

Microbial Quality of infant formula milk powder in Baghdad City

Shiamee Mohammed Abdelreda, Dr. Reyam Naji Ajmi

Abstract— Due to the importance of infant feeding, which depends essentially of milk, which is the first in food chain and the purposes of ensuring the health and safety by getting the best product with the standard specifications to contribute to the growth of the natural progression of this study was to examine the microbiological quality of infant milk powder. A total of five types of milk powder traded in local markets and pharmacies in Baghdad, which are used as substitutes for breast milk during the first day of opening the box powders such as Total Coliform count (TC), Total Salmonella count (TS), Total plate Count (TPC) and Yeasts & Molds Count (YMC). Results were obtained respectively; Total plate count ($<0.05 \times 10^3$ cfu/g) were significantly higher than Total Coliform count ($<0.05 \pm 1.0 \times 10^2$ cfu/g) and Total Salmonella count ($<0.05 \pm 1.0 \times 1.0$ cfu/g) and Yeasts and Molds ($<0.05 \pm 1.0 \times 1.0$ cfu/g). All samples of infant formula of milk powders having non-significant difference. The obtained averaged results compared to Iraqi Quality Standards (IQS) values ($<0.05 \pm 1.0 \times 10^2$ cfu/g). Microorganisms in the infant formula cannot grow due to low humidity. Consisted studied milk samples to maintain health standards during production, so the present study was designed to assess the microbiological quality of the milk powders. This result indicated that all milk powder companies understudied are following the legal and quality standard although Total Count were within the range of IQS (Iraqi Quality Standards) and USA Environmental Protection Agency USEPA and as indicates the hygienic condition of Infant formula milk powders without risk level for human health.

Keywords: Microbiological Quality, Infant Milk Powder, Enterobacteriaceae Count, Yeasts and Molds count

1 INTRODUCTION

Milk powder is generally considered a good microbiological quality production; however, several factors may contribute to changes in the physical and chemical properties that reduce the shelf life and commercial value [1]. Various researchers agree that the health conditions under which raw milk production is the main factor affecting the quality of powder [2, 3, 4,5]. The degree of storage and transport temperatures may also affect milk powder properties, especially solubility and pH indicator [6]. Milk is a food complement, high nutritional value makes it an ideal way for rapid multiplication of bacteria, especially of unhealthy production and storage temperatures [7,8] milk powder quality to be free of disease-causing bacteria and toxic substances harmful, Microbiological analyzes critical to assess the safety, quality, conformation with the standards and specifications, and regulatory compliance [7].

Critical microbiological analyzes to assess the safety and quality conformation with the standards of milk powder in unsanitary conditions must not contain more than 5.0×10^2 bacteria per milliliter [9]. Total bacterial count reflects the hygiene

practices used in the production, processing and dealing with milk as an indicator health standards during the production, processing. Pathogenic bacteria in infant formula, such as Staphylococcus aureus, Escherichia coli, Streptococcus aecalis, Streptococcus pyogenes, Pseudomonas, Proteus and Salmonella is considered unacceptable. The presence of Molds and Yeast would indicate unsatisfactory hygiene conditions [10,11] Even though food intolerance (abdominal bloating, vomiting, diarrhea, or gastric retention) is frequent in the feeding process of infants [11]. Has found scarcely salmonella in infant formula powder [11,12,13,14,15,16,17,18]. Salmonella cause a range of diseases in humans. Coliform and E. coli and Total count is used as an indicator of reliable pollution refers to the possibility of the introduction of pathogenic micro-organisms to disease in milk [7,8] E. coli Top 500 cells / ml indicates a lack of hygiene while cleaning equipment [9]. The border accepted coli count in the milk must be less than 100 cells / ml [19,20,21], but some prefer the count of 50 cells / ml [22,23], differs from the growth of bacteria from one type of bacteria to another. While one bacterial species may grow under certain conditions, other types may weaken. These conditions are interrelated and include the ability nutrient availability, and levels of oxygen and moisture, and the level of other gases, having pH inhibitors, temperature [23]. Coliforms can cause damage to the milk because they ferment lactose with the production of acid and gas and can also lead to a deterioration of the milk proteins [19]. Yeasts and fungi in the milk may create a risk to human health products [24,25], many of these

- 1st Shemaa Mohammed Abdelreda, Master Student, Department of Biology Science, AL-Mustansaryah University, Baghdad 496. Iraq
- 2nd Dr. Reyam Naji Ajmi, Department of Biology Science, AL-Mustansaryah University, POX 46079 Iraq-Baghdad
Email : reyam80a@yahoo.com

Microbial can cause an imbalance in the formative characteristics of the milk and microbial growth is a major concern objects [26,27,28,29].

Many Studies have shown that people who are allergic to mold suffer from asthma, and health effects in infants has evolved to have respiratory symptoms where is Penicillium signs for infants, such as a persistent cough Wi increased exposure increases the risk of respiratory symptoms during the first year of life [30] exposure to molds can cause a number of health issues such as throat irritation, nasal congestion, coughing, wheezing, eye irritation and skin irritation as well as in some cases. Exposure to mold may also cause anaphylaxis depending on the time as well as chronic lung disease [31].

Yeast and mold contamination are important in milk, while the yeast does not lead to food poisoning, it does not cause food to spoil types. A different template produces toxic substances that have been identified as mycotoxins. Some mutagenic and carcinogenic, some special offer organ toxicity and some Sam [26,30], yeasts produce toxic compounds called mycotoxins, secondary metabolites produced by fungi under certain environmental conditions [31].

2 MATERIALS AND METHODS

It was isolated and identify some Microbial Quality of infant formula milk powder of milk in five selected types of local markets in the city of Baghdad - Iraq (city coordinates: 33 ° 20'00 "N 44 ° 26'00" E) and the disclosure of The main components as total plate count, total coliforms count (TC) salmonella (TS), yeast and mold count (YMC) according to Iraqi quality stander (IQS) No.2006/3/2270 [32].

2 EXPERIMENTAL PROCEDURES

2-1 PREPARATION OF MILK SAMPLES:

Infant formula milk powder (50 g) for each one was diluted in warm (45oC) sterile diluents peptone water solution (90 ml) to make primary dilution (10^1). Then a series up to 10^{-5} dilution was prepared by transferring primary dilution (1 ml) into test tube containing sterile diluents (9 ml) to obtain 10^2 dilution and repeating the operations with sterile diluents (9 ml) using the 10^2 and further dilutions to obtain 10^3 , 10^2 and/or 10^3 .

2-2 ENUMERATION OF TOTAL VIABLE BACTERIA:

Total plate count was used for enumeration of total viable bacteria. . For the determination of TVC (1) ml of each dilution was transferred using sterile pipette and spreader on plate count agar using a sterile petri dish for each sample. The petri dish was then kept in an incubator at 37oC for24-48 hours. Following incubation, plates exhibiting 30-300 colonies were counted. The average number of colonies in a particular dilution was multiplied by the dilution factor to obtain the TVC.

The TVC was expressed as the number of organism of colony forming units per ml (CFU/ml) of samples according to [33].

2-3 ENUMERATION OF TOTAL COLIFORM BACTERIA

The medium used for coliform was violet red bile lactose agar (VRB) (1) ml of each dilution samples was transferred using sterile pipette and spreaded on (VRB) AGAR using a sterile petri dish for each sample. The petri dish were then kept in an incubator at 37oC for24-48 hours.. After incubation, typical pinkish and centrally red colonies were counted.

2-4 SALMONELLA

The medium used for salmonella was Sabouraud dextrose agar (SDA) (1) ml of each dilution milk samples was transferred using sterile pipette into two tubes of(1)Selenite cysteine broth inoculated for 24 hr at 35CO.[2]Tetrathionate brilliant green broth(TBG) inoculated for 24 hr at 42,5CO AFTER incubation broth was streaked onto xylose lysine desoxycholate agar(XLD)agar and incubated for a further 24 hours at 37 OC. The colonies were examined for the characteristic pinkish color of Salmonella with or without hydrogen sulphite Suspected colonies were then subjected to indole, motility, oxidase, urease, citrate and sugar fermentation tests for identification of Salmonella spp.

2-5 YEASTS AND MOLDS GENERAL COUNTING METHOD

The medium used for fungi and yeast was Sabouraud dextrose agar(SDA) (1) ml of each dilution milk samples was transferred using sterile pipette on(SDG)agar using a sterile petri dish for each sample the petri dish were then kept in an incubator at 24co for4-5days.

2-6 STATISTICAL METHOD

The statistical analysis was performed according to the AOAC Protocol [34,35] was assessed using different measures of statistical sigma plot and coefficient of determination, interclass correlation coefficient and concordance correlation coefficient, mean prediction error the concentration was the concentration of the same milk component standard method. The coefficient of determination, r^2 , was calculated where N is the total number of paired observations. A value of $r^2 = 1$ indicates 100% precision between the methods.[33] was calculated to determine overall. The mean prediction error (Pe) was computed to describe the predictive performance of the methods and to compare prediction methods to the standard method and to one another [28,29]. Data plotting method by used to analyses the agreement between the standard methods and according to Iraqi quality stander (IQS) No.2006/3/2270 [32]. The 98% limits of agreement were calculated as the mean.

3- RESULTS AND DISCUSSION

The results were conducted to assess the quality of public health quality of milk powders and infant, microbes it has been observed. Although the microorganisms in infant formula due to decline. The moisture content cannot grow .Total plate count (<0.05 0.1×10^3 cfu/g) were significantly higher than Total Coliform count ($<0.05 \pm 1.0 \times 10^2$ cfu/g) and Total

Salmonella count ($<0.05 \pm 1.0 \times 10^2$ cfu/g) and Yeasts and Molds ($<0.05 \pm 1.0$ cfu/g). All samples of infant formula of milk powders having non-significant difference. The obtained averaged results compared to Iraqi quality standard (IQS) values ($<0.05 \pm 1.0 \times 10^2$ cfu/g). When comparing the results obtained with previous studies have proved that are of great importance because it is the index of health standards during storage and marketing, A study conducted by [36] less than total viable count to three types ($3.9 \times 10^3 \pm 3.0 \times 10^2$ cfu/g) ($3.4 \times 10^3 \pm 5.0 \times 10^2$ cfu/g), ($3.3 \times 10^2 \pm 6.1 \times 10^2$ cfu/g) respectively, observed in the infant formula is lower than too reported by [36,37,38] ($5.6 \times 10^2 \pm 4.3 \times 10^3$), ($8.7 \times 10^3 \pm 0 \times 10^2$) and same results [38]. Enterobacteriaceae count was not significantly ($p > 0.05$) study is lower than reported by [39,40] (13×10^3). It is the general concept that Enterobacteriaceae are not present in Infant formula proved by various researches. The Infant formula powder is packed hygienically in large sterilized containers and bags. However, transportation some damaged containers and bags have been observed, probably they can contaminate the milk powders non found yeasts and moulds that's were non-significant. However, the mean value them in the present study is lower than the results presented by [38] ($>10^3/100$ g) in powder milk and reported by [41] i.e $<10^3/g$ in milk powder. Presence of yeasts and moulds in milk or milk products, molds may create hazard to one's health, produce an allergen and an irritant to human health [42,43].

Sample No.	Total plate count	Total Coliform count	Total Mold and Yeast count	Total Salmonella count	Deviation in folds from ISI standard Sample (a) (b) = (x) ÷ (a)
1	Non	Non	Non	Non	-20 <0.05
2	Non	Non	Non	Non	-20 <0.05
3	Non	Non	Non	Non	-20 <0.05
4	Non	Non	Non	Non	-20 <0.05
5	Non	Non	Non	Non	-20 <0.05
Observed	3000	3000	3500	3000	-20 <0.05
Allowed Max Value	1×10^4	1×10^2	Zero	Zero	-20 <0.05

a = Observed Values
x = (Standard Value of IQS = < 50000 cfu/g)
IQS = Iraqi quality Standards

Table (1): shows the results Microbial Quality (cfu /g) in different infant formula samples compared to IQS standards

4 RECOMMENDATION

Microorganisms in the infant formula cannot grow because of the low moisture content does not play any direct role in the damage it. But it occurs in milk powder for infants each is of

great importance and serves as an indicator of the health standards of the province during the production, processing and handling. Children's milk provides high nutritional value that can support a wide range of bacteria as well as yeast and mold growth and reproduction substrate

In order to obtain information about the characteristics of quantity and quality needed to milk powder, raw materials, and this can be achieved by taking advantage of this information that has been obtained from the search by selecting potential in a wide range of applications in the examination of basic foodstuffs it has included the monitoring process, and determine the quality of milk origin, related to a study conducted in the laboratory and pilot-scale with the consumption and import, which is growing in size due to the lack of local and national product, due to the greed of merchants and critical cases of significant economic losses therefore, we recommend using these techniques that allow rapid analysis the large size of the samples.

In the production and consumption of the product line for the purpose of surveillance and use in helping to food safety and quality standards and affordable. To implement these decisions by detecting minute in infant formula organisms to ensure food quality and safety

ACKNOWLEDGMENT

This work was supported by the Fund from Science and UNESCO (United Nations Educational, Scientific and Cultural Organization) in Paris. The author is extremely grateful to all the people for their cooperation and help to get data.

REFERENCES

- [1] Cousins, C.M.; Bramley, A.J. Microbiologia de la leche cruda. In: Robinson, R.K. (ed). Microbiologia lactologica. V.1. Acribia, Zaragoza, 1987, p.109-150.
- [2] Lück, H. Control de la calidad de la industria lactologica. In: Robinson, R.K. (ed). Microbiologia lactologica. V.2. Acribia, Zaragoza, 1987, p.255- 294.
- [3] Muir, D.D.; Griffiths, M.W.; Phillips, J.D.; Sweetsur, A.W.M.; West, I.G. Effect of the bacterial quality of raw milk on the bacterial quality and some other properties of low-heat and high-heat dried milk. J. Soc. Dairy Technol., 39: 115-118, 1986.
- [4] Woodhall, M. The application of hazard analysis and critical control point system to milk powder manufacture. J. Soc. Dairy Technol., 42: 102-105, 1989.
- [5] Griffiths, M.W.; Phillips, J.D.; West, I.G.; Sweetsur, A.W.M.; Muir, D.D. The quality of skim-milk powder produced from raw milk stored at 2 degree. Food Microbiol., 5: 89-96, 1988.
- [6] Jayaro B, Henning DR. J Dairy Sci, 2001; 84:2157-2162.
- [7] Kim, H., Hardy, J., Novak, G., Ramet, J. P. and Weber, F. 1983. Off-tastes in raw and reconstituted milk. FAO Animal Production and Health Paper, 35: 2.

- [8] OECD 2005. Dairy policy reform and trade liberalization. Organisation for economic co-operation and development, p. 98, OECD Publishing
- [9] O'Connor CB. Rural dairy technology. ILCA training manual. Addis Ababa, Ethiopia: International Livestock Research Institute; 1994. p. 133.
- [10] Dardes de Almeida C, Rozolen C, Goulart A, Kopelman B. Is Breast Milk Collected at Home Suitable for Raw Consumption by Neonates in Brazilian Public Neonatal Intensive Care Units? *J Hum Lact* 2006;22:418-25.
- [11] Serafini A, André M, Rodrigues M, Kipnis A, Carvalho C, ET al. Microbiological quality of human milk from a Brazilian milk bank. *Rev Saude Publica* 2003;37(6):775-9.
- [12] Pickett, G. and Agate, G.H. (1967) Outbreak of salmonellosis due to a lactose-fermenting variant of *Salmonella* Newington. *Morbidity and Mortality* 16:18.
- [13] Rowe, B., Begg, N.T., Hutchinson, D.N., Dawkins, H.C., Gilbert, R.J., Jacob, M., Hales, B.H., Rae, F.A. and Jepson, M. (1987) *Salmonella* ealing infections associated with consumption of infant dried milk. *Lancet* 2(8564):900-903.
- [14] CDC (Centers for Disease Control and Prevention). (1993) *Salmonella* Tennessee in powdered milk products and infant formula - Canada and the United States. *MMWR Mortal. Wkly. Rep* 42:516-517
- [15] Usera, M.A., Echeita, A., Aladuena, A., Blanco, M.C., Reymundo, R., Prieto, M.I., Tello, O., Cano, R., Herrera, D. and Martinez-Navarro, F. (1996) Interregional foodborne salmonellosis outbreak due to powdered infant formula contaminated with lactose-fermenting *Salmonella* virchow. *Eur J Epidemiol* 12(4):377-381.
- [16] Threlfall, E.J., Ward, L.R., Hampton, M.D., Ridley, A.M., Rowe, B., Roberts, D., Gilbert, R.J., Van Someren, P., Wall, P.G. and Grimont, P. (1998) Molecular fingerprinting defines a strain of *Salmonella* enteric serotype Anatum responsible for an international outbreak associated with formula-dried milk. *Epidemiol Infect* 121(2):289-293.
- [17] Olsen, S.J., Bishop, R., Brenner, F.W., Roels, T.H., Bean, N., Tauxe, R.V. and Slutsker, L. (2001) The changing epidemiology of salmonella: trends in serotypes isolated from humans in the United States, 1987-1997. *J Infect Dis* 183(5):753-761
- [18] Bornemann, R., Zerr, D.M., Heath, J., Koehler, J., Grandjean, M., Pallipamu, R. and Duchin, J. (2002) An outbreak of *Salmonella* serotype Saintpaul in a children's hospital. *Infect Control Hosp. Epidemiol* 23(11):671-676.
- [19] Shojaei ZA, Yadollahi A (2008). Physicochemical and Microbiological Quality of raw milk, Pasteurized and UHT milks in Shops. *Asian J. Sci. Res.*, 1(5): 532-538.
- [20] Douglas JR (2003). Bulk tank cultures are the dairy man best friend. University of Wisconsin Milking Res. Inst. Lab., (Report) No. 2223.
- [21] Boor KJ, Brown DP, Murphy SC, Bandler DK (1998). Microbial and chemical quality of raw milk in New York State. *J. Dairy Sci.*, 81: 1743-1748.
- [22] College of Agric and Life Science (CALS) (2001). Department of Food Science. Cornell University. Dairy Science facts - Milk quality improvement program (1984 -2001).
- [23] J. D. Eifert, C. Gennings, W. H. Carter, S. E. Duncan and C. R. Hackney, "Predictive Model with Improved Statistical Analysis of Interactive Factors Affecting the Growth of *Staphylococcus aureus*," *Journal of Food Protection*, Vol. 59, No. 6, 1996, pp. 608-614.
- [24] United States Department of Agriculture Agricultural Marketing Service, "Commodity Areas," 2013. <http://www.ams.usda.gov/AMSV1.0/dairy>
- [25] ICMSF (International Commission on Microbiological Specifications for Foods) 1978. *Microorganisms in Foods. 1. Their significance and methods of enumeration*. Toronto .
- [26] Kaper, J.B., J.P. Nataro and H.L.T. Mobley, 2004. Pathogenic *Escherichia coli*. *Nat. Rev. Microbiol.*, 2: 123-140.
- [27] Singh T. K., Drake M. A., Cadwallader K. R. (2003). Flavour of Cheddar cheese: a chemical and sensory perspective. *Comp. Rev. Food Sci. Food Saf.* 2, 139-162
- [28] López-Malo A., Maris Alzamora S., Palou E. (2005). *Aspergillus flavus* growth in the presence of chemical preservatives and naturally occurring antimicrobial compounds. *Int. J. Food Microbiol.* 99, 119-128 [10.1016/j.jfoodmicro. .08.010](https://doi.org/10.1016/j.jfoodmicro.2005.08.010).
- [29] Rydlo T., Miltz J., Mor A. (2006). Eukaryotic antimicrobial peptides: promises and premises in food safety. *J. Food Sci.* 71, 125-135
- [30] Jump up^ Gent, Janneane. "Levels of Household Mold Associated with Respiratory Symptoms in the First Year of Life in a Cohort at Risk for Asthma" (PDF). Department of Epidemiology and Public Health, Yale University. Retrieved November 18, 2011.
- [31] Jump up^ Cohen, Aaron. "WHO Guidelines for Indoor Air Quality: Dampness and Mould" (PDF). World Health Organization. Retrieved November 18, 2011.
- [32] Iraqi quality stander. 2006/3/2270. Microbiological limits in food part three. Enumeration and Identification of microbiological groups in foods .
- [33] FAO/WHO. 1973. Code of principles concerning milk and milk products (7th Ed.) Standard for whole milk powder, partly skimmed milk powder and skimmed milk powder. No. AS. FAO/WHO.

[34] International Dairy Federation 1987b. ISO 6731:1989 IDF 021B:1987 Milk, cream and evaporated milk—determination of total solids content (reference method). Brussels, International Dairy Federation. 2 p.

[35] ISO (1995). International Organization for Standardization. Recommendation of the meeting of the ISO of meat and meat products. ISO/TC-34/SC.6. The Netherlands. pp. 10-18

[36] Yadav, J.S., S. Grover and V.K. Batish, 1993. Microbiology of dried milks. A Comprehensive Dairy Microbiology. B.V. Gupta, metropolitan book Co Pvt. Ltd. pp: 315-349.

[37] Rueckert, A., R.S. Ronimus and H.W. Morgan, 2005. Rapid differentiation and enumeration of the total, viable vegetative cell and spore content of thermophilic bacilli in milk powders with reference to *Anoxybacillus flavithermus*. J. Applied Microbiol., 99: 1246-1255.

[38] Flint, S., J-L. Drocourt, K. Walker, B. Stevenson M. Dwyer, I. Clarke and D. McGill, 2006. A rapid, two-hour method for the enumeration of total viable bacteria in samples from commercial milk powder and whey protein concentrate powder manufacturing plants. Int. Dairy J., 16: 379-384.

[39] Taha, S., M. Naguib and A. Ghani, 1972. Dried milk microbiology. J. Milk. Food. Technol., 36: 559.

[40] Parihar, P. and L. Parihar, 2008. Dairy microbiology Chapter, 3. Microbes. Agrobios India, pp: 46-50.

[41] Ceittao., M.F, F. De, I. Dalzani and H. Mazzoni, 1973. Cited Dairy Science. Abstract, 39, 6388.

[42] Springer S, Annibale D. Necrotizing enterocolitis. [Accessed: 03/22/2012]. Available at: <http://emedicine.medscape.com/article/977956-overview>.

[43] Deak, T., 2008. Yeast in Specific Types of Foods. In Handbook of Food Spoilage Yeast, Deak, T. (Ed.). 2nd Edn., CRC Press Taylor and Francis Group, Boca Raton,.