Original Article

Detection of Microbial Contamination in Some Types of Processed Cheeses available In Local Market.

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Abstract:

Background: Cheese has an outstanding nutritional quality, but is also an efficient vehicle for transmission of diseases to humans and is an excellent medium for bacterial growth and an important source of bacterial infection. when consumed all without pasteurization Salmonella spp. are one of the most frequently reported causes of bacterial foodborne worldwide.

Objective: This study was carried out to study the microbiological contamination of processed cheese.

Material and Methods: A total of 13 samples of processed cheese were randomly collected from supermarkets in Baghdad, IRAQ. Elven grams of cheese were added to 99ml of sterile diluted peptone water in a flask and shaken well to make 10^{-1} dilution .Further dilution were made. 0.1 ml was used to incubate culture media (selective) incubated at 37C°and 42C° for 24 hours. Colonies were counted. Bactrac 4300 was used to confirm diagnosis of Salmonella spp.

Result: Although none of tested samples contained Salmonella spp. Or Staphylococcus aureus or Escherichia coli either other bacteria using conventional method, one sample of them been contaminated by Salmonella spp. using Bactrac 4300 system.

Conclusion: It can be concluded that processed cheese contaminated by this pathogen in this area as well in other countries and might constitute a risk for contamination and Bactrac 4300 a new method used for confirmation.

Keyword: Processed cheese, Microbiological test, Salmonella.

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Introduction:

Processed cheese is a dairy product differing from the natural cheese in that it is not made directly from milk, although the main ingredient is the natural cheese. Processed cheese can be made from a simple basic set of ingredients consisting of cheese, water, salt, different type of proteins, fat, stabilizer, flavorings and added minerals (1). The microbiological quality of dairy product is influenced by the initial flora of raw milk, processing conditions and post-heat treatment contamination (2). Undesirable microbes that can cause spoilage of dairy product include Gram – negative psychrotrophs, Coliforms, Lactic acid Bacteria, Yeast and Molds. In addition, various bacteria of public health concern such as Salmonella spp., Listeria monocytogenes, Campylobacter jejuni , Yersinia enterocolitica , pathogenic strains of Escherichia coli and enterotoxigenic strains of Staphylococcus aureus , Bacillus spp., Clostridium spp., enterococci may also found in a dairy product (3). For this reason, increased emphasis should be placed on the microbiological examination of dairy foods. Microbiological analysis are critical for the assessment of quality and safety, conformation with standards and specifications and regulatory compliance (4). The processed cheese is not a preserved food, but a semi-preserved food with limited shelf life of 3- 4 months , especially when the product is packaged in plastic foils , while products stored in metal cans may have longer shelf lives of 6 – 12 months at room temperature (5). Recently processed cheese is imported from neighboring countries according to standards set by the government. Therefore, this study carried out to study the microbiological contamination of the processed cheese available in the local market.

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Materials and Methods:

Collection of samples: A total number of 13 processed cheese samples were collected from the supermarkets of Baghdad, IRAQ. All samples were analyzed before their expiry date.

All samples were immediately transferred to the microbiology laboratory at Market Research and Consumer Protection Center ,Baghdad University ,and stored at -18C° at deep freezer until use(table1).

Prepration of samples :The media used were in a dehydrated form and prepared according to the manufacturer's instructions. Elven grams of cheese were added to 99ml of sterile distilled peptone water in a flask and shaken well to make 10^{-1} dilution. Further dilutions were prepared in sterile distilled peptone water. Prepared samples were serially diluted (10^{-6}) in sterile water and used to enumerated bacteria in specific culture medium.

Estimation of microbial count: This was carried out

according to the methods described by (6and 7), which include the following methods:

a). Total Plate count : Total plate count were enumerated by pour plate method using standard plate count .Diluted samples were cultured on plate count Agar by using one ml of each dilution (10^{-6}), which added to petri-dish and incubated at 37C° for 24 hours, colonies were counted.

b).Enumeration of Staphylococci:

For the enumeration of coagulase positive Staphylococcus (Staphylococcus aureus), the mannitol salt agar was used for confirmation and incubated at 37C° for 24 - 48 hours, developed colonies were counted.

b).Enumeration of Escherichia coli:

For the enumeration of (Escherichia coli), Macconkey agar was used for confirmation and incubated at 37C° for 24 - 48 hours, developed colonies were counted, Suspected colonies were confirmed by TSI agar and indole, methyl red, Voges- Proskauer, citrate (IMViC) tests

C)-Salmonella Detection:

For the detection of Salmonella in cheese samples, prepared samples with (10^{-6}) dilution, (0.1ml) mixture was used to inoculate culture media. The methods of isolation of Salmonella spp. can be done by inoculating samples on Selenite or tetrathionate broth at 42C for 24 hours than subculture by using Salmonella Shigella agar (SSA) or brilliant green agar and incubated at 42C° overnight. All suspected colonies were submitted to the standard biochemical reactions which are Triple sugar iron (TSI). agar, Lysine decarboxylase (LIA), Urease ,Indole, methyl red, Simmon citrate utilization to confirm whether they belong to Salmonella spp. (8).Culture of Salmonella may produce colonies with large, glassy black centers some of them many appear as almost completely black colonies.

D)-Confirmation of Salmonella spp. by using Bactrac 4300:-

This equipment was used to confirm the diagnosis of salmonella in cheese sample (9). Broth media was used 201C Bimedia specific for detection of Salmonella spp. Incubated by measuring cells at 40C for 24 hours .

Table :(1) Collected cheese samples from Baghdad markets

No	Sample Name	origin	Date of production	Date of expiry	Volume /gram
1	The two cows	Egypt	2014/9/20	2015/9/19	200
2	Teama	Egypt	2014/10/13	2015/10/12	133
3	mini	Turkey	2014/10/2	2015/10/1	100
4	Kiri	Boland	2014/8/4	2015/6/1	100
5	Al-sabah	Egypt	2014/9/20	2015/9/19	100
6	Alvita	Egypt	2014/11/15	2015/11/14	100
7	Happy morning	Egypt	2014/9/25	2015/9/24	100
8	The two cows	Egypt	2014/8/15	2015/8/14	100
9	Dawn	Egypt	2014/12/7	2015/12/6	100
10	Dairyland	Egypt	2014/10/11	2015/10/10	400
11	Beefa	Australia	2014/5/12	2015/5/11	200
12	Buda	Hungary	2014/12/10	2015/12/9	200
13	Hajdu	Hungary	2014/10/26	2016/10/25	113

Statistical Analysis: Statistical significance was assessed by using least significant differences – LSD (T-test) P – value 0.05 was considered significance.

Results:

Salmonella spp., Escherichia coli, and Staphylococcus aureus were not detected in any processed cheese samples under study using conventional methods (table 2).While one sample of processed cheese (Beefa) was contaminated with Salmonella spp. By using Bactrac 4300 system. (table 3) (figure1) .

Table (2): Isolation of microbial species identified in the processed cheese samples.

No	Trade Mark of cheese samples	Total Count Bacteria CFu/g	Escherichia coli	Staphylococci CFu/g	Salmonella
1	The two cows	1×10^2	Nil	Nil	Nil
2	Teama	3×10^1	Nil	Nil	Nil
3	Mini	2×10^2	Nil	Nil	Nil
4	Kiri	1×10^1	Nil	Nil	Nil
5	Al-sabah	4×10^1	Nil	Nil	Nil
6	Alvita	12×10^2	Nil	Nil	Nil
7	Happy morning	9×10^1	Nil	Nil	Nil
8	The two cows	2×10^2	Nil	Nil	Nil
9	Dawn	1×10^1	Nil	Nil	Nil
10	Dairyland	6×10^2	Nil	Nil	Nil
11	Beefa	14×10^3	Nil	Nil	Nil
12	Buda	3×10^3	Nil	Nil	Nil
13	Hajdu	5×10^1	Nil	Nil	Nil

*Nil = NO growth

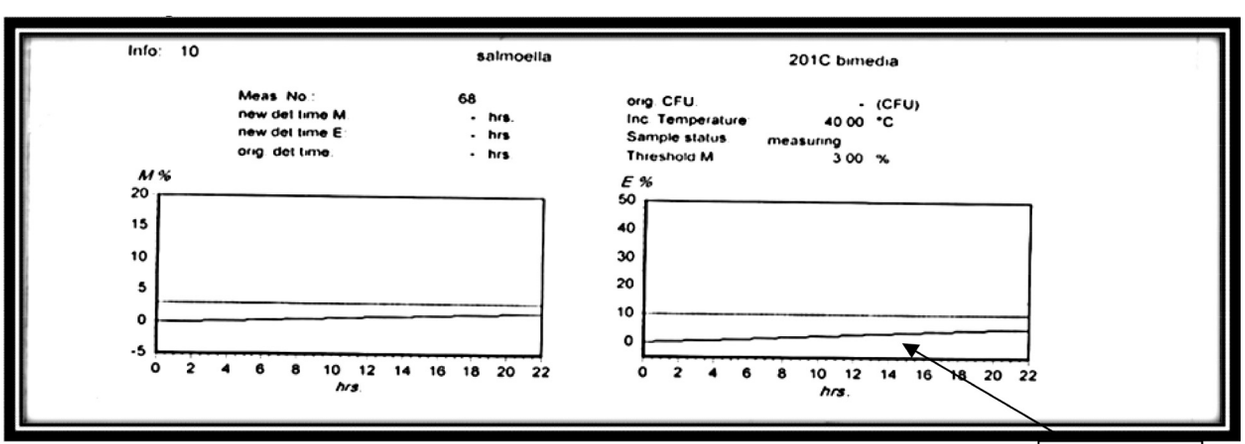
**Positive = growth

Table (3): Detection of Salmonella spp. by using Bactrac 4300

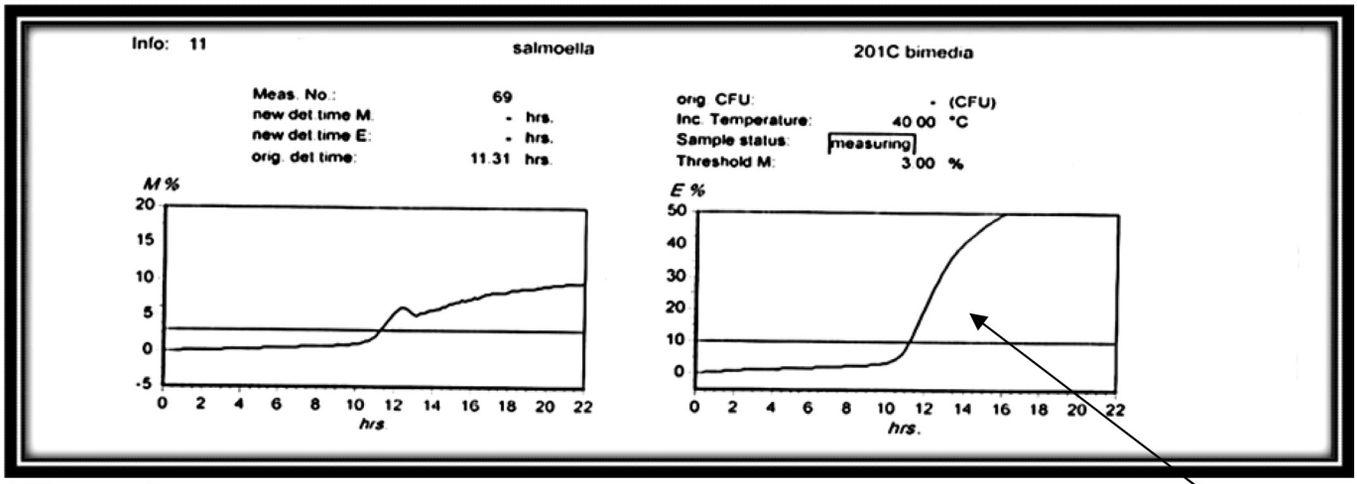
No	Sample of cheese	Salmonella
1	The two cows	Nil
٢	Teama	Nil
3	mini	Nil
4	Kiri	Nil
٥	Al-sabah	Nil
٦	Alvita	Nil
٧	Happy morning	Nil
٨	The two cows	Nil
٩	Dawn	Nil
١٠	Dairyland	Nil
١١	Beefa	positive
١٢	Buda	Nil
١٣	Hajdu	Nil

*Nil = NO growth

**Positive = growth



*(A): No growth



***(B): growth**

Figure (1): The Negative and Positive Result by Using Bactrac 4300 for Detection of Salmonella spp.

Discussion:

The microbiological results in our study indicated that the processed cheese sample collected during this investigation were of good microbiological quality since, Escherichia coli ,bacteria were not detected are other microorganisms like Salmonella and Staphylococcus aureus .Our results are in agreement with findings of other (10,11).The microbiological examination of processed cheese revealed good hygienic quality of the processed cheese under study that might due to the heat treatment of cheese, or the processed cheese might have been imported shortly before investigation or the storage conditions were good (12). The low percentage of contamination may be due to the season in which the study was performed, where the low temperatures of winter can effect the growth of species that usually grow out 37°C (13). So it is highly recommendation that strict hygienic conditions should be adopted during manufacturing and handling of such products, besides that local markets and processing should be periodically inspected by specialists (14). This system Bactrac4300 (Sy-lab impedance Analysis) used in study for the first time for detection of microorganism including Salmonella spp. using this technique rather conventional method indicate the easy and detailed documentation ,automation a cultural methods, reduced analysis times and saving on materials and work load (9).

Conclusion:

It can be concluded that processed cheese contaminated by this pathogen in this area as well in other countries and might constitute a risk for contamination and Bactrac 4300 a new method used for conformation

Reference:

1-Lee,S.K;Buwalde,R.J;Euston;S.R.and Mckenna,A.B.Changes in the rheology and microstructure of processed cheese during cocking.Lebensm.Wiss.V.Technol.2003.36;339-345.

2-Richter,R.C,Ledford,Ail and Murphy,S.C. Compendium of methods for the microbiological examination of foods. America Public Health Association, washinton.D.C,chapter 45.2005,pp.837-856.

3-Tatini,S.R;Kauppi,K.L.Encyclopedia of diary sciences .Roginski,H; Fuquay,J.W.(eds).Vol.(Academic pressand Elsevier science , Amsterdam,Boston,London.2003.pp.74-79.

4-Verga,L.Microbiological quality of commercial dairy products .Applied microbiology. 2007,vol.56,487-494.

5-Schar,W.and Bosst, J. Chemical and physicochemal changes in processed cheese and readymade fondue during storage. A review. Lebensm - wiss.2002,V-technol.35;15-20.

6-Honghtby, A.G; Maturin, L.J.and Koenig, K.E. Microbiological count methods . In ; Marshall RT(ed);standard methods of the examinations of dairy product.16th edition,Ed., Washington DC, American Public Health Association.2002,pp213-246 .

7-APHA.Compendium of methods for the Microbiological Examination of Food.3red.washinton.D.C,America Public Health Association. 2000,pp-220-230.

8-ArifE.D.Isolation and identification of salmonella species from local cheese in Sulaimania province.Al-anbar,J.Vet. Sci.2012, vol. 5;82-84.

9-Bactrac4300.Microbiology of food and animal feed-method validation.2013,FDIS published.

10-Nour, Eldiam, M.S.A,El Zubeir, I.E.Comparison of microbiological quality of processed and non-processed

Sudanese white cheese .Res.J.Microbiol.2006,(1); 273-279.

11-Suleiman,T;Mohamed Abdalla,M;Hassun,N;ElHaj,M. Chemical and microbiological evaluation of processed cheese available in Khartoum market,sudan.American.J.of food and Nutrition.2011,(1);28-33pects

12-Kapoor,R.and Metzger,L.E.Processed cheese :scientific and technological aspects A-review .Comp .Rev.Food.Sci. and Safety .2008 ,7:194-214.

13-Sliq,S;Hona,S.and Younes,A. Detection and isolation of Salmonella in some Syrian processed cheese.J.Agr. Sci.2010,26:321-350.

14-Ali,H.H.Salmonella serotypes isolated from local cheese in Mosul city. Iraq J.Vet.Sci.2005,(19);27-30.