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EFFECT OF PEDIOCOCCUS ACIDILACTICI AND ITS BACTERIOCIN ON SOFT CHEESE QUALITY AND VALIDITY

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A B S T R A C T

Lactic acid bacteria (LAB) are widely applied in the food industry and can produce antimicrobial substances, as bacteriocins. *Pediococcus acidilactici* is a homofermentative LAB, used to produce more safe fermented food due to production of bacteriocin named pediocin that inhibits several spoilage and pathogenic microrganisms. The present study was done as a trial to prolong the shelf life and enhance the quality of soft cheese by using *pediococcus acidilactici* and its bacteriocin (pediocin). Three different treatments of soft cheese were prepared: C, control samples (without addition either *Pediococcus acidilactici* or its extracted pediocin), addition of P (*Pediococcus acidilactici* 1%) and BP (crude pediocin extracted from *pediococcus acidilactici*), then all treated samples were freshly examined and during cold storage time in refrigerator at (5-7°C) till signs of spoilage were detected for their organoleptic, acidity indices and microbiological status. Results revealed that bacteriocin extracted from *pediococcus acidilactici* (BP) extended the shelf life of soft cheese to 34 days with maintaining good organoleptic characteristics, and samples treated with *Pediococcus acidilactici* strain extended the shelf life to 29 days, while control samples to 18 days.

KEYWORDS: Soft cheese**.** *Pediococcus acidilactici.* bacteriocin

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1. INTRODUCTION

Lactic acid bacteria (LAB) have acquired considerable relevance in the food industry and in public health. They are widely used in fermented foods, have a long history of safe use, and are commonly given the Generally Recognized as Safe (GRAS) status [1]. *Pediococcus acidilactici* is commonly found in fermented vegetables, fermented dairy products and meat [2]. It has been found as members of the normal flora of the alimentary tract, including the oral cavity and gastro intestinal tract, and is used as probiotics in the food industry [3]. The preservative ability of LAB in food is attributed mainly to the production of antimicrobial substances, including organic acid, hydrogen peroxide and bacteriocins [4]. Bacteriocins are ribosomally synthesized, biologically active peptides or proteins with antagonistic activity against specific microorganisms [5]. The incorporation of these compounds as biopreservative ingredient into model food has been shown to be effective in the control of pathogenic and spoilage microorganisms [6]. Although bacteriocins can be produced in the food during fermentation, bacteriocins by LAB can be produced in much higher amounts in vitro during fermentations under optimal physical and chemical conditions [7]. Several publications have reported that bacteriocinogenic Pediococci strains could be used as bioprotective cultures for food manufacturing processes [8]. Pediocin

produced by *pediococcus acidilactici* is another well-studied bacteriocin that will likely be the second LAB bacteriocin to be widely used in the food industry [9]. Cheese is a vital dairy product with best nutritional value and health care function, and it is widely popular in many countries in the world with good taste and diverse flavor and has a long history in the human diet [10]. As, soft cheese is highly perishable and thus has a short shelf life, even at refrigerated temperature [11], the objectives of the present work was a trial to prolong shelf life of soft cheese by application of *pediococcus acidilactici* and its extracted bacteriocin and monitoring the quality of cheese by evaluation the organoleptic, acidity indices and microbiological changes in soft cheese during refrigeratd storage.

2. MATERIALS AND METHODS

2.1. Activation of pediococcus acidilactici strain

Lyophpilized single strain *Pediococcus acidilactici* ATCC 25740 was obtained from Cairo-MIRCEN (Microbiological Resource Center) Faculty of Agriculture, Ain Shams University, Cairo, Egypt. The strain was activated on 9ml sterile MRS broth (Biolife, Italy) at 37ºC/24hr, then three culture transfers were performed to activate the culture [12]. The strain was subcultured into reconstituted sterile skim milk powder and incubated at 37 °C for 24 hr for further activation of the bacterial strain till obtain the suitable probiotic concentration of 1×10^{12} cfu/ ml and then kept in refrigerator within 24hr [13].

2.2. Extraction of crude bacteriocin from pediococcus acidilactici strain

Ten ml of activated culture were separately inoculated into one liter of MRS broth under aseptic condition and incubated at 37°C /16 hr as described by [14]. Then the cultures was adjusted to pH 2.0 by adding HCL 1N then cultures were heated in water bath at 100ºC for 5min.The cells were harvested by centrifugation at 10,000 rpm for 20 min at 4ºC and re-centrifugated under the same conditions. The supernatants containing bacteriocin extracts were collected sterilized by using 0.45 µm –pore size Seitz filter with single sheet to eliminate the possible presence of viable bacterial cells and obtain cell free supernatant [15].

2.2.1. Detection of bacteriocin titer

The titer of bacteriocin was quantified by agar well diffusion method [16]. Indicator pathogenic microorganisms, *Bacillus subtilus*, *Staphylococcus aureus* and *Escherichia coli* at the concentration of 1×10^{6} cfu/ml were inoculated into Muller Hinton agar poured into sterilized Petri dishes then leaving the plates for solidification. Wells were made on the solidified agar with sterile cork borer (10 mm in diameter) then inoculated with 100 µl of two fold serial dilutions from bacteriocin extracts [17]. The plates were incubated at 37 ºC/24 hr and then examined for clear circular inhibition zone around the wells. The titer of inhibition was defined as the reciprocal of the highest dilution showing definite inhibition zone and was expressed as activity units (AU) per ml. The activity unit (AU/ml) was calculated according to the following Formula: AU/ml = (1000 / V) x 2^{y} Where, AU is arbitrary unit of bacteriocin activity, y is the number of the last dilution showing inhibition and V is the volume of the supernatant (µl) which inoculated in each well. Bacteriocin activity was recorded as positive if the width of the clear inhibition zone around the colonies of the producer was 2 mm or larger [18]. The experiment was repeated 3 times.

2.3. Preparation of soft cheese

A total of eighteen liter of fresh raw mixed milk of cows and buffalos (1:1) were obtained from the herd of Faculty of Veterinary Medicine, Benha University. A soft cheese

was manufactured according to the method described by Abdalla and Ahmed [19] with some modification according to Shakeel-Ur-Rehman *etal.*, [20] as follows: Raw milk was fortified with milk protein concentrate (70% protein), heat treated at 80°C for 30 minutes then calcium chloride (0.02%), glucono delta lactone 1.7/L% (v/v) and Rennet (0.01v/v)were added then salted by sodium chloride (5%) at 37°C, The bulk volume of milk was divided into 3 groups (2 liter each) and inoculated by the following:
P: *Pediococcus acidilactici* ATCC 25740 (1%).
BP: bacteriocin of *pediococcus acidilactici* ATCC 25740 (100 AU ml^{-1}). C: without inoculation. Then they were incubated at 37 °C for 1hr till curd formation occurs and left for whey separation overnight, then packaged in sterile plastic cups and stored at refrigerator temperature (5-7°C). The cheese samples were examined organoleptically, acidity indices and microbiologically at zero time, 7, 14, 17, 21, 23, 26, 29, 32 and 34 day till signs of spoilage were detected. The experiment was repeated 3 times and the average results for each treatment were recorded.

2.4. Organoleptic evaluation

Cheese samples were evaluated for flavor (45 points), body and texture (35 points) and appearance (20 points) with overall score 100 points [21].

2.5. Acidity indices

2.5.1. Determination of pH

The pH values were determined using pH meter (Jenway model 3510 made in UK) [22].

2.5.2. Titratable acidity (T.A%)

Acidity in soft cheese was estimated by standard method for titratble acidity method according to [23].

2.6. Microbiological examination

2.6.1. Preparation of serial dilutions

Soft cheese samples were homogenized with sterile solution (2% w/v) of sodium citrate. One ml from cheese homogenate was added to 9 ml of sterile peptone water (1%) from which ten fold serial dilutions were prepared [24].

2.6.2. Determination of Pediococcus acidilactici count

Pediococcus acidilactici count was determined using MRS agar and was incubated at 37 ºC for 48hr [24].

2.6.3. Determination of Coliform count

The coliform count was done using most probable number (MPN) by using 3 fermentation tubes containing MacConkey broth supplemented with Durham's tubes and were incubated at 37 ºC / 24 hr [24].

2.6.4. Determination of total yeast and mould count

They were determined using Sabouraud dextrose agar medium supplemented with chloramphenicol and chlortetracycline (100 mg of each) and the plates were incubated at 25 ºC for 5-7 days as described by IDF [25].

2.7. Statistical analysis

Experiments were done three trials on different times. The statistical analysis was applied by using SPSS 16.0 Analysis of Variance (ANOVA) for windows [26].

3. RESULTS

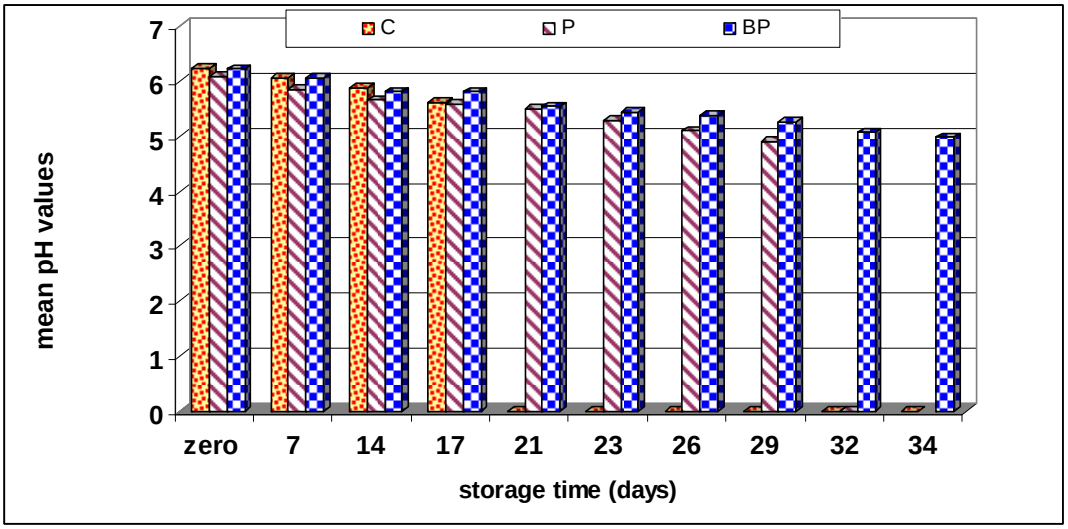
Table (1) showed significant differences ($p \leq 0.05$) in the mean overall score between BP samples and the other treatments at the zero day ,after 7, 14, 17, 21,23,26,29,32 and 34 day of storage . Also, BP samples showed significant difference in flavour, body and texture compared to other treatments. On the other hand, there is no significant differences ($p > 0.05$) between groubs in appearance during time of storage. Figure (1) observed the relatively high pH values at zero day.

Table (1): The mean values of organoleptic overall scores for the examined cheese samples during their refrigerated storage.

Storage time (days)	Flavour (45)			Body and texture (35)			Appearance(20)			Overall score(100)		
	C	P	BP	C	P	BP	C	P	BP	C	P	BP*
Zero	39.26±0.53[b]	39.18±0.12[b]	41.79±0.26[a]	30.64±0.36[b]	31.65±0.43[b]	32.52±0.30[a]	18.47±0.20[a]	18.38±0.29[a]	18.93±0.35[a]	88.36±0.56[b]	89.22±0.64[b]	93.24±0.15[a]
7	39.61±0.8[b]	39.46±0.23[b]	41.77±0.22[a]	30.45±0.34[b]	31.31±0.44[b]	32.90±0.66[a]	17.80±0.15[b]	18.40±0.22[a]	18.40±0.19[a]	87.87±1.13[c]	89.17 ±0.43[b]	93.07±0.87[a]
14	35.65±0.80[c]	39.32±0.76[b]	42.07±0.81[a]	29.53±0.58[c]	31.94±0.09[b]	32.53±0.35[a]	17.03±0.55[b]	18.17±0.90[a]	18.60±0.31[a]	82.55±1.26[c]	88.76±0.99[b]	39.20±0.31[a]
17	33.34±0.78[c]	39.56±0.50[b]	41.80±0.15[a]	26.44±0.72[c]	31.44±0.74[b]	32.43±0.81[a]	15.83±0.23[c]	18.17±0.67[a]	17.62±0.81[a]	75.61±1.51[c]	89.17±1.60[b]	91.85±0.88[a]
21	S	40.24±0.45[a]	41.65±0.43[a]	S	31.34 ±0.70[a]	32.72±0.40[a]	S	17.80±0.26[a]	17.41±0.82[a]	S	89.08±0.75[b]	91.34±0.48[a]
23		40.07±0.60[a]	41.17±0.18[a]		31.43±0.32[a]	31.20±0.21[a]		16.97±0.48[a]	17.17±0.27[a]		87.73±0.40[a]	88.30±0.23[a][b]
26		39.33±0.60[b]	39.93±0.18[a]		29.50±0.38[b]	30.33±0.35[a]		16.50±0.79[a]	16.97±0.38[a]		85.33±0.97[b]	87.23±0.73[a]
29		36.86±0.52[b]	38.97±0.48[a]		27.77±0.83[b]	30.18±0.63[a]		14.27±0.35[b]	16.40±0.55[a]		78.90±0.37[b]	85.55±0.43[a]
32		S	37.43±0.43		S	26.76±0.87		s	16.05±0.61		S	77.62±0.95
34			34.87±0.55			26.26±0.57			15.68±31			76.72±0.82

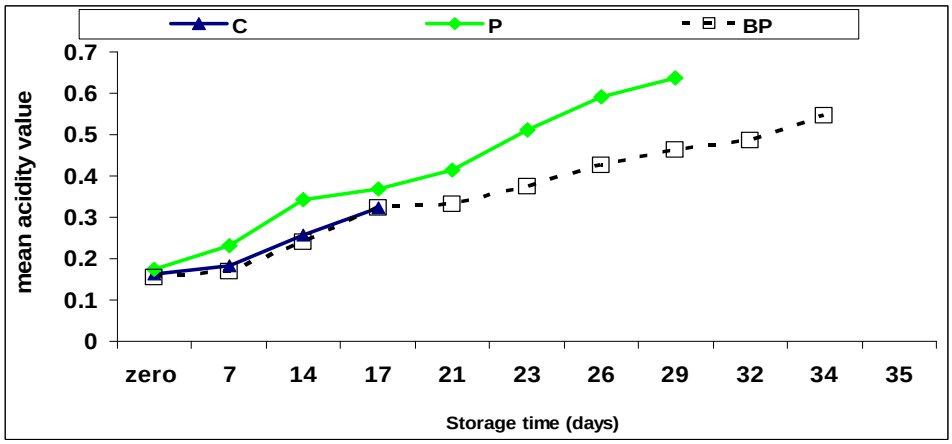
C: control samples. P: *Pediococcus acidilactici* (1%).
BP: *Pediococcus acidilactici* bacteriocin (100AU/ml).
N.B: Results shown are means of triplicates of each treatment.
a, b, c: Mean values in the same row having different superscripts significantly different ($p \leq 0.05$).
S: The spoilage samples, *S.E.: Standard Error ND: Not detected

Figure (1): The mean pH values for the examined cheese samples during their refrigerated storage.



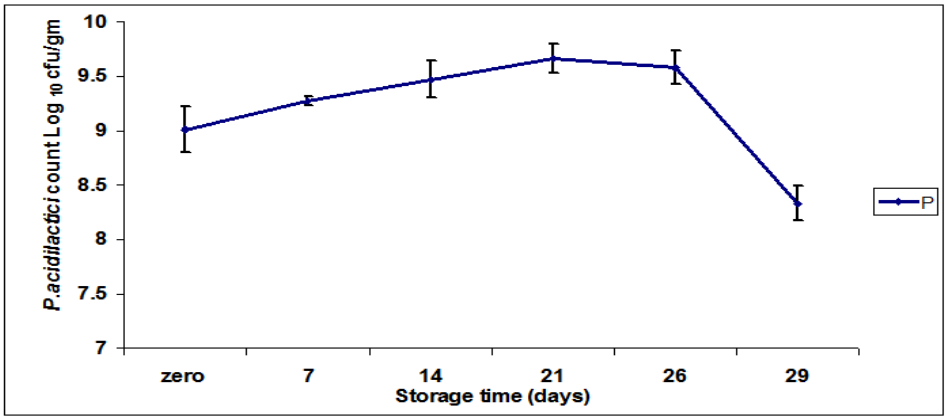
C: control samples. P: *Samples treated with Pediococcus acidilactici* (1%).
BP: *Pediococcus acidilactici* bacteriocin (100AU/ml).
N.B: Results shown are means of triplicates of each treatment.

Figure (2): The mean titratable acidity values of the examined cheese samples during their refrigerated storage.



C: control samples. P: *Samples treated with Pediococcus acidilactici* (1%).
BP: *Samples treated with Pediococcus acidilactici* bacteriocin (100AU/ml).
N.B: Results shown are means of triplicates of each treatment.

Figure (3) Viability *Pediococcus acidilactici* in *Pediococcus acidilactici* treated soft cheese samples during their refrigerated storage.



P: 1% *pediococcus acidilactici.*
N.B: Results shown are means of triplicates of each treatment.

Table (2) The mean values of yeast and mould count (Log_{10} cfu/g) for the examined cheese samples during their refrigerated storage.

Storage time (days)	Yeast count (mean± S.E)			Mould count		
	C	P	BP*	C	P	BP
Zero	ND	ND	ND	ND	ND	ND
7	ND	ND	ND	ND	ND	ND
14	2.17±0.06	ND	ND	ND	ND	ND
17	2.45±0.04	ND	ND	1.10	ND	ND
21	S	1.49±0.12	ND	S	ND	ND
23		1.81±0.07[a]	1.62±0.09[a]		ND	ND
26		1.98±0.04[a]	1.81±0.06[b]		1.00	ND
29		2.00±0.01[a]	1.96±0.03[b]		S	ND
32		S	2.15±0.04[b]			ND
34			2.36±0.07			ND

C: control samples. P: *Samples treated with Pediococcus acidilactici* (1%).
BP: *Samples treated with Pediococcus acidilactici* bacteriocin (100AU/ml).
N.B: Results shown are means of triplicates of each treatment.
a, b, c: Mean values in the same row having different superscripts significantly different ($p \leq 0.05$).
S: The spoilage samples
*S.E.: Standard Error

There were decrease in pH values during the storage period in all treatments till the end of shelf life, while BP cheese samples showed higher pH values compared with other treatments with significant differences ($p \leq 0.05$). Figure (2) showed titratable acidity of all cheese samples showed the opposite trait to pH values and BP showed the least titratable acidity with significant difference($p \leq 0.05$). Figure (3) showed increase in *Pediococcus acidilactici* count during storage period with maintaining probiotic level. The results in this study showed coliform failed to be detected in all the examined cheese samples in either fresh or during cold storage till end of shelf life. Table (2) showed there were significant differences in yeast count ($p \leq 0.05$) between BP and P samples. Mold appeared in each treatment prior to spoilage.

4. DISCUSSION

The overall sensory evaluation of soft cheese during refrigerated storage was shown in table (1). The scores slightly were increased within the first 14 day of storage then decreased till end of storage, this result was agreed with that reported by Deghidi *et al.*, [27]. The mean overall organoleptic scores for *Pediococcus acidilactici* treated cheese were 89.22±0.64, 89.17 ±0.43, 88.76±0.99, 89.17±1.60, 89.08±0.7587 .73±0.40, 85.33±0.97 and 78.90±0.37at zero day, 7, 14, 17, 21, 23, 26 and 29 days of storage, respectively. This result agreed with that reported by Verachia , [28] who found that Pediococcus strain when added to cheese milk along with starter culture, enhanced the flavour of cheese. Also the addition of pediococci reduced the time needed for cheese maturation as well as improved its flavour due to acceleration formation of volatile compounds [29]. Bacteriocin treated soft cheese samples showed the highest overall organoleptic scores from the day of production, till the end of shelf life (at 34 days) with mean values of 93.24±0.15 and 76.72±0.82, respectively (table,1). On the other hand, the mean overall organoleptic scores for control samples showed the minimum overall organoleptic scores with mean values of 88.36±0.56, 87.87±1.13, 82.55±1.26 and 75.61±1.51 at zero day, 7, 14 and 17 days, respectively. This result nearly agreed with that reported by Kebary *et al.*, [30]. Concerning with the pH informs precisely about the freshness state of milk and milk products and is used as indicator of acidity [31]. The mean pH values of the examined soft cheese treated samples at zero time were 6.24±0.06, 6.09±0.06 and 6.22±0.06 for C, P and BPcheese samples, respectively (Fig. 1). This result agreed with that reported by sobeih *et al.*, [32]. The relatively high pH values at zero day of cheese manufacture may be attributed to the time of drainage as the retention of calcium phosphate increased within the curd matrix, which act as a buffering agent against the developed acidity of cheese [33]. There were decrease in pH values during the storage period in all treatments till the end of shelf life, while BP cheese samples showed higher pH values compared with other treatments with mean values of 6.05±0.06, 5.80±0.05, 5.70±0.06, 5.54±0.03, 5.42±0.02, 5.37±0.04, 5.25±0.04, 5.07±0.03 and 4.97±0.04 after 7, 14 , 17, 21, 23, 26, 29, 32 and 34 days of refrigeration storage, respectively(Fig.1). This result may be due to the fact that bacteriocin has inhibitory effect on lactic acid bacteria [34]. Control cheese samples showed the lowest pH with mean values of 6.05±0.06, 5.87±0.06 and 5.32±0.08 after 7, 14 and 17 days of refrigeration storage, respectively. The low pH may be due to absence of starter culture which has a role in developing a rapid acidification of milk through the production of lactic acid [35]. Figure (2) showed titratable acidity of all cheese samples showed the opposite trait to pH values which increased as the storage period progressed up to the end of storage period. These results were in agreement with

those reported by Elewa *etal.,* [36]. Bacteriocin extracted from *pediococcus acidilactici* (BP) showed the lowest acidity with mean values of 0.156 ±0.01 at zero day and 0.54±0.02 at 34th day of storage. This result agreed with those reported by Ibrahim and Elbarary [37] who found T.A% of pasteurized milk treated with bactriocin of *lactobacillus acidophilus* had less acidity than other treatments. *Pediococcus acidilactici* count was decreased from inoculation level during soft cheese preparation 10^{12} to reach 10^{9} cfu/g at the zero time in P as cheese samples shown in Fig (3). This decrease may be attributed to hydrolysis of GDL and formation of gluconic acid which slow down the growth of starter probiotic bacteria [38]. Glucono delta lactone (GDL) is a permitted food additive as acidulant. It is characterized by slow acid development and it has advantages of preserving food by lowering pH without other effects arising from the use of starters to develop acidity. Glucono delta lactone has been used in manufacture of many cheese varieties in replacement of starter culture [39]. During storage time, the viable counts of *pediococcus acidilactici* were gradually increased with mean values of 9.27±0.04, 9.47±0.17, and 9.66±0.13 Log_{10} cfu/g at 7th , 14th and 21st days of storage, respectively. Reaching maximum count at 26th day of storage with mean values of 9.58± 0.16 Log_{10} cfu/g then decreased to 8.33±0.16 Log_{10} cfu/g. These results agreed with those reported by Verachia, [28] who found that pediococci populations gradually increased to a final concentration of about a quarter of flora of ripened cheese. It is of great importance to said that when selecting bacteria for their physiological effects, they should stay viable during the whole shelf-life of the food product and resist the acidic environment of the stomach and to bile salts in the small intestine to be probiotic food [40]. It has also been suggested that the food product should contain at least 10^{6} cfu/g or equivalent to 7 Log_{10} cfu/g of the probiotic bacterial strain and it must be consumed approximately 300–400 g per week such product [41]. The count of *Pediococcus acidilactici* at the end of shelf life still above the recommended level of probiotic (8.33± 0.16 Log_{10} cfu/g). These results came in accordance with those reported by Mehanna, *et al.,* [42]. *Pediococcus acidilactici* has a health benefit in improving the health condition of patient has constipation, diarrhea, relieving stress, enhancing immune response and prevent colonization of the small intestine caused by some pathogens like Shigella, Salmonella, *Clostridium difficile* and *Escherichia coli* in the small intestine [43]. Coliform are routinely used as indicator to the hygienic quality of food products. Their presence indicates careless methods of production, handling of processed food products and the use of insufficient sanitized equipment. Moreover, Coliforms are killed during pasteurization and if they are present in the product they would be a result of post pasteurization contamination [44] and [45]. The results in this study showed coliform failed to be detected in all the examined cheese samples in either when fresh or during refrigerated storage till end of shelf life. These results agreed with those reported by Degheidi *et al.,* [27] and Gould, [46]. These results came in accordance with those reported by EOSQ [47] whom stated that coliform should not exceed 10 cfu/ g or 1 Log_{10} cfu/g of soft cheese. Regarding to yeast and moulds which play an important role in spoilage of dairy product primarily in fermented milks and cheeses [48], Contamination with undesirable moulds has been a serious problem to the dairy industry resulting in huge economic losses in cheese and other fermented foods [49]. From table (2) it shown that yeast began to appear in P cheese samples at the 20th days of refrigerated storage with the mean values of 1.49±0.12 and increase slightly till end of shelf life at the 29th day with mean value of

2.00±0.01 Log_{10} cfu/g. This result agreed with those reported by Degheidi *et al.*, [27] who found that moulds and yeasts began to appear after 20th days of storage in probiotic treated white soft cheese. This may be attributed to that *Pediococcus acidilactici* had inhibitory effect on yeast growth [50] These counts came in accordance with those reported by EOSQ [47] whom stated that yeast count should not exceed 400 cfu/ g of cheese. Yeasts were detected in C cheese samples at 14th day of storage and increased in the count till the end of shelf life in refrigerated storage with a mean value of 2.17±0.06 and 2.45±0.04 Log_{10} cfu/ g, respectively (table 2). But BP cheese samples showed the absence of yeast till 23 days of storage begin to grow at day 23rd with mean value of 1.62±0.09 Log_{10} cfu /g then increased to 2.36±0.07 Log_{10} cfu/ g at 34th day of refrigerated storage. It may be referred to inhibitory effect of crude bacteriocin extracted from *P.acidilactici* against yeast and mold growth [51]. While, mould was not detected in all treated cheese samples and begin to appear just prior to spoilage in control samples at day 17th with mean value of 1.10 Log_{10} cfu/g and in P samples at day 29th of refrigerated storage with mean value of 1.00 Log_{10} cfu/ g (table 2) . The low counts of mould agreed with those reported by EOSQ [47] whom stated that mould count should not exceed 10 cfu/g of cheese. On the other hand, BP showed no mould growth during refrigerated storage from zero time till end of refrigerated storage period. These findings came in agreement with those of Effat, [52]. This may due to the use of antifungal metabolites produced by a *Pediococcus acidilacti* strain [53].

Conclusion

The present study showed that the addition of *pediococcus acidilactici* culture 1% in soft cheese improved its quality during refrigerated storage as well as extended the shelf life up to 29 days. While, Pediocin; the crude bacteriocin extracted from *Pediococcus acidilactici*; can used safetly in soft cheese manufacture due to its antibacterial and antifungal effect and extend the shelf life up to 34 days of storage with maintaining good organolptic properties.

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تأثير سلاله البيديوكوكسس اسيديلاكتسى والبكتريوسين الناتج منه على جوده وصلاحيه الجبن الطري

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الملخص العربي

تعتبر سلالات بكتريا حمض اللاكتيك الاكثر تطبيقا فى صناعه الأغذية ومنها سلاله *Pediococcus acidilactici* حيث لها القدرة على انتاج مواد مضادة للميكروبات مثل البكتريوسين. لذلك تمت هذه الدراسة كمحاوله لزيادة فتره الصلاحية وجوده الجبن الطري باستخدام سلاله البيديوكوكسس اسيديلاكتسى والبكتريوسين الناتج منه كمواد حافظه من أصل حيوي. تم تحضير ثلاث معاملات مختلفة للجبن الطري. المجموعة الضابطة بدون اضافه السلالة البيديوكوكسس اسيديلاكتسى او البكتريوسين لها ومجموعه مضاف اليها 1% من سلاله البيديوكوكس اسيديلاكتسى ومجموعه مضاف اليها البكتريوسين المستخلص من هذه السلالة. ثم تم تخزين هذه العينات بالثلاجة حتى ظهور علامات الفساد من نمو فطريات وخمائر. وقد تم تحليل هذه العينات عند يوم انتاجها وخلال فتره التخزين تقييما حسيا وقياس الحموضه والاس الهيدروجينى وميكروبيولوجيا . وقد وجدت النتائج ان الجبن الطري المصنع باستخدام سلاله البيديوكوكسس اسيديلاكتسى زادت فتره الصلاحية به الى 29 يوم بينما زادت فتره الصلاحية للجبن المصنع باستخدام بكتريوسين البيديوكوكسس اسيديلاكتسى الى 34 يوم مع الحفاظ على خصائص الحسية والمكروبيولوجية جيده. بينما كانت فتره صلاحيه المجموعة الضابطة الى 18 يوم. ولقد تنافصت قيمه الاس الهيدروجينى وتزايدت نسبه الحموضة فى جميع العينات اثناء فتره التخزين. وبالنظر الى عينات الجبن المضاف لها البيديوكوكسس اسيديلاكتسى فقد استطاع لها البيديوكوكسس اسيديلاكتسى البقاء حيا فى عينات الجبن بمتوسط قدره 0.21± 9.01 عند يوم التصنيع الى ان وصل $8.33\ Log_{10}\ cfug^{-1}$ عند اليوم ال29 من التخزين بالثلاجة.
كما اوضحت النتائج خلو جميع العينات من الخمائر منذ بداية التصنيع ولكن ظهرت عند اليوم 23 فى العينات المحقونة بالبكتريوسين بمتوسط قدره 0.09± 1.62 بينما ظهرت عند اليوم 21 فى العينات المحقونة بسلاله البيديوكوكسس اسيديلاكتسى بمتوسط قدره0.12 ± 1.49ثم بدا العدد بالزيادة حتى نهاية فتره الصلاحية بينما المجموعة الضابطة ظهر فى اليوم ال14 وال17 فقط بمتوسط قدره 2.17± 0.06 و 0.04±2.45 على التوالي. وظهرت الفطريات فى المجموعة الضابطة عند اليوم ال17 وظهرت بعينات البيديوكوكسس اسيديلاكتسى عند اليوم ال29 بينما لم تظهر بالعينات التي تحتوى على البكتريوسين المستخلص من سلاله البيديوكوكسس اسيديلاكتسى وكان العد يطابق المواصفات القياسية المصرية لا يزيد عن 400 خليه للخمائر ولا يزيد عن 10 خلايا للفطريات لكل جرام . لم تظهر المجموعة القولونبه اثناء يوم الانتاج او اثناء فتره التخزين مما يدل على وجود الاشتراطات الصحية اثناء التصنيع والتخزين. ويستنتج من هذ الدراسة ان اضافه سلاله البيديوكوكسس اسيديلاكتسى الى الجبن الطرى تحسن الخصائص الحسيه للجبن مع المحافظة على العدد اعلى من الموصي به للحصول على فوائد صحيه وزاد الصلاحية الى 29 يوم. كما ان البكتريوسين الناتج منها زاد فتره الصلاحية للجبن الطرى الى 34 يوم مع المحافظة على خصائص حسيه جيده .

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