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EFFECT OF BUFFALO AND COW SKIM MILK FLOURIDIZATION ON 2. SOME CHARACTERISTICS OF CASEIN MICELLES, PROTEOLYTIC DIGESTIBILITY AND MICROBIOLOGICAL PROPERTIES

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ABSTRACT

Fluoridated buffalo's and cow's skim milk with different concentration of sodium fluoride NaF (5 – 20 p.p.m. F) and their effect on the characteristics of casein micelles, proteolytic digestibility and microbiological properties were investigated. Added NaF reduced the weight and size of casein micelles and this effect increased by increasing the concentration of NaF. After cold storage (5ºC±1/48 hrs) the reduction of weight and size of casein micelles of cow's skim milk contained 20 p.p.m. F was markedly higher than that of buffaloe's skim milk. Increasing the concentration of NaF significantly caused increase ($P \leq 0.05$) in the rate of proteolytic digestion of buffalo's milk samples by trypsin and commercial (Alphintern®) enzymes compared with cow's ones. Growth of lactic acid cultures and pathogenic bacteria in buffalo's and cow's skim milk did was not affected by fluoride supplementation.

Key words : *buffalo's skim milk; casein micelles, lactic acid cultures NaF; pathogenic bacteria , proteolytic digestion, weight and size,*

1. INTRODUCTION

The element fluorine occurs widely as fluoride. Many studies in different parts of the world have confirmed the close relationship between fluoride and dental caries. Water and food are the main source of fluoride absorbed by the body. (American Academy of Pediatrics,1979, 1986, El–awamry *et al.,* 1989 and El–Gabry and Darwish , 2003).

Addition of fluoride to centralized water supplies is an effective method for improving the dental health of a population. However, benefits of water fluoridation are limited by the lack of centralized water supplies in much of the world and difficulties in controlling dosages for infants and children. The benefits of dietary fluoride have been demonstrated only for infants and children up to the age of 12 (Al–Khateeb *et al*., 1991, Levy *et al.,* 1995). Milk is a normal component of the diet of this age group. However, not milk but also dairy products are poor sources of fluoride (Garrec and Plebin, 1986 and Hassan *et al.,* 1999). The supplementation of milk with fluoride is an effective alternative to water fluoridation (Davis, 1979; Pakhomoy, 1996, Banoczy, 1997 and Bian *et al*., 2004). The advantages of fluoridated milk include better dosage control and consumption of other nutrients in milk (*e.g.*, calcium, phosphorus and vitamin D) that are necessary for optimum bone and tooth development (Davis, 1975a,b and White 1987). The fluoride content of milk may vary according to the fluoride content of animal feed, species of milk, drinking water, the surrounding environment (*e.g.* high fluoride content of the atmosphere due to emissions from aluminium smelters). Processing of milk and milk products and supplementation or fortification of milk with fluoride were reported by some authors (Junkkarinen and Kreula, 1976, Maylan and Krook, 1982, Shehata *et al.,* 1985, Gerrec and Plebin, 1986 and Wheeler *et al.,*

1988a,b). Fluoride in milk exists in two chemical forms: as ionic or free fluoride and bound fluoride. Ionic fluoride refers to fluoride ion still in solution, while bound fluoride refers to the fluoride that has formed a complex with other elements or compounds (calcium or protein for example). In milk, the casein is present primary in the form of colloidal particles (calcium caseinate phosphate complex) ~50- 600 nm in size known as casein micelles. Much researches have focused on the effect of processing conditions such as temperature, pH, ionic strength, addition or removal of minerals and concentration of salt on the molecular weight and size of aggregates (Grufferty and Fox, 1985, Udabage *et al.,* 2000; Anema and Li 2003, Philippe *et al*., 2005 and Huppertz and Fox, 2006).

The proteolytic digestibility of milk protein is a complex phenomenon that involves significant changes in the structure, function and application of peptides derived from milk protein. These have been extensively studied (Diaz *et al.,* 1996 and Kim *et al.,* 2007). The casein phosphopeptides (CPP) derived from milk caseins by tryptic digestibility associated with amorphous calcium phosphate (ACP) forming stable complexes. These peptides (CPPACP) are able to incorphour fluoride inos. These complexes designated CPPACFP, have the potential to provide superior clinical efficacy in preventing dental caries (Cross *et al*., 2004 and Kecik *et a*l. , 2008).

Microorganisms vary as regards their tolerance to salt as well as their power of adaptability to salt tolerances. Previous studies have been carried out on the effect of salts on the growth of various microorganisms that are of important to dairy and public health (Perfileve *et al*. 1982, El–Gazzar and Marth, 1991 and Shehata *et al.,* 1997).

However, no cited studies were reported on characteristics of casein micelles, proteolytic enzyme and microbiological evaluation of fluoridated and contaminated buffaloe's skim milk with fluorine. Therefore, the objective of the present study was to evaluate the common levels existed (5-20 ppm Fó) in fluoridated or contaminated buffaloe's skim milk on weight, size of casein micelles, proteolytic digestion and growth of lactic acid cultures and pathogenic bacteria compared with those of cow's skim milk.

2. MATERIAIS AND METHODS

2.1. Materials

2.1.1. Milk samples

Fresh raw bulk buffalo's and cow's milk was obtained separately from the hards of the Fac. Agric., Cairo Univ. Milk samples were separated twice using a cream separator. The residual fat was removed by centrifugation at 3000 rpm for 15 min. at 5ºC.

2.1.2. Sodium fluoride solution (NaF)

Analytical grade sodium fluoride (Sigma Chemical Co.) was dried and 2.210 g weight dissolved in distilled deionized water and made to 1 L(1000 mg Fó /L).

2.1.3. Enzymes

2.1.3.1. Trypsin

Pancreatic trypsin (3.0 Anson trypsin unit) was obtained from Nova Nordisk A/ S Denmark .

2.1.3.2. Pepsin

Pepsin cryst (100 mμ/mg) was obtained from Merck, Germany.

2.1.3.3. Alphintern®, (commercial Enzyme)

Each table contained chymotrypin (300E.A.U) and trypsin (300E.A.U) was obtained from Amoon Pharmacea Trial Industries Co., Cairo, Egypt; under license from Lab Leuroquin France.

2.1.4. Microorganisms

2.1.4.1. Lactic acid cultures

Yoghurt culture *Lactobacillus acidophilus, Bifido bacterium, Lactobacillus casei* and *Lactobacillus para casei* were obtained from the Dairy Sci. and Technology Dept., Fac. Agric., Cairo Univ**.**

2.1.4.2. Pathogenic microorganisms

Gram negative(*Escherichia coli* and *Salamonella typhinurium*)and gram positive (*Staphylococeus aureus* and *Listeria monacytogenes*) were obtained from the Dept. of Agriculture Microbiology Fac. Agric., Cairo Univ.

2.1.5. Preparation of fluoridated milk

Buffalo's, cow's skim milk and mixture of the two (1:1 v/v) of known added fluoride concentrations (5,10,15 and 20 ppm Fó) were prepared by the addition of appropriate volumes of aqueous sodium fluoride (NaF

1000 ppm F ó) to known volumes of milk with stirring and were left standing for one hour to equilibrate at room temperature 25ºC. The same procedure was used for preparation fluoridated reconstituted skim milk powder (10 %). The control samples were perpetrated without adding NaF.

2.2. Methods of analysis

2.2.1. Determination of weight and size

For measuring the average weight and size of the casein particles, the method of Dyatchenko and Voldavento (1953) was followed, The optical density was measured using a Unicam 8625 UV/VIS spectrophotometer (Ati Laicam, England) and read at 545 nm. The casein content of the milk samples was determinated by the Kjeldahl method as modified by Rowland (1938).

2.2.2.Determination of proteolytic digestibility

The proteolytie digestibility of milk samples was carried out with the use of trypsin, pepsin and Alphintern® enzymes as described by Datta Roy (1981&1982). The optical density of α-amino nitrogen at 575 nm was estimated using a Unicam 8625 UV/VIS spectrophotometer (Ati Laicam, England) according to the method of Lee and Takahashi (1966).

2.2.3. Microbiological analysis

Growth rates of fermented cultures and pathogenic organisms were determined in sterilized fluoridated buffalo's and cow's skim milk (5 to 20 ppm F) at 121ºC/5 min. The lactic organisms were inoculated at a concentration of 1.0 % while pathogenic organisms at a concentration of approximaately 10.000 cfu/ml. The inoculated milk was incubated without agitation at 37ºC for all lactic acid bacteria except yoghurt culture at 40ºC for 4 to 16 hrs. The samples of pathogenic organisms were incubated at 32ºC to 37ºC for 1 to 4 days.

2.2.4. Statistical analysis

Experiments were repeated in triplicates and each analysis in triplicates. Data were analyzed statistically using the general modal of SAS (Statistical analysis system, 2000) at 5 % level of significance.

3. RESULTS AND DISCUSSION

3.1. Weight and size of casein micelles

Table (1) reveals the weight and size of casein micelles of different milk species containing NaF up to 20 p.p.m Fó. In control samples, reconstituted cow's skim milk had higher weight and size than other milk samples. This could be attributable to attachment of denatured whey proteins onto the outside K-casein layer of the casein micelles, increasing their hydroyonmic size (Oldfield *et al*., 2005 and Martin *et al*., 2007). However adding up to 20 p.p.m. F as NaF progressively reduced the weight and size of casein micelles of different treated milk samples.

The obtained data show that there was no significant effect of various concentrations of NaFó (5-20 p.p. F) on the weight of casein micelles while there are significant differences between milk species. The reduction of weight was in ordered reconstituted skim milk powder > buffalo's and cow's skim milk 1: 1 > cow's skim milk > buffalo's skim milk.

As regards the size, it is clear that the casein micelles size of all samples progressively decreased with increasing of NaF concentration. The analysis of variance showed that there were significant differences ($P \leq 0.05$) among the concentrations of NaF added and milk species. The previous studies showed that changes in the weight and size of casein micelles were related to many factors such as composition of milk, stage of lactation, seasonal variation, individuality, pH and salt balance (Abd El-Salam *et a.l*., 1978, Ismail *et a.l*., 1978, Salama *et a.l*., 1978a,b Youseff *et al*., 1983 a,b and Salam *et al*., 1983).

The casein micelles weight and size of buffaloe's and cow's skim milk containing Fó as NaF (5–20 p.p.m) during storage at 5±1ºC for 24-48 hrs are shown in Table (2). Supplementation of Fó up to 20 p.p.m. progressively decreased the weight and size of casein micelles during storage at 5±1ºC for 24 hrs and lowered by increasing storage period from 24 to 48 hrs. The changes in the weight and size of casein micelles could be related to many factors such as added fluoride forms a reversible ionic complex with casein micelles (Beddows, 1982 and Beddows and Blake, 1982). The reversibility of the casein micelles structure and salt

Table (1): Effect of milk fluoridization (5-20 ppm F as Na F ó) on the weight and size of casein micelles.

Type of milk	Weight (million units)						Size (nm)					
	Concentration of F (p.p.m.)						Concentration of F ó) (p.p.m.)					
	Control	5	10	15	20	Mean	Control	5	10	15	20	Mean
Buffalo's	640.25	638.55	629.07	625.28	619.34	630.09[b]	85.91[C]	85.47[D]	84.42[E]	83.89[F]	83.06[H]	84.55[a]
Cow's	522.63	513.95	509.56	507.06	505.05	511.65[d]	70.17[K]	68.94[L]	68.45[M]	68.03[N]	67.74[O]	68.66[c]
B:C (1:1)	649.91	630.11	628.76	622.42	620.41	630.32[a]	87.20[B]	84.54[E]	84.41[E]	83.46[G]	83.23[H]	84.56[a]
RSP	655.08	617.02	615.51	593.36	591.27	614.45[c]	87.92[A]	82.80[I]	82.59[I]	79.92[J]	79.32[J]	82.39[b]
Mean	616.97	599.41	595.72	587.03	584.01		82.80[a]	80.44[b]	79.97[C]	78.67[d]	78.343[e]	
S.E.	20.6837	18.8378	18.9172	18.0772	17.7889		2.7692	2.535	2.5287	2.4205	2.3867	

RSP= Reconstituted skim milk powder.
Different superscripts (A,B,C, a,b,c,…) at the same column are significantly different (P≤ 0.05).
S.E = Standard Error.

balance change was produced by cooling at 4ºC, (Dalgleish and Law 1989, and Udabage *et a.l,* 2000). Also, loss of free fluoride in stored milk is a result of its binding to Ca^{++} released from the protein (Weiczorek *et al.,* 1992). Furthermore, ionic strength increase (*i.e.* on adding NaF) leads to a reduction in the ionic activity of calcium phosphate which causes an increase in solubility and dissociation of calcium phosphate (Walstra 2003).

Statistical analysis in Table (2) reveals that there were significant variations between concentrations of added NaF and species of milk. Cow's skim milk samples were higher with significant effect (P≤ .05) than those of buffalo's samples. The reductions of weight and size of cow's milk containing 20 p.p.m. F as NaF was 7.2 % compared with 2.6% in buffalo's samples.

3.2. Proteolytic digestibility enzymes

Comparison of digestibility rate by different enzymes of buffalo's and cow's skim milk containing 5-20 p.p.m. F^- as NaF is given in Table (3). The rate of proteolysis is known to depend on the enzyme specifically and the conformation of proteins. Trypsin specifically attacks the carboxy group of lysine and arginine while pepsin specifically attacks the carboxy group of tryptophan, phenylalanine, tyrosine, methionine and leucine. The changes in conformation of milk proteins alter the number of accessible peptide bonds after the rate of proteolysis (Kim *et al*., 2007 and Qi *et al*., 2007). The results show that the addition of NaF enhanced the rate of digestibility by trypsin and commercial enzymes of buffalo's and cow's skim milk at any concentration of NaF (5-20 ppm F as NaF) and incubation time (30-60 min.). It also revealed that the significant effect of NaF was more clear with buffalo samples than those of cows. Furthermore, the presence of NaF did not improve the rate of peptic digestibility of buffalo and cow samples. The significant differences in the relative digestibility of milk samples attributable to interactions between milk proteins and NaF may affect the backbone conformations of the protein and the amino acids (AA) between inside and outside of the protein. Therefore, the small amount of NaF present can probably allow the better exposed functional groups of the AA side chains to enzyme attack. Datta Roy (1981, 1982) showed that heating and freezing bring about a drastic changes in the three–dimensional conformation of the milk proteins so as to expose the carboxy group of lysine, arginine and tryptophan, phenylalanine tyrosine methionine for trypsin and pepsin to attack respectively of the milk species (cow, buffalo and goat).

Statistical analysis of these data (Table 3) support the view that the rate of commercial enzyme digestibility of buffaloe skim milk supplemented with NaF was significantly higher ($P \leq 0.05$) than other enzymes.

Therefore, It is evident, that NaF plays a role in the structural organizations of milk protein, which affect the enzymatic liberation of amino acids.

3.3. Microbiological properties

Data collected during the growth of pure cultures in fluoridated buffalo and cow skim milk samples (5-20 ppm Fó as NaF) show that the microbial activity in fluoridated milk was similar to that in normal milk samples (Table4,5).However, acid production by any of the starter culture organisms tested. Nearly similar findings were reported by (Frank and Christein, 1985, Ivanova *et al*. 1989). Also, in this respect, Rusoff and Koniknoff (1975) reported that fluoridated milk containing the proper levels of fluoride can be used in cheese making without affecting the cheese culture, and no differences were detected in flavor, color or taste. While, Assali and White (1985) evaluated the effect of either 4 or 40 ppm sodium fluoride on fermented dairy products (butter milk, yoghurt, cottage cheese) and stated that the higher concentration of fluoride significantly retarded acid production in the starter cultures and adversely affected the quality of the fermented production. On the other hand, previous reports showed that the rate of growth of lactic acid, probiotic and pathogenic bacteria was affected by othersalts (Minor and Marth 1972, Shehata *et al*., 1995 and Mohamoud 1997). Hussein (2004) indicated that replacement of sodium chloride with potassium cholride improved the viability of either free cells or encapsulated bifidobacteria.

Table (2): Effect of milk fluoridization (5-20 p.p.m. F^{-}) on weight and size of casein micelles during storage at 5±1ºC.

Concentration of F (p.p.m.)	Weight (million units)			Size (nm)		
	BSM	CSM	Mean	BSM	CSM	Mean
	At zero time					
Control	640.24 ^{A}	522.63 ^{N}	581.43 ^{a}	85.91 ^{A}	70.17 ^{L}	78.04 ^{a}
5	636.55 ^{C}	513.92 ^{O}	575.24 ^{b}	85.47 ^{C}	68.93 ^{M}	77.20 ^{b}
10	628.07 ^{G}	509.06 ^{P}	568.81 ^{c}	84.42 ^{G}	68.45 ^{N}	76.43 ^{d}
15	625.27 ^{J}	807.06 ^{G}	566.17 ^{d}	83.89 ^{I}	68.02 ^{O}	75.95 ^{d}
20	619.34 ^{L}	505.05 ^{R}	562.19 ^{f}	83.06 ^{K}	67.74 ^{P}	75.40 ^{f}
Mean	629.89 ^{b}	462.86 ^{d}		84.54 ^{b}	61.304 ^{d}	
	After 24 h					
Control	639.31 ^{B}	490.39 ^{S}	564.85 ^{e}	85.73 ^{B}	65.84 ^{Q}	75.78 ^{e}
5	632.24 ^{E}	444.37 ^{T}	538.31 ^{g}	84.85 ^{E}	59.22 ^{P}	72.03 ^{g}
10	631.17 ^{F}	435.47 ^{u}	533.52 ^{h}	84.69 ^{F}	58.42 ^{S}	71.55 ^{h}
15	627.25 ^{H}	431.82 ^{v}	529.53 ^{j}	84.15 ^{H}	57.92 ^{T}	71.03 ^{j}
20	626.20 ^{H}	426.15 ^{x}	526.18 ^{l}	84.06 ^{H}	57.20 ^{V}	70.63 ^{k}
Mean	631.23 ^{a}	445.64 ^{e}		84.64 ^{a}	59.72 ^{e}	
	After 48 h					
Control	635.02 ^{D}	429.37 ^{W}	532.19 ^{i}	85.21 ^{D}	57.60 ^{u}	71.40 ^{i}
5	627.87 ^{HG}	425.79 ^{X}	526.83 ^{k}	84.22 ^{H}	57.12 ^{v}	70.67 ^{k}
10	623.34 ^{K}	424.71 ^{Y}	524.83 ^{m}	83.63 ^{J}	57.28 ^{v}	70.45 ^{l}
15	619.06 ^{L}	413.88 ^{Z}	516.47 ^{n}	83.04 ^{K}	55.51 ^{w}	69.28 ^{m}
20	617.74 ^{M}	407.08 ^{A}	512.41 ^{n}	82.91 ^{K}	54.62 ^{x}	68.76 ^{n}
Mean	624.61 ^{C}	420.17 ^{f}	522.38 ^{c}	83.80 ^{C}	56.43 ^{f}	70.12c
General mean	628.58 ^{a}	459.15 ^{b}		84.35 ^{a}	61.60 ^{b}	
S.E.	1.2844	7.6578		0.1731	1.0287	

BSM= Buffalo's skim milk.
CSM= cow's skim milk.
Different superscripts (A,B,C, a,b,c,…) at the same column are significantly different ($P \leq 0.05$).
S.E. = Standard Error.

Table (3) : Effect of milk fluoridization (5-20 p.p.m. F^{-}) on the rate of proteolytic digestibity with different enzymes*.

F^{-} as (p.p.m).	Trypsin			Pepsin			Alphintern® (commercial enzyme)		
	BSM	**CSM**	**Mean**	**BSM**	**CSM**	**Mean**	BSM	CSM	Mean
	At zero time								
Control	0.3800^{T}	0.6235^{P}	0.5017^{f}	0.3800^{A}	0.6235^{S}	0.5017^{n}	0.3800Y	0.6235^{U}	0.5017^{m}
5	0.4200^{S}	0.6650^{O}	0.5425^{k}	0.42000^{Z}	0.6650^{Q}	0.5425^{m}	0.4200^{X}	0.6650^{T}	0.5425^{l}
10	0.4300^{S}	0.6790^{NO}	0.5520^{k}	0.4300^{X}	0.6790^{P}	0.5522^{l}	0.4300^{X}	0.6745^{T}	0.5522^{l}
15	0.48550^{R}	0.7055^{MN}	0.5805^{j}	0.4855^{T}	0.7055^{O}	0.5955^{k}	0.4855W	0.7055^{S}	0.5955^{k}
20	0.5655^{G}	0.7805^{L}	0.6730^{i}	0.5655^{F}	0.7805^{K}	0.6730^{l}	0.5655^{V}	0.7805^{R}	0.6730^{j}
Mean	0.4502^{f}	0.6920^{e}		0.4562^{f}	0.6920^{d}		0.4562^{f}	0.6920^{e}	
	At 30 min.								
Control	0.7290^{M}	0.8045^{L}	0.7667^{h}	0.524^{W}	0.7705^{L}	0.6472^{j}	0.9505^{G}	1.060^{O}	1.0052^{j}
5	0.8020^{L}	0.990^{H}	0.8960^{g}	0.532^{V}	0.8120^{S}	0.6720^{i}	0.9705^{P}	0.0825^{N}	1.0265^{hs}
10	0.9490^{JK}	1.0235^{H}	0.9862^{f}	0.552^{U}	0.8235^{I}	0.6877^{h}	1.1235^{M}	1.2225^{K}	1.1730^{g}
15	0.9690^{JI}	1.050^{EF}	1.0095^{f}	0.623^{S}	0.8435^{H}	0.7333^{g}	1.1720^{L}	1.249^{J}	1.2105^{f}
20	1.3430^{DE}	1.1015^{cq}	1.1177^{e}	0.711^{N}	0.8880^{F}	0.7995^{d}	1.4640^{E}	1.4315^{G}	1.4477^{c}
Mean	0.9166^{d}	0.9939^{c}		0.588^{f}	0.8275^{f}		1.1361^{d}	1.2091^{C}	
	At 60 min.								
Control	0.9295^{K}	1.074^{GH}	1.0017^{f}	0.6215^{S}	0.8525^{G}	0.7370^{f}	1.3390^{I}	1.4470^{F}	1.3932^{e}
5	1.0425^{GH}	1.2610^{C}	1.1517^{d}	0.6580^{R}	0.8940^{E}	0.7760^{e}	1.3510^{I}	1.4680^{E}	1.4095^{d}
10	1.1615^{D}	1.2610^{C}	1.2112^{c}	0.7540^{M}	0.9130^{D}	0.8337^{c}	1.3860^{H}	1.4945^{D}	1.4403^{c}
15	1.2345^{C}	1.3315^{B}	1.2830^{b}	0.8095^{J}	0.9295^{C}	0.8695^{b}	1.4540^{EF}	1.5735^{C}	1.5137^{b}
20	1.4240^{A}	1.3490^{B}	1.3865^{a}	0.9420^{B}	1.0310^{A}	0.9865^{a}	1.9775 A	1.8225^{B}	1.8000^{a}
Mean	1.1584^{b}	1.2553^{a}	1.2068^{a}	0.7571^{C}	0.9240^{a}	0.8405^{a}	1.5015 b	1.5612A	1.5314^{a}
General mean	0.8417^{b}	0.9797^{a}		0.6005^{b}	0.8138^{a}		1.0312^{b}	1.1534^{s}	
S.E.	0.0598	0.0458		0.0279	0.0202		0.0870	0.0698	

* As optical density at 570 nm, BSM= Buffalo's skim milk . CSM= Cow's skim milk .

Different superscripts (A,B,C, a,b,c,…) at the same column are significantly different (P≤ 0.05).

S.E. = Standard Error.

Table (4): Effect of sodium fluoride supplementation of milk (5-20 p.p.m Fó) on the growth of some lactic acid bacteria.

Fó as (p.p.m.)	Incubation time (hr)	Yoghurt culture		*L. acidophilus*		*B. bacterium*		***L. casei***		***L. para casei***	
		BSM	**CSM**	**BSM**	**CSM**	**BSM**	**CSM**	**BSM**	**CSM**	**BSM**	**CSM**
Control	4	+	+	+	+	+	+	+	+	+	+
	16	+	+	+	+	+	+	+	+	+	+
5	4	+	+	-	-	-	-	+	-	+	-
	16	+	+	+	+	+	+	+	+	+	+
10	4	+	+	-	-	+	+	+	-	+	-
	16	+	+	-	-	+	+	+	-	+	-
15	4	+	+	+	+	+	+	+	+	+	+
	16	+	+	-	-	-	+	-	-	-	-
20	4	+	+	-	-	-	+	-	-	-	-
	16	+	+	+	+	+	+	+	+	+	+

Buffalo's skim milk.

CSM= Cow's skim milk.

- = No growth.

Table (5): Effect of sodium fluoride supplementation of milk (5-20 p.p.m. Fó) on the growth of some pathogenic bacteria.

Fó as (p.p.m.)	Species of milk	Gram negative bacteria		Gram positive bacteria		
		E. coli *	*Salmonella*** *typhinurium*	*Staphylococcus *aureus*	*Listeria **** *monocytogenes*	
Control	BSM	+++	+++	+	+++	(2)
	CSM	+	+	+	+++	(4)
5	BSM	+++	+++	+++	+++	(2)
	CSM	+	+	+	+++	(4)
10	BSM	+++	+++	+++	++	(4)
	CSM	+	+	+	+++	(4)
15	BSM	+++	++	+++	++	(4)
	CSM	+	+	–	+++	(4)
20	BSM	+++ (gas)	++	+++	++	(4)
	CSM	+ (gas)	+	+	+++	(4)

* = Growth after 1 day **= Growth after 4 day. *** = Growth after (2–4) day.

+++ = Strong growth ++ = Medium growth + = Fair growth - = No growth.

To conclude, the species of milk and the concentration of NaF will lead to differences in growth behavior of organisms studied and consequentially affected the microbiological quality of fluoridated milk. So, the ambient consideration of the healthy nutritional and technological characteristics make sodium fluoride supplementation a very attractive process in dairy industry.

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تأثير إضافة الفلوريد للبن الفرز الجاموسى والبقرى
ثانيا: على بعض خواص جسيمات الكازين والتحلل البروتيني والصفات الميكروبيولوجية

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ملخص

تناولت هذه الدراسة تأثير إضافة فلوريد الصوديوم بتركيزات مختلفة (5- 20 جزء في المليون) على وزن وحجم جسيمات الكازين ومعدل الهضم بالإنزيمات المختلفة – التربسين – البيسين – التربسين التجاري (Alphintern®) ونمو بعض بكتريا حامض اللاكتيك والبكتريا المرضية لكل من اللبن الفرز الجاموسي والبقري.

أوضحت الدراسة النتائج الاتية :

أدت إضافة فلوريد الصوديوم بالتركيزات المختلفة إلى كل من اللبن الفرز الجاموسي والبقري والخليط بنسبة 1 : 1 حجم / حجم واللبن الفرز المجفف المعاد تركيبه إلى حدوث انخفاض فى كل من وزن وحجم جسيمات الكازين – وكان هذا التاثير اكثر وضوحا فى اللبن الفرز المعاد تركيبه بالمقارنة بالألبان الاخري.

كان معدل الانخفاض فى وزن وحجم جسيمات كازين اللبن الفرز البقري المحتوي على 20 جزء في المليون فلوريد الصوديوم والمبرد على درجة حرارة 5ºم لمدة 48 ساعة اكثر من مثيله اللبن الفرز الجاموسي.

حدوث زيادة معنوية في معدل الهضم بإنزيمات التربسين والـ (Alphintern®) للبن الفرز الجاموسي مقارنة باللبن الفرز البقري بزيادة تركيزات فلوريد الصوديوم المضافة للبن .

لم يتأثر نمو بعض بكتيريا حامض اللاكتيك والبكتيريا المرضية بإضافة فلوريد الصوديوم بالتركيزات المختلفة لكل من اللبن الفرز الجاموسي والبقري.

يتضح وبصفة عامة أن إضافة فلوريد الصوديوم للبن يحدث تغيرات عديدة لها أهمية تكنولوجية وصحية.

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