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Bioscience Research

Print ISSN: 1811-9506 Online ISSN: 2218-3973

Journal by Innovative Scientific Information & Services Network

RESEARCH ARTICLE BIOSCIENCE RESEARCH, 2020 17(1): 282-287. **OPEN ACCESS**

Detection of certain microbial and chemical contamination in some traditional local dairy in Baghdad, Iraq: A case study

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In this study, locally produced cheese and yogurt samples were collected from ten different quarter of Baghdad city period May 2019, to detection of microbial contamination by the isolation, identification of morphological characteristics and biochemical tests of coliform, *Staphylococcus aureus*, Salmonella spp. and yeasts. The findings of microbial contamination referred that all samples of yogurt was out accepted limits in Iraqi microbiological specifications for coliform, yeasts and *S. aureus*, however, all samples were negative of Salmonella spp. contaminations, all samples of cheese under acceptable limit according to Iraqi standards specifications for coliform and *S. aureus*, with detection of Salmonella spp. in 3 cheese samples. The determinations of heavy metals i.e. lead and cadmium in the samples was done by atomic absorption spectrophotometer technique. The results related to Pb level showed that all samples were out of the accepted limits in the standards specifications, except yogurt sample (Bayaa quarter), while the level of Cd was lower and under accepted limit in all samples.

Keywords: Microbial and chemical contamination, local dairy products, cheese, yogurt, heavy metals

INTRODUCTION

The local products of cheese, cream and yogurt are one of the most important staple in resource they are produced by animal local breeders with direct marketing to the consumers. Dairy products are considered as one of the most essential foods considering their contents from protein, fat, vitamins and essential minerals for the life of human (Aziz, 2015). These products have suitable nutritional media for the growth of several microorganisms owning their an appropriate water activity, carbohydrates and protein, as well as other growth factors such environment is suitable for the growth and cultivation of microorganisms leading to multiple biological and chemical changes. These changes are responsible for the deterioration of quality and nutritional value of product (Ledenbach and Marshall, 2009). The hygienic conditions necessary for safety of production are not expected to be applied by the local producers due to absence of hygienic measures followed in manufacturing of milk and derivatives as a result of the absence of a healthy culture (Kumar and Prasad, 2010; Thaker et al., 2013). The contamination of these products may be due to exposure to various sources of chemical and biological pollutants which may begin from raw milk, production steps, packaging, storage,

handling and marketing to the consumer (Can and Çelik, 2012; Meshref et al., 2014). The milk using in the manufacturing of these products may be the causes that lead to possible another probable cause is the common infection of cow s udder leading to mastitis caused by Staphylococci, Streptococci and *Escherichia* coli (Baylis, 2009). The problem of contamination of dairy products by heavy metals is one of the obstacles that affect the health of the consumer that may be due to milking process, production steps and marketing (Bakircioglu et al., 2011; Shahbazi et al., 2016). Unfortunately, the heavy metals pollution *i.e.* cadmium and lead cannot be removed from milk and milk products during the production process is which represent a risk to the consumer health (Noaman and Al-Azzawi, 2016).

Due to the several traditional dairy products that are available in the local markets of Iraq, and the versatility ignorance in the preparation process, hence, the current study was designed to fulfill these goals

MATERIALS AND METHODS

Samples collection

Ten sample of each cheese and yogurt were collected from different quarter of Baghdad city which included Sadriya, Al-Amil, Bab Al-Mu'adham, Al-Shoula, Bayaa, Ragheba Khatoun, Ghazaliyah, Kasra, Kadhimiya and Adhamiya during May 2019. All samples were placed in an ice cooled box and transferred to the laboratory for tests which were performed with three replicates per test.

Detection of microbial contamination

The isolation, identification of morphological characteristics and biochemical tests of coliform, *Staphylococcus aureus*, Salmonella spp. and yeasts were carried out as described by others Alsoufi et al. (2016); Al-Musawi et al. (2018).

Determination of heavy metals concentration in cheeses and yogurt

Determination of heavy metals (lead and cadmium) in cheese and yogurt were carried out by putting 0.5gm of sample in 50mL digestion tube with addition of 5mL of concentrated nitric acid as a digestion solution, then, the mixture is heated at 80ºC for 2-3 hour until digestion of mixture become clear. Later, evaporated the surplus nitric acid. Was and cooled at room temperature, then, a dilute of 25mL by 0.2mol.L^{-1} of nitric acid was filtered through Whatman filter paper No. 1. Final solution was used of heavy metals was determined using Shimadzu AA-7000 atomic absorption spectrophotometer according to Al-Soufi et al. (2012); Abu-Almaaly (2017).

Statistical analysis

Data were statistically evaluated as mean ± standard error (SE), minimum and maximum values. All calculations were performed with the SPSS program (version 16, 2007).

RESULTS

The lead and cadmium concentrations (mg/kg) of yogurt and cheeses products showed differences within the samples, the highest and the lowest concentration of Pb in yogurt were 0.4659±0.0131 and 0.0147±0.0017 in Al-Amil and Bayaa quarters, respectively, while Cd was under detective limit (U.D.L) in samples of Al-Amil, Bab Al-Mu'adham, Kasra and Adhamiya area, the highest and the lowest Cd concentration of Bayaa and Ghazaliyah quarters were 0.00210±0.000163 and 0.0001±0, respectively. The highest and the lowest concentration of Pb in cheese were 0.0592±0.0018 and 0.0233±0.0019 in Sadriya and Kasra quarters, respectively, while Cd was under detective limit in samples of Sadriya, Al-Shoula, Bayaa, Ghazaliyah and Kasra quarters, the highest and the lowest concentration Cd of Ragheba Khatoun and Adhamiya quarters were 0.0002±0.000082 and 0.001±0.000163, respectively (Table 1).

Microbial contamination in yogurt and cheeses

The highest and the lowest count of coliform in yogurt were 130×10^3 and 5.3×10^3cfu/mL in Bayaa and Adhamiya quarters, respectively, the mean count for all yogurt samples was 36.7×10^3cfu/mL, while yeasts count was 58.3×10^3 and 15.3×10^3cfu/mL in Bayaa and Kadhimiya quarters, respectively, the mean count for all yogurt samples was 31.7×10^3cfu/mL, whilst, Salmonella spp. was no detectable in all yogurt samples. The highest and the lowest count of coliform in cheese were 22.7×10^6 and 2×10^6cfu/gm in Bab Al-Mu'adham and Ragheba Khatoun quarters, respectively, the mean count for all cheese samples was 11.3×10^6cfu/gm, while *S. aureus* count were 33.3×10^5 and 2.7×10^5cfu/gm in Kadhimiya and Al-Shoula quarters, respectively, the mean count for all cheese samples was 11.8×10^5cfu/gm,

respectively, however Salmonella ssp. was detectable (+) in cheese samples of Sadriya, Bab Al-Mu'adham and Bayaa (Table 2).

Table 1:Concentration of the Pb and Cd concentration (mg/kg) in yogurt and cheese.

No. of Samples	Quarters of study	Metal concentration (mg/kg)			
		Yogurt		Cheese	
		Pb	Cd	Pb	Cd
1	Sadriya	0.4238±0.0064	0.00013±0.000047	0.0592±0.0018	*U.D.L
2	Al-Amil	0.4659±0.0131	*U.D.L	0.0395±0.0018	0.0005±0.000163
3	Bab Al-Mu'adham	0.0249±0.0026	*U.D.L	0.0523±0.0011	0.0007±0.000163
4	Al-Shoula	0.0272±0.0017	0.00017±0.000047	0.0436±0.0024	*U.D.L
5	Bayaa	0.0147±0.0017	0.00210±0.000163	0.0404±0.0014	*U.D.L
6	Ragheba Khatoun	0.0249±0.0069	0.00047±0.000125	0.0269±0.0016	0.0002±0.000082
7	Ghazaliyah	0.0374±0.0017	*U.D.L	0.0544±0.0017	*U.D.L
8	Kasra	0.0455±0.0022	*U.D.L	0.0233±0.0019	*U.D.L
9	Kadhimiya	0.0488±0.0015	0.00017±0.000047	0.0248±0.0018	0.0003±0.000047
10	Adhamiya	0.0423±0.0013	*U.D.L	0.0347±0.0010	0.001±0.000163

*U.D.L.= under detective limit

Table 2:Microbial mean count for coliform, yeasts and Salmonella spp. and *S. aureus* in yogurt and cheese.

No. of Samples	Quarters of study	Microbial count					
		Yogurt (cfu/mL)			Cheese (cfu/gm)		
		Coliform ($\times 10^3$)	Yeats ($\times 10^3$)	Salmonella spp.	Coliform ($\times 10^6$)	*S. aureus* ($\times 10^5$)	Salmonella spp.
1	Sadriya	57	35	-	15.3	23.3	+
2	Al-Amil	2.7	20.3	-	5.7	5.3	-
3	Bab Al-Mu'adham	6.3	25	-	22.7	26.7	+
4	Al-Shoula	37	43.7	-	17.3	2.7	-
5	Bayaa	130	58.3	-	14.7	12.7	+
6	Ragheba Khatoun	47	41.7	-	2	4	-
7	Ghazaliyah	53	31.3	-	5.7	4.3	-
8	Kasra	5.7	25.7	-	6.3	3.3	-
9	Kadhimiya	23	15.3	-	18.7	33.3	-
10	Adhamiya	5.3	20.7	-	5.3	3	-
Mean count		36.7	31.7	-	11.3	11.8	30%

- Non detectable
+ Detectable

DISCUSSION

Heavy metals concentration in yogurt and cheeses

The Iraqi standards specifications No. 693/1/1988.(1988); No. 4079/2010. (2010) did not refer to level of lead and cadmium in cheese, while, the standards specifications of codex alimentarius commission (2015) and GCC Standardization Organization (2017) it refer that maximum level of lead, must be not more than 0.02 mg/kg, while it did not referred to the level of cadmium. The results of Pb level refer that all samples of yogurt and cheese are out of the limits allowed in the standards specifications, except yogurt sample (Bayaa) 0.0147±0.0017 mg/kg that it in the allowed level in the standards specifications, according to the level of Cd, the results recorded was lower and under detective limit in all samples.

The variation in lead and cadmium content in cheese and yogurt samples is due to the presence this metal in the can which is using in milk collection or that using in production and transportation, In this regard, the Iraqi standard specification No. 238/1984 (1984) indicated that it should be completely free of cadmium and lead because it will be transferred to the food placed therein, also, the pollution will be happened during the marketing from the contaminated environment.

In this regard, Ayar et al. (2009) was explain that the reasons of dairy products contamination by lead is due to the lead piping and lead-lined tanks in water supplies, and he refer that Pb content were 0.92 and 0.136 mg/kg for White cheese and yogurt (Ayran), respectively. Also Bakircioglu et al. (2011) observed that the lead content of cheese samples packing in tin containers were higher than other samples in plastic containers, and he attributed contamination to cheese making process and refer that the high temperature during cheese production might occur release of heavy metals from the equipment

using in this industries. While Meshref et al. (2014) noted that lead concentrations was in the ranges of 0.194-0.6495 ppm for kareish cheese, and he attributed the reason for this is that the presence of Pb in milk and dairy products may be due to environmental sources. However Suturovic´ et al. (2014) found that all samples of pasteurized and fermented milk products were compatible with Serbian regulations (Official Gazette SRY (No. 5/92 & 32/, 2002) that gives maximum contents of heavy metals in milk is 0.01 and 0.1 mg/kg for cadmium and lead, respectively, and he refer that the probable cause of heavy metals contamination in yogurt and cheeses are milk, equipment that use in production methods, water that used in cleaning process, packaging materials and marketing and handling. Whereas Shahbazi et al. (2016) refer that the ranges of mean Pb content 14.5 and 7.54 µg/kg in cheese and yogurt, respectively, and he found that the metals content of dairy products that collected in the winter were higher than summer season, and this it might have from the rainfall and consequently the washed down of wastes and contaminant of soil, so, considering the contamination above the limits in the standard specifications of yogurt and cheese samples by lead and cadmium, a control of metal content must be applied during the production process, marketing and all steps for these products from farm to consumer for prevent contamination with heavy metals.

Microbial contamination in yogurt and cheeses

According to Iraqi standard specification No. 2270/5/2006 (2006), the microbiological limits (cfu/mL) for yogurt and labena for coliform, yeasts and salmonella spp. were (1×10^1 and 1×10^2), (1×10^2 and 1×10^3) and (0 and 0) for the good and acceptable quality, respectively, while the microbiological limits (cfu/gm) for hard and semi hard cheese was (1×10^2 and 1×10^3) for *S. aureus*, (1×10^2 and 1×10^3) for Coliform and (0 and 0) for salmonella spp. for the good and acceptable quality, respectively. The results refer that all samples of yogurt and cheese are out the limits allowed in the Iraqi standards specifications for microbiological limits of coliform, yeasts and *S. aureus*, but all samples were empty of salmonella spp. yogurt, and it was detect of Salmonella spp. in 3 cheese samples (Sadriya, Bab Al-Mu'adham and Bayaa).

The presence of high microbial contamination in dairy products is due to the use of raw milk containing high numbers of bacteria, or to the fact that the heat treatments carried out on raw milk were not enough to completely eliminate the contamination (Ledenbach and Marshall, 2009; Thaker et al., 2013). Therefore, several studies have been conducted in this field, such as Baylis (2009) whose refer that raw milk cheeses may be a main source of food borne pathogens and it's a potential vehicle for Verocytotoxin (Shigatoxin)-producing *E. coli* (VTEC⁄STEC), and he explain that the many studies in the world show that *E. coli* O157 strains can survive the different stages of the cheese making process and milk products. While Kumar and Prasad (2010) they was found that Indian milk products, such as Dahi (traditional yogurt or fermented milk product) and Khoa (is a dairy product lower in moisture than typical fresh cheeses such as ricotta) were contamination with Staphylococcus and *E. coli*, and they refer that this may be due to unhygienic production conditions that lead to produce a dairy products which may cause food poisoning and pose a threat to public health of consumer. Also, when Can and Çelik (2012) were investigating the presence of enterotoxigenic and antimicrobial resistant *S. aureus* in Turkish cheeses, they observed presence of it in cheese samples, which it cause constitute a potential risk for public health, and they stressed the need to take the necessary steps for better control of cheese contamination sources and taking hygienic measures is necessary for food safety. Whilst Thaker et al. (2013) were could to isolation of 10 isolates 6.25% of *S. aureus* from 160 samples of Indian milk and milk products, and they found that this isolates were variably resistant to the antibiotics tested, which clearly refer a possibility of potential public health hazard by this bacteria that it contamination of milk and milk products with other pathogenic bacteria that is mainly due to unhygienic processing, handling and unhygienic environment.

General, the manufacturing of dairy products in homes, fields or small factories that are not controlled by health, as well as lack of attention to hygiene, good packaging and marketing in unsanitary conditions exposure of consumers to serious risks

CONCLUSION

Due to the lack of health control role, these products have become popular in the local markets. This increases the risk of consumers being exposed to many health risks; therefore it will be possible to control it by application of

standards specifications and sanitary control of these products by authorities to ensure high dairy products quality from farm to consumer.

CONFLICT OF INTEREST

The authors declared that present study was performed in absence of any conflict of interest.

ACKNOWLEGEMENT

The authors acknowledge the support provided by University of Baghdad and University of Al-Mustansiriah, Iraq through the using of laboratories of Department of Evaluating Products, Market Research and Consumer Protection Center, University of Baghdad and laboratories of Department of Science, College of Basic Education, University of Al-Mustansiriah.

AUTHOR CONTRIBUTIONS

Mohammed Abdulrazzaq Alsoufi: designed the study, interpreted the data, and drafted the manuscript.

Raghad Akram Aziz: involved in collection of data and also contributed in manuscript preparation.

Raffat Ahmed Abu-almaaly: conduct microbial and chemical analysis.

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