Influence of probiotic on microbiological quality of kariesh cheese

Esmat, I. awad[1]., Marwa, A. Ibrahim[2]and Manal. I. El-Shaer[2]

[1]Food Control Department, Faculty of Veterinary Medicine, Zagazig University, Egypt.
[2]Animal Health Research Institute, Zagazig, Egypt.
Esmatawad2@yahoo.com

Abstract: Thirty random milk samples were collected from dairy farms at Sharkia Governorate; each sample was divided into two parts. The first part was used as control while the second one was inoculated with active culture of two probiotic strains (*Lactobacillus casei* (ATCC- 334) and *Lactobacillus plantarum* (ATCC-8014)) in ratio 2% of milk. Both parts were manufactured into Kariesh cheese followed by microbiological analysis. The results indicated that out of examined control kariesh cheese samples 30(100%) were showed bacterial growth, the minimum was 1.0×10^8cfu/gm, the maximum was 8.8×10^{10} cfu/gm and the mean value was $7.86\times10^9\pm3.05\times10^9$ cfu/gm. While fresh kariesh cheese with probiotics showed that count was ranged from 9.0×10^5 to 3.0×10^7 with a mean value of $6.42\times10^6\pm1.29\times10^6$. There was significance difference between two groups ($P < 0.05$). All examined control kariesh cheese samples were contaminated with *Coliforms* while only 24(80%) of examined kariesh samples with probiotics were harbor *Coliforms* and the count were ranged from 2.0×10^3 and 2.0×10^3 to 1.9×10^5 and 1.2×10^5 with a mean value of $4.66\times10^4\pm9.3\times10^3$ and $1.47\times10^4\pm4.6\times10^3$ for control samples and samples with probiotics respectively *Staphylococcus aureus* count in control and kariesh samples with probiotics. All control kariesh cheese samples were positive for *Staphylococcus aureus*, the count was ranged from 1.0×10^4 to 2.9×10^7 and the mean value was $4.2\times10^6\pm1.2\times10^6$. Out of examined kariesh cheese samples with probiotics 21(70%) were contaminated with *Staphylococcus aureus*, The high level of contamination was 8.0×10^5 , the low level was 2.0×10^3 and the mean value was $9.4\times10^4\pm3.7\times10^4$. There was significance difference between two groups ($P<0.05$). 20% of examined control cheese samples contained Coagulase positive *Staphylococcus aureus* while none of kariesh cheese with probiotics was contaminated by such organism. *Yersinia enterocolitica* failed to be detected in neither control samples nor Kariesh samples with probiotics. The obtained results indicated that kariesh cheese is of inferior quality and hazardous food, as it is considered as source of food borne illness and addition of probiotics leads to significance decrease in total bacterial and staphylococcus counts also coagulase positive *Staphylococcus aureus* failed to be detected in kariesh samples with probiotics. So there is a great need for rising up, developing and spreading the use of probiotics where kariesh cheese is made as a natural and gentle way to preserve food, with a view to improving the quality, hygiene and safety of food.

[Esmat, I. awad., Marwa, A. Ibrahim and Manal. I. El-Shaer. **Influence of probiotic on microbiological quality of kariesh cheese.** *Life Sci J* 2014;11(1):394-397] (ISSN: 1097-8135). http://www.lifesciencesite.com. 60

Keywords: Probiotics; kariesh cheese; microorganisms

1.Introduction

Kariesh cheese is one of the most popular local types of fresh soft cheese of the Egyptian cities. The increasing demand for it by Egyptian consumers is mainly attributed to its high protein content and low price. Kariesh cheese is made from defatted or skims cow or buffalo milk, or a mixture of both. Cow or buffalo are milked directly into special earthenware pots, which are kept undisturbed in a suitable place to allow the fat to rise to the surface forming a cream layer and the partly skimmed milk sours and clots. Then the cream layer is removed, and the curd is poured onto a mat which is tied and hung with its contents, to allow the whey drain until the desired texture of the cheese is obtained. Finally, the cheese is cut into suitable pieces, and salted then it is ready to be consumed as fresh cheese. The shelf-life of the fresh cheese is between one and two weeks (Abou-Donia, 1984. and Aldo, *et al.,* 2013). It is generally made of raw skimmed buffaloes' or cow's milk which is often of poor bacteriological quality owing to the high microbial load present in raw milk and the objectionable condition under which it was produced (Salwa *et al.,* 2012). Kariesh cheese contains amount of sugar, some water, soluble vitamins and most of calcium and phosphorus. The quality and composition of kariesh cheese may vary considerably due to such factors as the quality and composition of the clotted skimmed milk, the method of manufacture, the time required to complete the whey drain, the quality of salt added and the method of handling finished cheese (Aldo *et al.,* 2013).

A recent trend in cheese manufacture is production of natural flavoured cheese made in short time with highly nutritive value and good microbiological quality as for human consumption. (Hosny *et al.,* 2011) Kariesh cheese is sold uncovered and without container where the risk of contamination

is high so it is considered as a good medium for the growth of different types of spoilage and pathogenic microorganisms.(Dawood *et al.,* 2006).

Probiotic may reduce the incidence of disease or decrease the severity of disease outbreaks. Probiotics are defined as live microorganisms that administered in adequate amount produce a health benefits to the host. (Reid *et al.,* 2003). The mechanism used include the production of inhibitory substances against pathogens, competition for essential nutrients and adhesion sites, the supply of essential nutrients and enzymes and the modulation of interaction with the environment and the development of beneficial immune response.(Gomez and Balcazar, 2008). Several visions have been proposed to improve kariesh cheese including therapeutic or probiotic effects of kariesh cheese by using cultures containing Bifidobacterium sp. (Abd-Elhamid, 2012). Recently there has been significant commercial interest in using lactic acid bacteria and propionibacteria as natural food preservative to enhance food safety and stability as anti microbial systems possessed by these bacteria offer potential food effective natural preservation methods. (Effat, 2000). Lactobacilli have the longest history as biotherapeutic agents (probiotics) and are still the most common ingredients among those intended for consumption by farm animals. This choice of probiotic bacteria seems appropriate because the normal gastrointestinal microbiota of these animals is particularly rich in lactobacilli (Tannock, 1997).

2. Material and Methods

Materials:

1- Milk samples

Thirty random milk samples were collected from dairy farms at Sharkia Governorate; each sample was divided into two parts.

2-Probiotic strains

Two probiotic strains *Lactobacillus casei* (ATCC-334) and *Lactobacillus plantarum* (ATCC-8014).

Methods:

1- Culture preparation:

The two probiotic strains *Lactobacillus casei* (ATCC- 334) and *Lactobacillus plantarum* (ATCC-8014) were grown in MRS broth (biolife) at 30^0C for 48hours in anaerobic condition(Zambou *et al.,* 2004)

2- Kariesh cheese making

The first part was used as control while the second one was inoculated with active culture of two probiotic strains in ratio 2% of milk. Both parts were manufactured into Kariesh cheese according to method adopted by Fahmi, (1960).

3- Cheese analysis

25 gram cheese was homogenized for one minute with 225ml sodium citrate (2%). Total plate count on plate count agar (Oxoid), Total *Coliform* on violet red bile agar (Oxoid), *Staphylococcus aureus* on Baired Parker medium (Oxoid) and *Yersinia enterocolitica* on CIN medium (Oxoid). Methods of microbiological analysis were conducted according to APHA, (1992).

3. Results and Discussion

Concerning to total colony count listed in table (1) out of examined control kariesh cheese samples 30(100%) were showed bacterial growth, the minimum was 1.0×10^8cfu/gm, the maximum was 8.8×10^{10} cfu/gm and the mean value was $7.86\times10^9\pm3.05\times10^9$ cfu/gm. While fresh kariesh cheese with probiotics showed that count was ranged from 9.0×10^5 to 3.0×10^7 with a mean value of $6.42\times10^6\pm1.29\times10^6$. There was significance difference between two groups ($P < 0.05$). These results were in agreement with that reported by Abou Dawood *et al.* (2005) and Hosny *et al.* (2011).

Table (2) showed that all examined control kariesh cheese samples were contaminated with *Coliforms* while only 24(80%) of examined kariesh samples with probiotics were harbor *Coliforms* and the count were ranged from 2.0×10^3 and 2.0×10^3 to 1.9×10^5 and 1.2×10^5 with a mean value of $4.66\times10^4\pm9.3\times10^3$ and $1.47\times10^4\pm4.6\times10^3$ for control samples and samples with probiotics respectively Slightly higher counts were reported by Hosny *et al.* (2011) while Ahmed, (1988) recorded nearly similar counts. There was no difference between two groups ($P > 0.05$). *E. coli* can become acid resistant and survive acid extremes similar to the human gastric barrier (Benjamin and Datta, 1995). Sindt*, et al. (2002)* concluded that *E. coli* and total *coliforms* provides inclusive information that encompasses several pathogenic strains of bacteria and is an appropriate model to determine reduction strategies for controlling acid-resistant, pathogenic, enteric bacteria.

Table (3) summarized *Staphylococcus aureus* count in control and kariesh samples with probiotics. All control kariesh cheese samples were positive for *Staphylococcus aureus*, the count was ranged from 1.0×10^4 to 2.9×10^7 and the mean value was $4.2\times10^6\pm1.2\times10^6$. Out of examined kariesh cheese samples with probiotics 21(70%) were contaminated with *Staphylococcus aureus*, The high level of contamination was 8.0×10^5 , the low level was 2.0×10^3 and the mean value was $9.4\times10^4\pm3.7\times10^4$. There was significance difference between two group ($P<0.05$).

These findings were nearly similar to those reported by Abou Dawood *et al.* (2005) while Kaldes, (1997) and Hosny *et al.* (2011) reported lower counts.

Table (1): Total colony count of kariesh cheese samples (n=30).

	Total colony count				
	Positive samples		Minimum	maximum	Mean ±SEM
	NO	%			
Control group	30	100%	1.0×10^{8}	8.8×10^{10}	$7.86\times10^{9}\pm3.05\times10^{9}$
Fresh kariesh cheese with probiotics	30	100%	9.0×10^{5}	3.0×10^{7}	$6.42\times10^{6}\pm1.29\times10^{6}$

Table (2): Coliforms count of Kariesh cheese samples (n=30).

	Coliforms count				
	Positive samples		Minimum	Maximum	Mean ±SEM
	NO	%			
Control group	30	100%	2.0×10^{3}	1.9×10^{5}	$4.66\times10^{4}\pm9.3\times10^{3}$
Fresh kariesh cheese with probiotics	24	80%	2.0×10^{3}	1.2×10^{5}	$1.47\times10^{4}\pm4.6\times10^{3}$

Table (3): Staphylococcus aureus count of kariesh cheese samples (n=30).

	Staphylococcus aureus count				
	Positive samples		Minimum	Maximum	Mean ±SEM
	NO	%			
Control group	30	100%	1.0×10^{4}	2.9×10^{7}	$4.2\times10^{6}\pm1.2\times10^{6}$
Fresh kariesh cheese with probiotics	21	70%	2.0×10^{3}	8.0×10^{5}	$9.4\times10^{4}\pm3.7\times10^{4}$

Table (4): Incidence of coagulase positive Staphylococcus aureus in examined kariesh cheese samples (n=30).

	Coagulase positive Staph. aureus	
	Positive samples	
	NO	%
Control group	6.0	20.0%
Fresh kariesh cheese with probiotics	0.0	0.0%

The results presented in table (4) stated that 20% of examined control cheese samples contained Coagulase positive *Staphylococcus aureus* while none of kariesh cheese with probiotics was contaminated by such organism. *Yersinia enterocolitica* failed to be detected in neither control samples nor Kariesh samples with probiotics. El-Gamal *et al.* (2013) failed to detect Yersinia enterocolitica from examined cheese and yoghurt samples. The Egyptian standards for kariesh cheese did not mention an account on the acceptable level of total bacterial count but indicated that *Coliforms* are less than 10cfu/gm and absence of *coagulase positive Staphylococcus aureus* .*Lactobacillus spp.* showed a broad inhibitory spectrum against *Staphylococcus aureus* and *E. coli* and the inhibitory substance of certain lactobacilli were bacteriocin and lactic acid. (Ryan *et al.,* 2008 and Amin *et al.,* 2009). The obtained results indicated that kariesh cheese is of inferior quality and hazardous food, as it is considered as source of food borne illness and addition of probiotics leads to significance decrease in total bacterial and staphylococcus counts also coagulase positive *Staphylococcus aureus* failed to be detected in kariesh samples with probiotics. So there is a great need for rising up, developing and spreading the use of probiotics where kariesh cheese is made as a natural and gentle way to preserve food, with a view to improving the quality, hygiene and safety of food.

References:

1. Abd-Elhamid, A.M. (2012). Production of functional Karish cheese by microencapsulation of *Bifidobacterium adolescentis* ATCC 15704. Advance J. Food Sci. Technol., 4 (2): 112-117.
2. Abou-Dawood A.I., Taha S.H., Mohamed M.A. (2005). Chemical and microbiological quality of raw milk, soft and hard cheeses collected from some districts at Giza government. Egypt. J. Dairy Sci., 33, 201–214.
3. Abou-Donia, S. A. (1984). Egyptian fresh fermented milk products. New Zealand Journal of Dairy Science and Technology, Vol. 19, 7-18.
4. Ahmed, A.A.H.(1988).Behavior of virulent Yersinia enterocolitica in Domiati cheese .Assiut Vet. Med., 22:81.
5. Aldo T., Fady A. A. and Ola A.H. O. (2013). History, Processing and Quality Enhancement of Traditional Egyptian Kariesh Cheese: A Review. Food Science and Technology 1(1): 1-6, 2013.
6. Amin, M., M. Jorfi, A.D. Khosravi, A.R. S. and Sheikh, A. F. (2009). Isolation and identification of *Lactobacillus casei* and *Lactobacillus plantarum*

from plants by PCR and detection of their antibacterial activity. J. Biol. Sci., 9: 810-814.
7. APHA, American Public Health Association (1992). Standard Methods for the Examination of Dairy Products , Amer. Publ. Halth Assoc. Inc. 12th Ed., New York.
8. Benjamin, M. M., and A. R. Datta. (1995). Acid tolerance of enterohemorrhagic *Escherichia coli*. Appl. Environ. Microbiol. 61: 1669–1672.
9. Dawood, M.; Abdou, R. and Salem, G. (2006).Effect of irradiation of skim milk on the quality of
10. karish cheese. Egyptian Journal of Dairy Science 32(4) 500-509.
11. Effat B.A. (2000). Antifungal substances from some lactic acid bacteria and propionibacteria for use as food preservatives. J. Agric. Sci. Mansoura Univ., 25, 6291–6304.
12. Egyptian Standards for Kareish and Domiati Cheese (2000) Egyptian Organization for Standards and Quality Controls. ES:1008/2000.
13. El-Gamal, M. S., El Dairouty,R. K., Okda,A. Y., Sahar H. S. and El-Shamy,S. M.(2013). Incidence and Interrelation of *Cronobacter sakazakii* and Other Foodborne Bacteria in Some Milk Products and Infant Formula Milks in Cairo and Giza Area. World Applied Sciences Journal 26 (9): 1129-1141.
14. Fahmi, A. H. (**1960**). Kariesh cheese. J. Agric. Sci. **13**: 1.
15. Gomez G.D. and Balcázar, J. L. (2008). Areview on the interactions between gut microbiota and innate immunity of fish, FEMS Immunol. Med. Microbiol. 52: 145–154.
16. Hosny I.M., W.I. El Kholy, H.A. Murad and El Dairouty, R. K. (**2011**). Antimicrobial activity of Curcumin upon pathogenic microorganisms during manufacture and storage of a novel style cheese (karishcum). Journal of American sci., 2011; 7(5).
17. Kaldes Y.Th. (1997). Microbiological examination of soft cheeses manufactured in Minia city. Assiut Vet. Med. J., 38, 39–46.
18. Salwa, A. A.,Farag, D. E. and Galal, E. (2012). Effect of Gamma Irradiation on the Quality and Safety of Egyptian Karish Cheese. Journal of American Science 2012; 8(10).
19. Sindt, J. J., Drouillard,J.S., Thippareddi, H., Phebus, R. K., Lambert,D. L., Montgomery, S. P., Farran, T. B., LaBrune, H. J., Higgins, J. J. and Ethington, R. T.(2002). Evaluation of finishing performance, carcass characteristics, acid-resistant E. coli and total coliforms from steers fed combinations of wet corn gluten feed and steam-flaked corn**.** J ANIM SCI**.,** 80 (12) 3328-3335.
20. Reid, G. Reid, M. E. Sanders, H. R. Gaskins, G.R. Gibson, A. Mercenier, R. A. Rastall, M. B. Roberfroid, I. Rowland, C. Cherbut and Klaenhammer, T. R. (2003). Scientific paradigms for probiotics and prebiotics, J. Clin. Gastroenterol. 37:105–118.
21. Ryan, K.A., T. Jayaraman, P. Daly, C. Canchaya , S. Curran , F. Fang , E.M. Quigley and O'Toole, P. W. (2008). Isolation of lactobacilli with probiotics properties from the human stomach. Letters in Applied Microbiol., 47 (4): 269-274.
22. Tannock, G. W. (1997). Probiotic properties of lactic acid bacteria: plenty of scope for fundamental R& D. Trends in biotechnology , 15, 270- 274.
23. Zambou, N. F.,Nour El-Noda, A., Mbiapo, T. F. and Morsi, E. (2004). Effect of ropy and capsular exopolysaccharides producing strain of *Lactobacillus plantarum* 162RM on characteristics and functionality of fermented milk and soft Kareish type cheese. African Journal of Biotechnology Vol. 3 (10), pp. 512-518.

1/11/2014