولكن في كل الأحوال كانت الاعداد أكبر بكثير من الحد الأدني اللازم لاحداث التأثير الحيوي المطلوب.
- كانت كل عينات الجبن الطازج خالية تماماً من الخمائر والفطريات وبكتيريا الكوليفورم ولكن ظهرت الخمائر والفطريات تدريجياً خلال فترة التسوية وكانت الزيادة معنوية إحصائياً في كل العينات بتقدم فترة التسوية.

أوضحت نتائج التقييم الحسي أن جبن المقارنة مختلف العمر حاز نسبيأعلي أعلي الدرجات للمظهر العام واللون رغم تجانس اللون وجاذبيته في كل العينات أما بالنسبة للقوام والتركيب والنكهة فكانتأعلي الدرجات نسبياً لجبن المقارنة وجبن المعاملة رقم (٣) ولم تكن الاختلافات بينهما معنوية إحصائياً.