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OCCURRENCE OF SOME ENTERIC PATHOGENS IN RAW MILK AND SOME DAIRY PRODUCTS

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SUMMARY

110 samples of dairy products were collected from different localities and farmer's houses (40 raw milk samples, 20 raw cream samples, 30 Kariesh cheese samples and 20 yoghurt samples). Samples were examined microbiologically for the presence of enteric pathogens. In raw milk, *E. coli*, Shigella dysenteriae, Salmonella enterica indiana gr. B (04, 12-H, Z-H21, 7), Yersinia enterocolitica and klebsilla pneumoniae could be isolated from 27.5%, 7.5%, 2.5%, 5% and 2.5% of examined samples respectively. In raw cream, *E. coli* and Shigella dysenteriae could be isolated from 15% and 5% of examined samples respectively. In Kariesh cheese, *E. coli*, Shigella dysenteriae, Salmonella enterica indiana, yersinia enterocolitica and Klebsilla pneumoniae could be isolated from 43.3%, 16.6%, 3.3%, 3.3% and 6.6% of examined samples respectively. For yoghurt, *E. coli* and Yersinia enterocolitica could be isolated from 20% and 10% of examined samples respectively. From different 25 tested strains, 17 strains (68%) were positive for production of enterotoxin (7 strains of E. coli, 3 strains of Shigella dysenteriae, 2 strains of Salmonella enterica indiana, 3 strains of Klebsilla pneumoniae and 2 strains of Yersinia enterocolitica).

The public health importance of isolated organism as well as recommendation for prevention of microbial contamination of dairy products were also discussed.

INTRODUCTION

Milk and dairy products are subjected to contamination by microorganisms from different sources during production, processing, handling, packaging and storage resulting in an inferior quality rendering the product un safe (De Buyres, et al., 2001). If the microorganisms are pathogenic, their association with our food supply is critical from a public health point of view. The nutritional attrib-

utes of milk which make it an important part of the human diet are the same component that support the growth of pathogen that have been associated with milk and dairy products which leading to food born illness (Bryan, 1983).

Enteric pathogens represent one of the most important bacterial groups in foods. Members belonging to this group can survive and multiply or establish in the gastrointestinal tract of human, animals and birds. Under this term "enteric pathogens" several species of virulent microorganisms, such as pathogenic Salmonella, Shigella, Yersinia, *E. coli* and others that can cause gastrointestinal infections are involved (Ray, 1996).

Recent reports indicated that Salmonella spp., *E. coli* are considered the most frequent pathogens (Mead et al., 1999; White, et al., 2002) and they are responsible for many outbreaks (IFST, 1998).

Yersinia enterocolitica is a well established pathogen of humans, causing acute gastroenteritis, enterocolitis and mesenteric lymphadenitis, as well as a variety of extraintestinal disorders (Bottone, 1999).

Milk born infections are exclusively associated with consumption of raw milk or raw milk products (Ryser, 2000). Although the microbial growth is controlled in yoghurt by low pH, high acidity and starter culture, enteric pathogen can grow and multiply in this conditions (Lederberg, 2000).

Therefore, the aim of this study was to determine the incidence of enteric pathogens in raw milk, kariesh cheese, raw cream and yoghurt samples to assess health risks for consumers.

MATERIAL AND METHODS

Samples collection:

(40 raw milk, 20 raw cream, 30 kariesh cheese, 20 yoghurt) were collected randomly from different localities and farmer's houses making such products in the villages. Collected samples were transported in sterile air tight Jar to the laboratory to be examined bacteriologically immediately .

Preparation of samples:

Samples were prepared following the procedures described by American Public Health Association (APHA, 1992).

Isolation and identification of enteric pathogens were adopted according to (Varmam and Evans, 1991):

A loopful from previously prepared samples was streaked on sorbitol MacConkey agar and incubated at 35°C for 24h. Suspected colonies were picked up, spread on nutrient agar slants, reincubated at 37°C for 24 h for further identification.

Serotyping of salmonella:

By slide agglutination tests with Salmonella anti-

sera, according to Kauffmann – White serotyping scheme (Popoff & Minor, 1997).

Enterotoxin assays:

The ability to produce enterotoxins was assayed by the infant mouse test (Robins Brown et al., 1993).

The representative isolates were prepared to be injected through the abdominal wall into the milk-filled stomach infant mice which were 2-4 days, 3 infant mice were injected by sterile saline were used as negative control, after 4 hours, the mice were slaughtered and the entire intestine was removed the intestine and the remaining body were weighted to calculate the ratio of intestine weight (Gut weight) to remaining body weight.

If the Gut weight > 0.085, the test considered for the enterotoxins
Body weight

RESULTS

Table (1): Incidence of enteric pathogens in examined samples.

Type of samples	No. of sample	E. coli		Shigella dysenteriae		Salmonella enterica indiana gr.B (04, 12-H,Z-H, 1,7)		Yersinia enterocolitica		Klebsilla pneumoniae	
		Positive samples		Positive samples		Positive samples		Positive samples		Positive samples	
		No.	%	No.	%	No.	%	No.	%	No.	%
Raw milk	40	11	27.5	3	7.5	1	2.5	2	5	1	2.5
Raw cream	20	3	15	1	5	-	-	-	-	-	-
Kariesh cheese	30	13	43.3	5	16.6	1	3.3	1	3.3	2	6.6
Yoghurt	20	4	20	-	-	-	-	2	10	-	-

Table (2): Results of suckling mice assay of enteric pathogens.

Enteric pathogens species	Number of examined isolates	Gut weight (GW)	Body weight (BW)	GW/BW (ratio)
E. coli (10)	3	0.0734	1.2384	0.059
	2	0.1541	1.6598	0.093
	4	0.1732	1.8193	0.095
	1	0.1794	1.8245	0.098
Shigella dysenteriae (5)	2	0.0654	1.3473	0.048
	3	0.1742	1.8167	0.096
Salmonella enterica indiana (2)	1	0.1569	1.6981	0.092
	1	0.1549	1.7223	0.090
Klebsilla pneumoniae (3)	1	0.1732	1.8245	0.098
	2	0.1586	1.8125	0.087
Yersinia enterocolitica (5)	1	0.1054	1.4015	0.075
	2	0.1562	1.5874	0.081
	2	0.1506	1,6589	0.091
Control (Sterilebroth)		0.1346	1.5811	0.085

DISCUSSION

Provision of milk and milk products of good hygienic quality is desirable from consumer health point of view. This is one reason why milk testing and quality control include hygiene and microbial qualities (Giangiacomo, 2000).

The results recorded in Table (1) showed that for raw milk samples *E. coli*, Shigella dysenteriae, Salmonella enterica indiana gr. B (04, 12-H, Z-H21,7), Yersinia enterocolitica and Klebsilla pneumoniae could be recovered from 27.5%, 7.5%, 2.5%, 5% and 2.5% of examined samples respectively. These results are nearly similar to that obtained by Sharma et al., (1995), Deeb (1996), Ghazal (1997), Villar et al., (1999), Rser et al., (2000), El-Kosi (2001) and Zelalem et al., (2006). For raw cream, *E. coli* and Shigella dysenteriae could be recovered from 15% and 5% of examined samples respectively. These results are nearly similar to that obtained by Nazem (1991), Mauro et al., (1998) and El-Kosi (2001).

E. coli, Shigella dysenteriae, Salmonella enterica indiana gr. B (04, 12-H, Z-H21, 7), Yersinia enterocolitica and klebsilla pneumoniae could be isolated from 43.3%, 16.6%, 3.3%, 3.3% and 6.6% of kariesh cheese samples respectively. Nearly findings were obtained by Moustafa (1990), Al-

Ashmawy et al. (1994), Aman (1994), Vivegmis et al., (1998), Richard (2000) and El-Kosi (2001). *E. coli*, and Yersinia enterocolitica could be isolated from 20% and 10% of examined yoghurt samples respectively. These results nearly similar to that obtained by Salinas et al., (1984), Deeb (1996).

This high level of contamination of milk and milk products might be due to initial contamination originating from the udder surface, washing water, milking utensils and materials used for filtering the milk (Zelalem et al., 2006). Enteric pathogen are originated primarily from the lower intestine of man and animals. Hence, the principles source of them is mainly their intestinal contents. So, food contaminated directly or indirectly with faecal material can be potentially hazardous to consumers (Omore et al. 2001). In Table (2), we found that from different 25 tested strains, 17 strains (68%) were positive for production of enterotoxin, (7 strains of *E. coli*, 3 strains of Shigella dysenteriae, 2 strains of Salmonella enterica indiana, 3 strains of Klebsilla pneumoniae and 2 strains of Yersinia enterocolitica). Nearly similar results have been reported by Pai and Mors (1978), Cigugliano et al., (1982), Olson et al., (1985) and Ansam (2003). Contamination of milk and dairy product with enterotoxigenic strains of entric pathogens is considered as a mirror of the degree of disregard of numerous rules during processing and marketing.

It causes serious hazards to consumer's health. As, *E. coli* produced a variety of clinical symptoms including mild to sever diarrhea, haemorrahagic colitis, Haemolytic uremic syndrome and thrombotic thromocytopenic purpura haemorrahagica (Steinhart et al., 1996).

Enterotoxigenic strains of Yersinia enterocolitica cause gastroenteritis, terminal ileitis, mesenteric Lymphadenitis, erythemanodosum arthritis and eye infection (Varnam and Evans, 1991 and Ackers, et al. 2000). Enterotoxin produced by Klebsiella species induce mucisal damage leading exposure of Jejunal mucosa to bacterial invasion (Olson, et al. 1985).

From the obtained results we can concluded that products are exposed to extreme contamination with enteric pathogens constituting a publich health hazards. These hazards can be preventable with application of certain hygienic measures, pasteurization of milk used in production of various dairy products, prevention of post pasteurization contamination and using milk of high good microbiological quality. The consumer should have a higher degree of awareness and educator, so that he would not buy any suspicious food product.

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تواجد الميكروبات المعوية الممرضة في اللبن وبعض منتجات الألبان

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نظراً للأهمية الكبرى لمجموعة الميكروبات المعوية الممرضة قد أجريت هذه الدراسة لتحديد مدى تلوث بعض منتجات الألبان التقليدية قد تم فحص ١١٠ عينة (٤٠ عينة لبن خام ، ٢٠ عينة قشدة خام ، ٣٠ عينة جبن قريش ، ٢٠ عينة زبادى). أوضحت النتائج بالنسبة للبن الخام تواجد ميكروبات إيشريشيا كولاى ، شيجلا ديسنتريا ، السالمونيلا انتريكا انديانا ، يرسينيا انتيروكوليتيكا ، كليبسيلا نيمونى بنسب ٢٧,٥% ، ٧,٥% ، ٢,٥% ، ٥% على التوالى.

أما فى عينات القشدة الخام تم عزل ميكروب إيشريشيا كولاى ، شيجلا ديسنتريا بنسب ١٥% ، ٥% على التوالى وبالنسبة لعينات الجبن القريش تم عزل ميكروب إيشريشيا كولاى ، شيجلا ديسنتريا ، سالمونيلا انتريكا انديانا ، يرسينيا انتيروكوليتكا ، كليبسيلا نيمونى بنسب ٤٣,٣% ، ١٦,٦% ، ٣,٣% ، ٣,٣% ، ٦,٦% على التوالى.

أما فى عينات الزبادى تم عزل ميكروب إيشريشيا كولاى ويرسينيا انتيروكوليتكا بنسب ٢٠% ، ١٠% على التوالى وتم اختيار ٢٥ عترة مختلفة لعمل اختبار الضراوة ووجد أن ١٧ عترة لها القدرة على إنتاج السموم منها ٧ عترات من الايشريشيا كولاى ، ٣ عترات من شيجلا ديسنتريا ، ٢ عترة من السالمونيلا انتريكا انديانا ، ٣ عترات من الكليبسيلا نيمونى ، ٢ عترة من يرسينيا انتيروكوليتكا.

هذه النتائج تبين إلى أى مدى تتعرض تلك المنتجات للتلوث بمختلف أنواع الميكروبات المعوية الممرضة وما يمثله ذلك من خطورة على الصحة العامة ، ومن الممكن الحد من تلك الخطورة باتباع الإرشادات الصحية وبسترة اللبن قبل تصنيع منتجاته المختلفة.