

Full-text available at http://www.ajbrui.com & http://www.bioline.br/md

Received: January, 2006 Accepted (Revised): April, 2006 Published May 2006 Short communication

# **Extent** of **Microbial Contamination of Sausages sold** in two Nigerian cities

# Oluwafemi F. and Simisaye, M.T.

Department of Microbiology , University of Agriculture, P.M.B.2240,Abeokuta.

# ABSTRACT

Three shops were randomly selected in Abeokuta (South-West Nigeria) and Benin-City (South-South Nigeria) for the purchase of sausages which were then screened for microbial contamination. For the Abeokuta sausage samples the total aerobic counts ranged from 2.06-2.80 x  $10^6$ cfu/g; Staphylococcus aureus count :1.1- $1.47 \ x \ 10^6 \ cfu/g$ ; Enterobacteriaceae count: 1.57- 2.17 x  $10^6$  cfu/g ; lactic acid bacteria count(LAB) count :1.70 – 2.33 x  $10^6$  cfu/g. With respect to the sample from Benin-City, the total aerobic count ranged from  $3.54 \times 10^6$  cfu/g; S. aureus count: 1.8 x10<sup>5</sup>- 2 x 10<sup>7</sup>; Enterobacteriaceae count: 5.09 x 10<sup>8</sup> cfu/g; LAB count :1.3 -4.6 x 10<sup>8</sup> cfu/g. Probable organisms isolated from sausages sold in Abeokuta were E. coli, Streptococcus sp., Clostridium sp., Klebsiella sp., Shigella sp., Pseudomonas sp., Lactobacillus sp., and S. aureus while those organisms isolated from sausages sold in Benin-City include Salmonella, Proteus, Shigella, S. aureus, Klebsiella and Lactobacillus sp. Most of the sausages sampled were therefore considered to pose health risk to consumers, making it imperative to institute not only sanitary measures during processing, storage and marketing but also to ensure steady source of power supply. (Afr. J. Biomed. Res. 9:133 - 136 , May 2006)

Keywords: microbial contamination, sausages, Nigeria.

\*Address for Correspondence (e-mail): <u>foluwafemi2000@yahoo.co.uk</u>

African Index Medicus (WHO), CAB Abstracts, Global Health Abstracts, Asian Science Index, Index Veterinarius

# INTRODUCTION

Food-borne illnesses in human beings due to bacterial pathogens and their toxins are well documented worldwide (Hazariwala et al, (2002); Lin et al, (2002). Food-borne illness imposes a substantial economic and quality of life burden on society by way of acute morbidity and chronic sequela (Duff et al, 2003).Staphylococcal intoxication is a leading cause of food-borne intoxication and enterotoxigenic Staphylococcus strains have been isolated from foods implicated in illnesses (Adesiyun, 1995; Cencil et al, 2003). Salmonella spp has been reported by the United States Department of Agriculture Food Safety and Inspection Service (FSIS) as one of the most common causes of food-borne illness associated with meat and poultry products. Yersinia enterocolitica is a salt-tolerant, pschrotrophic rod that is widely distributed in nature, in aquatic and animal reservoir for human pathogenic strains (Hillers et al, 2003). In 1998, there was an increased number of reported cases of illness due to Listeria monocytogenes which the Centers for Disease Control and Prevention as well as state and local health departments in the U.S. attributed to the consumption of cooked hot dogs and deli meats (FSIS, 1999). Shehu and Adesiyun (1990) reported 39.5% of milk to be positive for E. coli. Enterotoxigenic Escherichia coli has been involved in food-borne illness and recovered from various food types, processed or raw (Firstenberg and Sullivan, 1997).

Campylobacter jejuni and C. coli, often responsible for causing Campylobacter enteritis (campylobacteriosis) in humans ,the most common bacterial form of acute infective diarrhea, are the most commonly reported bacterial cause of food-borne infections in the United States (Skirrow and Blaser, 1995; Altekruse et al, 1999). A number of foods in Nigeria have been reported to have high incidence of bacteria (Adesiyun, 1995), however, there is little /scanty information about the extent of microbial contamination of sausages sold in Nigerian supermarkets. The fact cannot be overemphasized that raw or pre-processed foods sold in supermarkets pose a direct health hazard to consumers if they contain an infective dose of pathogens or toxic levels of their toxins.

The purpose of this study was to evaluate the microbial contamination occurring on sausages at retail outlets in order to facilitate the assessment of

microbiological risks associated with them. The microbial estimates determined were total viable counts, *Staphylococcus aureus* counts, Enterobacteriaceae, pychrophilles and lactic acid bacteria in respect to microbiological quality of the sausages.

#### MATERIALS AND METHODS

**Source and collection of samples:** Three samples collected in Benin-City were from the University of Benin supermarket at Ugbowo, K-supermarket in Saponba and L-stores in Ring Road area. The Abeokuta samples (3) were from Ita –eko, Ibara and Onikolobo. The samples were put in sterile plastic containers and transferred to the laboratory ice-cooled within 2h of collection. The samples were collected from these sites randomly at the beginning, middle and at the end of each city

**Bacteriology: Total and Differential Counts:** One gram of each sausage sample was weighed into a mortar (that had been previously sterilized at  $160^{\circ}$ C for 1h) and ground with a sterile pestle until it became smooth and 9 ml of sterile distilled water was poured into the mortar. This was transferred to a test-tube followed by serial dilution up to  $10^{-7}$  dilution.

To determine total viable counts, 1 ml of each of  $10^{-5}$  and  $10^{-7}$  dilutions were plated on nutrient agar plates in triplicates. The plates were incubated at  $37^{\circ}$  C for 24hours. The same procedure was repeated for *Staphylococcus aureus* count, enterobacteriaceae count, lactic acid bacterial count on mannitol salt agar, MacConkey agar and De Man Rogosa Sharpe (MRS) agar respectively. Pschrophyllic count done for all samples in Benin-City. They were incubated on nutrient agar plates at  $4^{\circ}$  C for 48 h. For MRS agar, the plates were incubated at  $37^{\circ}$  C for 48-72 hours. Anaerobic count was done by incubating plates in an anaerobic jar for 24 h.

**Identification of Isolates:** The isolates obtained on plate counts were identified based on established conventional cultural, morphological and biochemical characterizations (Encinas *et al.*, 1996)

**Statistical Analysis:** All data were analyzed using the general linear model procedures of SAS and Analysis of Variance (ANOVA).

#### RESULTS

Mean total viable count, *Staphylococcus aureus* counts, enterobacteriaceae count, psychrophillic count, LAB counts are shown in Table 1 for sausages from Abeokuta and Table 2 for microbial counts of sausages from Benin-City. The three centers sampled in Abeokuta had total viable counts that were between 2.06-2.87x 10  $^{6}$ cfu/g (Table 1).This is an acceptable range for total viable count of organisms by the Public Health Laboratory Service (PHLS, 1996) but this was not the case for one location sampled in Benin-City. The sample from Ring Road area had total viable count of 4.8 x 10  $^{8}$ cfu/g (Table 2) which was above the PHLS approved (10 $^{6}$ -10  $^{7}$ cfu/g).

### Table1

Microbial counts of sausage samples(cfu/g) from three locations in Abeokuta

	Locations		
Counts	Onikolobo	Ita-eko	Ibara
Aerobic count	$2.5 \text{ x} 10^6$	$2.06 \times 10^6$	$2.87 \times 10^6$
S.aureus	$1.3 \text{ x} 10^6$	$1.1 \text{ x} 10^6$	$1.47 \text{ x} 10^6$
LAB count	$2.13 \times 10^{6}$	$1.7 \text{ x} 10^6$	$2.33 \times 10^6$
Coliform count	$1.7 \text{ x} 10^{6}$	$1.57 \text{ x} 10^6$	$2.17 \text{ x} 10^6$

#### Table 2.

Microbial counts of sausage samples (cfu/g) from three stores in Benin-City.

	Locations		
Counts	<b>Ring road</b>	Saponba	Ugbowo
		road	area
Aerobic count	$4.0 \text{ x} 10^8$	$3.5 \times 10^6$	$3.72 \text{ x}10^7$
S.aureus	$1.8 \text{ x} 10^5$	$3.3 \times 10^5$	$2.2 \text{ x} 10^7$
LAB count	$1.3 \text{ x} 10^4$	$3.7 \text{ x} 10^5$	$4.6  ext{ x10}^4$
<b>Coliform count</b>	9.6 x10 <sup>8</sup>	$5.0 \text{ x} 10^4$	$5.2 \text{ x} 10^7$
Psychrophillic	$3.0 \text{ x} 10^5$	$5.6 \times 10^6$	$4.1 \text{ x} 10^6$
count			

The enterobacteriaceae counts for all samples obtained from Abeokuta and Benin-City were above the limit specified by the British Standard Institute (BSI, 1991,1993) except samples collected from Saponba area of Benin-City and it was observed also that this was the sample with the highest LAB count.Coliform counts from Abeokuta were in the range of  $1.57 \times 10^6$ -2.17 x  $10^6$  while those from Benin-City were between  $5.0 \times 10^4$  -9.6 x  $10^8$  Although the specific coliform organisms were not cultured in this work, it is not unlikely with the high

counts that there will be some toxigenic strains of *E. coli, Salmonella* spp., *Campylobacter* and *Klebsiella* spp. The *S. aureus* count in all samples were within  $10^5-10^6$  cfu/g (Tables 1 and 2) except samples from one location (Ring Road area) that had 2.2 x  $10^7$  cfu/g which was significantly different from all samples and the approved value by PHLS and BSI.

Lactic acid bacteria(LAB) counts were highest in two samples-one from Benin and the other from Abeokuta (Tables 1 &2).These values were significantly(P< 0.005) higher than all other samples. Organisms isolated also indicated the presence of Lactobacillus species.

Probable isolates of microorganisms from sausages in Abeokuta were E. coli, Staphylococcus aureus, Streptococcus sp., Clostridium sp., Klebsiella sp., Shigella sp., Pseudomonas sp, Lactobacillus sp. In Benin-City, Salmonella sp., Proteus sp., Shigella *Staphylococcus* aureus, Klebsiella sp., sp. ,Lactobacillus sp. were isolated. All these microorganisms have been implicated in food-borne illnesses (Firstenberg and Sullivan, 1997; Hazariwala, 2002).

# DISCUSSION

The mortality associated with these pathogens is not well documented in Nigeria however, the economic impact of these illnesses is important (absenteeism, medical care, investigations, withdrawal of the contaminated products, loss of confidence in products). The high total viable counts from area such as the Ring Road area could be attributed to improper cleaning and sanitizing of equipment and poor employee hygiene within the store and more importantly due to erratic power supply in this area. The enterobacteriaceae counts for all samples obtained from Abeokuta and Benin-City were above the limit specified by the British Standard Institute (BSI, 1993) except samples collected from Saponba area of Benin-City and it was observed also that this was the sample with the highest LAB count. The BSI specified that enterobbacteriaceae count greater than 10<sup>4</sup>cfu/g is considered unsatisfactory. Adesiyun (1994)demonstrated gross contamination with. S. aureus and E. coli of preprocessed bovine milk in Trinidad. Shehu and Adesiyun (1990) reported E.coli in fermented Nigerian milk. Although the specific coliform organisms were not cultured in this work, it is not unlikely with the high counts that there will be some

toxigenic strains of E .coli, Salmonella spp., Campylobacter and Klebsiella spp. Food-borne salmonellosis has been associated with consumption of various foods especially meat and poultry products (Adesiyun, 1993). The high enterobacteriaceae counts is an indication of potential microbial contamination during processing, distribution and storage. Their presence in large numbers in food indicates inadequate processing/or recontamination due to cross contamination by raw materials, dirty equipment or hygienic handling (Ikeme, 1990). poor Enterobacteriaceae occur as normal flora of the intestinal tract. They are widely distributed in nature and this account for their presence in sausage. However, E. coli and Enterobacter spp have the potential to cause diarrhea (Volk, 1982). According to Zhao et al., (2003). The process of freezing reduces the numbers of some coliforms such as Campylobacter jejuni.

According to Kuku (1985),the presence of *S.aureus* could be as a result of it being a common organism on the skin, hands and boil and hence their presence in sausage may be as a result of contamination due to handling, processing, transportation and storage. Its presence in high numbers is a good indication of poor hygiene and temperature control. The presence of Staphylococci in high numbers in cured meat may indicate the presence of enterotoxin –producing strains of *S.aureus* (AS/NZS, 1999), thus the data generated are of great importance to inform public health authorities, to detect food-borne diseases outbreaks early and to implement and evaluate food safety programmes.

# REFERENCES

Adesiyun, A. A. (1993). Prevalence of *Listeria* spp., *Campylobacter* spp., *Salmonella* spp., and toxigenic *Escherichia coli* on meat and seafoods in Trinidad. Food Microbiol.10:345-403.

**Adesiyun A. A (1995):** .Bacteriologic quality of some Trinidadian ready-to-consume foods and drinks and possible health risks to consumers. J. of Food Protection 58(3):651-655).

Altekruse, S. F., Stern, N. J. Fields, P. I. and Swerdlow, D. L. (1999): *Campylobacter jejuni* – an emerging foodborne pathogen. Emerg. Infect. Dis.5(1)

**Autralian/New Zealand standards methods for Food Microbiology. (1999):** Guide to determine the equivalence of Food Microbiology nest method. AS/NZS 4659.

**British Standards Institute (1993):** Microbiological examination of food and animal feedingstuff.Enumeration

of Enterobacteriaceae.London.BSI (5763:part 10:1993(ISO 7402:1993).

**Cencil, G. B. T., Karama, M., Rossitto, P.V., Morgante, R. A. and Cullor, J. S. (2003).** Enterotoxin production by Staphylococcus aureus isolated from mastitic cows. Journal of Food Protection 66(9):1693-1696.

**Duff,S.B.,Scott,E.A.,Mafilios,M.S.,Todd,E.C.,Krilov,L.R.,G** eddes,A.M.andAckerman,S.J.(2003).Cost-effectiveness of a targeted disinfection program in household kitchens to prevent food-borne illnesses in the United States, Canada and the United Kingdom.Journal of food protection bb(II):2103-2115.

Encinas, J. P., Sanz, G. J., Garcia, A. M. R. Otero, A.(1996): Evaluation of different systems for the identification of Bacillus strains isolated from Spainish fermented sausages. Meat Science 42(2):127-131.

**Firstenberg, E. R. and Sullivan, N. M. (1997):** EZ Coli Rapid Detection System: a rapid method for the detection of *Escherichia coli* 0157 in milk and other foods. Journal of Food Protection 60(3):219-225.

**FSIS** (1998): Contamination with microorganisms, pathogen Food Safety and Inspection Service, Washington, D.C. Fed. Regist. 63: 1800-1802.

**FSIS (1999):** FSIS action plan for addressing *Listeria monocytogenes.* U.S. Department of Agriculture, Food Safety and Inspection Service, Washington, D, C.

Hazariwala, A. Sanders, Q. Hudson, C. R., Hofacre, C., Thayer, S. G. and Maurer, J. J. (2002): Distribution of Staphylococci enterotoxin genes among *Staphylococcus aureus* isolates from poultry and humans with invasive staphylococcal disease. Avian Diseases 46(1):132-136

Hillers, V. N. ,Medeiros, L., Kendall, P., Chen, G. and Di Mascola, S.(2003): Consumer food-handling behaviors associated with prevention of 13 foodborne illnesses. J Food Prot. 2003 Oct;66(10):1893-9.

**Ikeme, A. I. (1990):** Fermented sausage-dry and semi-dry. In Meat Science and Technology. 1<sup>st</sup> edition. The African fep publishers Limited Nigeria. Pp 210.

**Kuku, F. O.** (1985): Soilage of fruits, vegetable and tuber crop. Nigeria food Journal 2:1-3.

Shehu, L. M. and Adesiyun, A. A.(1990): Characteristics of strains of *Escherichia coli* isolated from locally-fermented milk (Nono) in Zaria, Nigeria.J. Food Protection 53:574-577.

**Skirrow, M B., and Blaser M J.(1995)**: *Campylobacte rjejun*i. Infections of the gastrointestinal tract. Blaser, M. J.,Smith, P.D., Ravdin, J. I., Greenberg, H. B. and Guerrant, R. L.(ed).Raven Press, Ltd, New York, p.825.

**Volk, W. A. (1982):** Essential of Medical Microbiology. 2<sup>nd</sup> ed .J. B. Lippincott Company, Philadelphia, pp. 369-377. **Zhao, T., Ezeike, G. O I., Doyle, M .P., Hung, Y. C. and Howell, R.S (2003)**: Reduction of Campylobacter jejuni on poultry by low-temperature treatment. J. Food Protection 66(4):652-6555.