PREVALENCE OF SALMONELLA IN MILK AND MILK PRODUCT IN DUTSE LOCAL GOVERNMENT AREA OF JIGAWA STATE

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ABSTRACT

The study was conducted in Dutse, Jigawa State to determine the Prevalence of Salmonella in milk and milk products. A total of two hundred and forty (240) samples were collected using systematic sampling technique. The samples which composed of raw milk (n=80), nono (n=80) and fura (n=80) were collected from different locations in the study area. All the samples were collected in sterile bottles, kept in their original package, labelled and transported quickly as possible in ice box to the microbiology laboratory of Faculty of Science, Federal University Dutse for the conventional identification of Salmonella. One milliliter (1ml) of each of the eighty (80) samples of fresh milk, nono and fura were used for serial dilution of 10^{-1} to 10^{-6} dilution factors. 10^{-2} dilution was inoculated into sterile petridishes prepared by manufacturer instructions and incubated at 370C for 24 hrs to observe for typical growth of salmonella. All samples that exhibited characteristic colonies of Salmonella were subcultured onto nutrient agar slants and then stored at 4^0C until required. Twenty-five (25 ml) of raw milk sample and also 25 grams of milk ~~additives~~ products were homogenized into 225 ml of 0.1% sterile buffered peptone water and incubated at 37°C for 24 h. One ml of the homogenate was added aseptically to 9 ml of Rappaport Vassilliadis (RV) broth and kept overnight at 35°C. Isolated organisms with supporting growth characteristics of Salmonella were subjected to various biochemical tests. Data obtained during the study were summarized into tables using descriptive statistics. The result of the study showed that, higher prevalence of 5.80% was obtained for Salmonella from samples of fresh milk, followed by nunu that had 4.60% while fura was least with 3.80%. The overall prevalence of Salmonella in milk samples was found to be 14.20%. The study revealed an overall prevalence of salmonella in fresh milk to be 87.5%. 25%, 18.75%, 18.75%, 12.5% and 12.5% of the overall prevalence were recorded in behind road safety, Katangare, Gyabiya, behind house of assembly quarters and metropolis respectively. The mean coli form count of the milk and milk additives from the two hundred and forty (240) samples ranged from $3.95x10^6$ in fresh milk to $5.51\ x10^5$each in nono. There was prevalence of salmonella species in a higher proportion in raw milk and its products such as nono and fura in the study area. It was recommended that, there should be implementation of hygienic measures during milking and manufacturing of dairy products to minimize the risk of human infections with Salmonella

Key words: Prevalence, salmonella, milk, additives, milk products

INTRODUCTION

Milk is a fresh, clean and normal mammary secretion obtained by milking of one or more dairy animals for nourishment of their young. In dairy animals, cow milk is the most consumed milk worldwide and serves as good source of animal proteins, fats, vitamins and minerals to the human body (Osterholm and Norgan, 2014). Despite rich in nutrients, fresh milk and its fermented products constitute a good medium for the multiplication of various bacteria, and hence serve as a medium for the transmission of milk borne diseases mostly *salmonellosis, brucellosis, tuberculosis, shigellosis* and *staphylococcosis* (Karon *et al*., 2017). Pathogenic bacteria in raw milk have been a major factor for public health concern. The main sources of contamination are the dairy cattle handlers and dairy equipments. Consumption of raw milk is considered to be the main cause of several outbreaks of *Salmonella spp*. However, raw cow milk forms the basis for most commonly sold local milk products in Nigeria. Some milk products in Nigeria include locally fermented skimmed milk known as "Nono", full creamed milk, "Kindirmo" local butter, "Man Shanu" and cheese, "Wara". Salmonella is a gram-negative bacterium, rod shaped, aerobic or facultative anaerobic, belonging to the family Enterobacteriaceae. It is a major pathogenic bacterium' inhabiting the

intestinal tract of humans and animals. *Salmonella spp* are important sources of bacterial contamination of the environment and the food chain, and are the leading causes of acute gastroenteritis in several countries (Osterholm and Norgan, 2014)

MATERIALS AND METHODS

The study was conducted in Dutse, Jigawa State. Dutse. A total of two hundred and forty (240) samples were collected using systematic sampling technique. The samples which composed of raw milk (n=80), nono (n=80) and fura (n=80) were collected from different locations in the study area. One milliliter (1ml) of each of the eighty (80) samples of fresh milk, nono and fura were used for serial dilution of 10^{-1} to 10^{-6} dilution factors. 10^{-2} dilution was inoculated into sterile petridishes prepared by manufacturer instructions and incubated at 370C for 24 hrs to observe for typical growth of salmonella. All samples that exhibited characteristic colonies of Salmonella were subcultured onto Nutrient agar slants and then stored at 4oC until required. Twenty-five ml of raw milk sample and also 25 grams of milk additives were homogenized into 225 ml of 0.1% sterile buffered peptone water and incubated at 37°C for 24 h. One ml of the homogenate was added aseptically to 9 ml of Rappaport Vassilliadis (RV) broth and kept overnight at 42°C. The colony forming unit on each plate were observed and counted after 24 hrs of incubation. Pure colonies of salmonella isolates were picked with bacteriological loop, smeared on to separates clean free grease glass slide with a drop of distilled water and fixed by gentle heating. Crystal violet was applied on each smear to stain for 1 minute and then washed with water. Isolated organisms with supporting growth characteristics of Salmonella were subjected to various biochemical tests. Biochemical tests such as Triple sugar iron (TSI), Simmons citrate and urease were used. The isolates of Salmonella were sub cultured onto the biochemical media, incubated at 37ºC for 24-48 hours to check for phenotypic changes within the media (Global Salm-Surv, 2003). Data were summarized into tables using descriptive statistics. Statistical Analysis System (SAS) version 9.3 was used to determine the Fisher's Exact Test for association between Salmonella and sources of samples. The $P < 0.05$ was considered significant.

RESULTS AND DISCUSSION

Table 1: prevalence of salmonella isolates in milk in relation to milk type

Milk type	Number of samples examined	Number positive for salmonella	Location specific prevalence (%)	Overall prevalence (%)	χ^2	p-value
Fresh milk	80	14	17.5	5.8	1.30	0.52^{NS}
Nunu	80	11	13.8	4.6		
Fura	80	9	11.3	3.8		
Total	240	34	-	14.2		

Table 1 above showed the prevalence of salmonella isolates in milk in relation to milk type. The number of samples from each milk product was 80, out of which the number of positive samples was found to be 14 in fresh milk, 11 in nunu and 9 in fura. Higher prevalence of 5.80% was obtained for Salmonella from samples of fresh milk, followed by nunu that had 4.60% while fura was least with 3.80%. The overall prevalence of Salmonella in milk samples was found to be 14.20% (Table 1). Several reports have documented the prevalence and distribution of Salmonella in milk, but there is paucity of information in the study area. The prevalence of Salmonella observed in milk products was 14.20%. This is in agreement with the findings of Hasan *et al*. (2018) who reported a prevalence of 11% in Iran, but lower than the 20% reported by Kuvandik *et al.* (2019) in Ethiopia and higher than 8.7% reported by Hasan *et al*. (2018) (2018) in Kanam local government area of Plateau State, Nigeria.

Table 2: Location based prevalence of salmonella isolates in fresh milk

Location	Number of samples	Number of positive samples	Prevalence (%)	χ^2	p-value
Behind road safety	16	4	25	1.21	0.88^{NS}
Katangare	16	3	18.75		
Gyabiya	16	3	18.75		
Behind house of assembly quarters	16	2	12.5		
Metropolis	16	2	12.5		
Total	**80**	**14**	**87.5**		

Table 2 shows the location based prevalence of salmonella isolates in fresh milk. The study revealed an overall prevalence of 87.5% as shown in Tables 2. 25%, 18.75%, 18.75%, 12.5% and 12.5% of the overall prevalence were recorded in Behind road safety, Katangare, Gyabiya, Behind house of assembly quarters and Metropolis respectively. The overall prevalence of 87.5% revealed by this study is higher than the 61.8% reported by Arii *et al*. (2002). However, it is on the lower side when compared with the 92.6% and 90.0% reported by Kuvandik *et al*. (2019) respectively. These variations may be explained by the differences in location, hygienic practices and the availability of potable water.

Table 3: Location based prevalence of salmonella isolates in Nono

Location	Number of samples	Number of positive samples	Prevalence (%)	$\chi 2$	p-value
Investment	20	2	10	0.38	0.95^{NS}
New market	20	2	10		
Laraba market	20	3	15		
Metropolis	20	2	10		
Total	**80**	**09**	**45**		

Table 3 shows the location-based prevalence of salmonella isolates in Nono. The study revealed an overall prevalence of 45.0% as shown in Tables 3. 10%, 10%, 15%, and 10% of the overall prevalence were recorded in Investment, New market, Laraba market, and Metropolis respectively. The result is an agreement with studies by Arii *et al*. (2002) who worked on 384 dairy cows and reported the overall prevalence to be 41.6% with 43.1%, 41.0%, 42.1% and 40.0% in cheese, butter, milk and yoghurt, respectively.

Table 4: Location based prevalence of salmonella isolates in fura

Location	Number of samples	Number of positive samples	Prevalence (%)	χ^2	p-value
Investment	20	4	20	1.16	0.76^{NS}
New market	20	2	10		
Laraba market	20	3	15		
Metropolis	20	2	10		

Total	**80**	**11**	**55**

Table 4 shows the location-based prevalence of salmonella isolates in fura. The study revealed an overall prevalence of 55.0% as shown in Tables 4. 20%, 10%, 15%, and 10% of the overall prevalence were recorded in Investment, New market, Laraba market, and Metropolis respectively.It has been reported that the unclean hands of workers, poor quality of milk, unhygienic conditions of the manufacturing unit and water supplied for washing the utensils could be the source for accelerating bacterial contamination of fura beside the post manufacturing contamination. The prevalence of the salmonella observed in this study could be due to the fact that fura being a good nutritive medium enhanced the growth of bacteria contaminant in the fura investigated (Osterholm and Norgan, 2014)

Table 5: Coli form count for milk and milk products

Sample	Number of samples	Number of positive samples	Coli form count
Fresh milk	80	14	$3.95x10^6$
Nono	80	9	$5.51\ x10^5$
Fura	80	11	$4.64\ x10^5$
Total	**240**	**34**	**$14.10\ x10^5$**

The mean coli form count of the milk and milk additives from the two hundred and forty (240) samples ranged from $3.95x10^6$ in fresh milk to $5.51\ x10^5$each in nono. The coli form count of milk and milk additives in this study gives an overview of how fit the product is for consumption. The study also gives an insight into the overall sanitation level of the whole process and the hygienic status of the products as the acceptable limits of coliform counts in milk should be less than 100 cell/mL (Majowicz *et al.,* 2020)

CONCLUSION AND RECOMMENDATION

From the study conducted, it can be concluded that there was prevalence of salmonella species in raw milk and its products such as nono and fura in the study area and pose a considerably public health concern. It is therefore important that there should be implementation of hygienic measures during milking and manufacturing of dairy products to minimize the risk of human infections with Salmonella

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