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DETECTION OF ESCHERICHIA COLI O157:H7 FROM "FURA DA NONO" SOLD IN ZARIA, KADUNA STATE, NIGERIA

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ABSTRACT

'Fura da nono' is a commonly food drink in northern part of Nigeria that is mostly consumed by almost all group of people in the country to include both young, adult and aged ones. The research work was aimed at determining the prevalence of E.coli O157:H7 from 'fura da nono'. A total of 228 ready-to-take 'fura da nono' samples were collected from Samaru, Sabongari and Tudun-wada markets. Proximate composition was carried out to determine the % moisture, ash content, protein content, fat and carbohydrate of the sample. Total viable count was also carried out in order to determine the microbial load. Escherichia coli O157:H7 was isolated using cultural method and confirmed using latex agglutination test. The results obtained revealed that the 'fura da nono' samples collected from Tudun-wada market had a count of $1.07x10^8 \pm 62.91$ cfu/ml, followed by that collected from Tudun-wada market has the least count of $6.65x10^7 \pm 24.90$ cfu/ml. The prevalence of E.coli O157:H7 was found to be 0.88% (2/228). As such, the food drink 'fura da nono' used for the study can be contaminated with pathogenic microorganism such as E.coli O157:H7 whose detection in food poses a threat to the health of the consumers. Strict hygienic measures should be adhered to when preparing 'fura da nono'.

Key words: fura da nono', E.coli O157:H7, Zaria

INTRODUCTION

'Fura da Nono' (fermented milk cereal mix) is a highly nutritious food drink which is a two in one product consisting of "fura" made from millet and "nono" a fermented milk product similar to yoghurt. In the market, 'fura' is mixed with 'nono' in a bowl for consumers. Depending on the consistency, the product is used as food and a refreshing drink to both group of people including the young and elderly (Umoh *et al.*, 1988; Musliu and Aliyu, 2012).

In Nigeria, the methods of processing and selling cow's milk and its products such as 'fura da nono'

exposes these products to the danger of microbial contamination both from spoilage and pathogenic microorganisms. Raw milk or processed milk is a well-known medium that supports the growth of several microorganisms, which results in the contamination of the product as well poses a health threat to the consumers (Oliver *et al.*, 2005).

Cattle are considered to be one of the primary sources of *E. coli* O157:H7 worldwide, as numerous studies have shown that *E. coli* O157:H7 prevalence is widespread in dairy and beef animals, and can be found in, and around cattle in most parts of the world without causing any disease symptoms (Muhammed *et al.*, 2009). Transmission is via the faecal–oral route, and most illness has been through consumption of contaminated raw green vegetables, undercooked meat as well as milk and its products such as 'fura da nono' (Lim *et al.*, 2010).

MATERIAL AND METHOD

Sample collection and analysis.

A total number of 228 samples of ready-to-take 'fura da nono' samples were collected from three different markets in Zaria town, Kaduna State Nigeria (76 samples from each market). The samples were transported in an ice bag to the Department of Microbiology, Ahmadu Bello University Zaria for the analysis for the analysis. A volume of 25ml of each of the samples was added to 225ml of buffered peptone water to make 1:10 dilution and marked as stock solution. From the stock solution, 1ml was pipetted into a 9ml of buffered peptone water to make the second dilution (10^{-2}) . The process was repeated until a dilution of 10^{-5} was reached. A volume of 0.1ml was pipette onto the surface of plate count agar and spread through out the plate using a bent glass rod to ensure evenly distribution of the aliquot in duplicate (Yusuf *et al.*,2018). The procedure was done for all the samples collected.

Proximate Composition.

The proximate composition of each of the samples awas determined using the methods described by AOAC 2000.

Detection of *E.coli* O157:H7.

Each of the samples was streaked on to the surface of Eosin Methylene Blue agar to detect the presence of a lactose fermentor. The colonies that appeared greenish metallic sheen with dark centres were presumptively picked, stored in a nutrient agar slant and considered as strains of *E.coli* before further confirmed using conventional biochemical test (Edward and Oluwafemi, 2017). The stored organism was subjected to biochemical test such as Indole test, Citrate utilization test, MRVP test (MacWilliams, 2015).. Those isolates that were confirmed to be strains of *E.coli* based on the mentioned biochemicals were lebelled and used for the detection of *E.coli* O157:H7. The isolates were then subcultured on to the surface of Sorbitol MacConkey agar for the detection of non sorbitol fermenting organism. The isolate that appears colourless were presumptively considered to be strains of *E.coli* O157:H7. This isolate was further subcultered into nutrient broth for 24hr and subjected to latex agglutination test to confirm the organism. The latex agglutination test was carried out based on the manufacturer's instruction (*E.coli* O157 antiserum (Oxoid) provided in the kit and the organism was considered positive when agglutination was seen in all the two wells lebelled for O157 and H7 (Yusuf *et al.*, 2018).

RESULT AND DISCUSSION

Proximate Composition of 'Fura da Nono' sample

The average proximate composition of 'fura da nono' as presented in Table 4.1 reavealed the average moisture content of (81.78%), ash content (0.59%), fat (4.97%), crude protein (3.77%) and carbohydrate (8.89%).

Total viable count of the 'fura da nono' samples

The research has shown the average count for samples collected from Samaru market to be $1.59 \times 10^8 \pm 62.91$ SD (cfu/mL), that of Tudun-Wada to be $1.07 \times 10^8 \pm 44.94$ SD (cfu/mL) and the count for Sabongari was found to be $6.65 \times 10^8 \pm 24.90$ SD (cfu/mL). The highest count was seen in the samples collected from Samaru market.

Occurance of E.coli O157:H7 in the'fura da nono' samples

The result revealed that out of the total number of 228 samples collected from the three sampling areas to include Samaru, Tudun-wada and Sabongari markets, 20 *E.coli* isolates were obtained and only 2 isolates were confirmed to be strains of *E.coli* O157:H7 based on latex agglutination test as presented in Plate I. The occurance of *E.coli* O157:H7 in Samaru market and Sabongari market was found to be 1.13% each, while zero prevalence in Tudun-wada market was observed as no *E.coli* O157:H7 was isolated from the samples collected in the market.

Parameter	Range of value (%)	Percentage (%)	
Moisture	78.90-84.78	9-84.78 81.78	
Ash	0.56-0.62	0.59	
Crude lipid	4.60-5.32	4.97	
Crude Protein	3.35-4.20	3.77	
Carbohydrate	5.18-12.59	8.89	

Table 1 Proximate composition of 'fura da nono' samples

Table 2 Total Viable Count of the 'Fura da Nono' samples

Market	No. of Samples Collected Average Count ±SD(cfu/	
Samaru	76	1.59 x 10 ⁸ ±62.91
Tudun-wada	76	1.07 x 10 ⁸ ±44.94

Sabongari	76	6.65 x 10 ⁷ ±24.90
TOTAL	228	

Market	No. of Samples Collected	No. of Samples with <i>E.coli</i> (%)	No. of Confirmed <i>E.coli</i> O157:H7	Frequency of occurance (%) of <i>E.coli</i> 0157:H7
Samaru	76	8 (10.52)	1	1.13
Tudun-wada	76	7 (9.21)	0	0
Sabongari	76	5 (6.57)	1	1.13
TOTAL	228	20(8.77)	2	0.88
χ^2 1.009	<i>P</i> -value	0.604		

Table 4.3 Frequency of occurance of *E coli* O157:H7 in 'Fura da Nono'

RESULT AND DISCUSSION

The proximate composition for a randomly selected sample indicates that the 'fura da nono' sample has a moisture content of 81.78%. The moisture content is a measure of water content of the product which accounts for the texture of the food drink. The value obtained in the study might be as a result of water added during the preparation as some of the sellers do add water to the mixture. The value from the study lower than the value of 84.92% obtained by Onoriode and Camilus (2017) in Port Harcourt, and higher than the value of 75% obtained by Nneoma (2019) in Abuja. The differences in the value can be attributed to the methods employed in the preparation of the product ('fura da nono'), season of the year at which the research was carried out.

The ash content indicates that the product was rich in minerals which make it a suitable environment for microbial growth. The ash content was found to be 0.59% which was similar to the value obtained by Nneoma (2019) in Abuja, and higher than the value of 0.48% obtained by Onoriode and Camilus (2017) in Port Harcourt. The differences in the values may be due to production processes as some Fulani women selling the product due heat the 'nono' at high temperature and this may cause the destruction of some essential components of the 'nono'.

The crude lipid value obtained from the study was 4.97%, and this indicates that the 'fura da nono' used contains enough fatty acid that can be channeled to metabolic activities through the processes of beta-oxidation. The value was higher than the value of 3.00 obtained by Nneoma (2019), and also lower than the value of 3.09 obtained in Port Harcourt by Onoriode and Camilus (2017). The protein content was found to be 3.77%, which indicate that 'fura da nono' is rich in nitrogen required for both the growth of human cells as well as the pathogenic organisms. As a result of the location, feeds of the animal and also the season of the year during which the study was carried out, the value was lower than that obtained in Abuja by Nneoma (2019) and higher than that obtained by Onoriode and Camilus (2017) in Port Harcourt. The carbohydrate value was found to be 8.89%, and it indicates the suitability of 'fura da nono' to be rich in sugars that are use for energy generation. The value obtained was lower than 14% obtained in Abuja by Nneoma (2019), and higher than the value obtained by Onoriode and Camilus (2017) of 7.70% in Port Harcourt. The differences in the values may be due to geographical location, production processes, sample size and season of the year as usually the number of the samples used during the research varied.

Also, the study reveals that 'Fura da Nono' sold locally in Zaria has a viable count higher than the standards set by both World Health Organization (WHO) and National Agency for Food and Drugs Administration and Control (NADAC) which suggests a value not exceeding $3x10^3$ cfu/ml. The counts range from $1.59x10^8\pm62.91$ SD cfu/ml in Samaru market, $6.65x10^7\pm24.90$ SD cfu/ml in Sabongari market and $1.07x10^8\pm44.94$ SD cfu/ml in Tudun-wada maket. The high counts seen may be due to the unhygienic practices during milking by those selling and handling the product, unclean environment where milking activity is carried out and probably failure to use portable water for cleaning the utensils used during milking and selling of the product.

Also, the environment where the consumers purchase the product and the fact that cattles do passbye the side of this areas and during the passage, the cow's dung might be carried to the area by air inform of aerosols. This shows the role of the environment in the contamination of the local product as suggested by Abid *et al.* (2009).

The prevalence of *E.coli* O157:H7 was found to be 0.88% (2/228) at χ^2 1.009, *P*-value0.604 which is not significant.

CONCLUSION

The value obtained was lower than that of 18% reported by Edia-Asuke and Sabah (2014) in Zaria, higher than the value of 1.3% also obtained by Ayuba (2015) in Zaria and also higher than that was obtained by Itelina and Agina (2010), a prevalence of 0.71% was seen in locally fermented milk (Nono) in Plateau State, Nigeria. The value obtained also was lower than the value obtained by Rubena *et al.*, (2013) value of 4.5% prevalence from Nasarawa State and also lower than what was obtained by Dahiru *et al* (2008) from Kano (value of 53%). This value obtained can be attributed to factors such as geographical location, sample size, nature of samples, methods employed as well as the time period during which the research was carried out; the research was carried out during rainy season.

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