Full Length Research Paper

The materialization of some enteropathogenic bacteria in simply and completely processed foods in Nigeria

Hammed Olla, Harshanti O. and Abubakar Bukar

Department of Medicine, Faculty of Sciences, Al-Hikmah University, Ilorin, Kwara, Nigeria.

Twelve (12) different food types consisting of six (6) fully processed and six (6) minimally processed ready – to – eat foods sourced from different areas of Kano metropolis were analysed for the presence of enteropathogenic bacteria from June – August, 2009. Enumeration of aerobic mesophilic bacteria using the aerobic plate count (APC) revealed that the mean count for all foods examined except zobo drink, exceeded the maximum acceptable limit (10^5 cfu/g/ml) set by the Food and Agricultural Organization (FAO) of the United Nations. The highest mean count of 1.60×10^7 cfu/g was obtained for tomato (minimally processed food), while the least count of 2.49×10^4 cfu/ml was recorded for zobo drink (fully processed food). Out of a total of 60 food samples analysed, *Escherichia coli* recorded the highest frequency of occurrence of 24 (46.6%), followed by *V. cholerae* with 15 (25.0%) while *Salmonella typhi* recorded the least occurrence rate of 6 (10.0%). Overall, the fully processed foods were observed to be less contaminated with enteropathogenic bacteria than the minimally processed foods. The results indicated that most of the ready – to – eat food samples examined in this study did not meet bacteriological quality standards. The implications of the results on human and environmental health are discussed.

Key words: Enteropathogenic, bacteria, ready - to - eat, food, Kano

INTRODUCTION

Food is any substance usually composed of carbohydrate, fats, proteins and water that can be eaten and/ or drunk by an animal or human for nutrition or pleasure (Wikipedia, 2007). Practically all the food we purchase or grow, be it fruit, vegetable, meat, cereal, meat, home made drinks and juices or dairy products habour a variety of microorganisms. This is not surprising when one considers that bacteria and fungi are ubiquitous and are especially plentiful in soil and around us (air) and could easily contaminate foods (www.microbiologyprocedure.com/food-microbiology/, 2009). From the microbial perspective, food can be view-ed as a fertile ecosystem in which these organisms vie for their nutrients (Nester et al., 2004). Micro-organisms on foods are not always undesirable because sometimes their growth results in a more pleasant taste or texture. For example, food manufacturers purposely encourage some microorganisms to flourish and produce desired flavours in some foods. However, contamination and growth of pathogens such as *Staphylococcus aureus, Salmonella* species*, Bacillus* species, *Pseudomonas aeruginosa, Clostridium* species, *Vibrio cholerae* and *Escherischia coli* can result in perceptible changes in quality of a food. Such foods can transmit a wide range of diseases in a condition termed food infection, where the food serves as a vehicle for the transfer of the pathogen to the consumer, in whom the pathogen grow and causes disease (Murray, 2003). Another condition that might arise is food intoxication, where the pathogens grow in the food and produce toxins that can then affect the consumer of the food (Prescott et al., 2008).

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*Correspondence author: E-mail: Ali_hammed25@yahoo.com

The occurrence of pathogenic microorganisms has always been attributed to several factors, which include contamination through water, soil, food processing equipments, food contact surfaces and most importantly food handlers (Shamsuddeen and Ameh, 2008; Kawo and Abdulmumin, 2009; Aboloma, 2008). Improper handling of food is responsible for most cases of food-borne diseases and intoxication, including improper use of preparation and storage temperatures, cross contamination and poor personal hygiene (Goncalves, 1998). When food handlers do not practice proper personal hygiene or correct food preparation, they may become vehicles for microorganisms, through their hands, mouth, skin among others (Silva et al., 2003: Bukar et al., 2009).

A high number of foods sold in our communities are contaminated to a large extent with pathogenic micro-organisms. Foods sold near polluted environments are prone to contamination by pathogenic microorganisms. Documented evidences have continued to link patho-genic microorganisms in food to incidences of food borne diseases and intoxications. The numbers of deaths due to pathogens that cause acute gastroenteritis in the United States is 1,381 of which 67% are attributed to food – borne transmission. Bacteria accounts for 72% and five pathogens account for over 90% of estimated food related deaths of which *Salmonella* has the highest percentage (31.0%) and *E. coli* had the lowest (3.0%), respectively (Paul et al., 2009). In a study by Udo et al. (2009) conducted at Calabar, South – South region of Nigeria, potential pathogens in ready – to – eat food were isolated, predominantly members of the family Entero-bacteriaceae. The isolation of *Salmonella* species in the study is very disturbing particularly as these samples were obtained from large wedding party receptions. In the study, a total of 150 salad food samples were analysed and in all, 85 (56.67%) were contaminated with bacteria; *E. coli* had 21 (24.71%), *Klebsiella aerogenes* 11 (12.84%) and *Salmonella typhimurium* 3 (3.53%). In ano-ther research, Oyeyi and Lum – Nwi (2008) reported an occurrence of *E. coli* in 27.7% of samples of street – vended foods in Bayero University campuses, Kano – Nigeria.

In developing countries such as Nigeria, there are serious concerns about sanitation of ready – to – eat foods, particularly as potable water is seldom available at preparation venues and fast food stands and also most food handlers lack basic knowledge of proper personal and environmental hygiene. It is in view of this that this study was conducted to evaluate the bacteriological quality of some ready – to – eat foods vis – a – vis enumeration of aerobic mesophilic bacteria and also determine the incidence of some enteropathogenic bacteria on the foods in Kano metropolis with a view to providing additional data on the incidence of such pathogenic microorganisms for necessary action on the part of food sellers, consumers and authorities.

MATERIALS AND METHODS

Sampling sites and description of some of the foods

A total of 60 samples comprising of twelve (12) different food types collected in 5 batches each were bacteriologically analysed. Sampling was carried out between June and August, 2009. Different foods, which include six (6) fully processed ready – to – eat (Awara, fried Groundnut, Rice and beans dish, Fura, Zobo drink and Bread) and six (6) minimally processed ready – to – eat (Tomato, Cabbage, Lettuce, Carrot and Ginger, Kunun zaki drinks) were sourced from different areas in Kano metropolis.

First category was purchased from Kabuga market near Bayero University, Kano old campus in Gwale Local Government Area (LGA), while the second category was purchased from Zangeru Fadama irrigation site, Kwakwaci, Fagge LGA of Kano State except all the drinks that were also purchased from Kabuga market.

Descriptions of some of the local foods are: Awara (fried soya bean cake), Fura (boiled millet ball to be mixed with cow milk), Zobo (drink made from *Hibiscus sabdariffa* calyxes), Ginger (drink made from water and grinded ginger rhizomes), Kunun zaki (a millet drink).

Sample Collection

Samples were collected in sterile polythene bag directly from sellers (vegetables), hawkers and food stalls (the other foods) and immediately brought to the laboratory for analysis.

Bacteriological analyses of the samples

Enumeration of aerobic mesophilic bacteria

Samples were prepared according to the method of FAO (1979). For solid samples, twenty five grams (25 g) of each sample was weighed and homogenized by blending in 225 ml of sterile buffered peptone water at 15,000 – 20,000 rpm, while for the liquid samples, 1 ml was introduced into 9 ml of the buffered peptone water in a test tube. Each was labeled as 1:10 (10^{-1}) dilution. One milliliter (1 ml) from the 10^{-1} dilution was added to 9 ml of sterile buffered peptone water and serially diluted to four other test tubes labeled (10^{-2} -10^{-5}). The procedure was carried out for each of the sample. One milliliter (1 ml) aliquot of each dilution was pipetted and added to appropriately labeled sterile Petri dish into which warm (45°C) nutrient agar (NA) was added and swirled so that the aliquot is evenly distributed, allowed to gel and the plates were then incubated aerobically at 37°C for 24 h and then observed. All discrete colonies were counted where possible and expressed in colony forming units/gram (cfu/g) for solid foods and colony forming units/milliliter (cfu/ml) of liquid food sample.

Isolation and characterization of Isolates

Bacterial isolates were isolated and characterized using standard procedures described by Cheesebrough (2000) for *E. coli* and *Salmonella typhi* and Buchanan and Gibbons (1974) for *V. cholerae*.

RESULTS AND DISCUSSION

Table 1 presents results of aerobic plate counts of the different foods analysed. It could be observed that tomato

Table 1: Range and mean bacterial counts (APC) of the food samples examined

S/N	Food types	No of samples examined	Range of bacterial count (cfu/g/ml)	Mean (cfu/g/ml)
Fully processed foods				
1	Awara	5	$3.20 \times 10^5 - 2.96 \times 10^5$	5.80×10^5
2	Fried Groundnut	5	$7.90 \times 10^3 - 1.72 \times 10^5$	2.87×10^5
3	Rice and Beans dish	5	$5.50 \times 10^3 - 1.20 \times 10^5$	3.00×10^5
4	Fura	5	$3.00 \times 10^4 - 2.24 \times 10^6$	2.65×10^6
5	Bread	5	$<30 - 2.64 \times 10^5$	4.20×10^5
6	Zobo drink	5	$<30 - 1.24 \times 10^4$	2.49×10^4
Minimally processed foods				
7	Tomato	5	$6.00 \times 10^3 - 1.32 \times 10^7$	1.60×10^7
8	Cabbage	5	$<30 - 2.80 \times 10^6$	3.40×10^6
9	Ginger drink	5	$<30 - 1.76 \times 10^6$	1.91×10^6
10	Lettuce	5	$< 30 – 1.40 \times 10^6$	1.75×10^6
11	Carrot	5	$5.60 \times 10^3 – 1.20 \times 10^6$	1.68×10^6
12	Kunun zaki drink	5	$< 30 – 9.80 \times 10^4$	1.01×10^5
	Total	**60**		

(a fruit vegetable) had the highest mean bacterial count of 1.60×10^7 cfu/g while Zobo (a drink) had the lowest count of 2.49×10^4 cfu/g/ml. Overall, the result shows that all the foods examined except Zobo drink had mean counts above 10^5 cfu/g/ml. According to Food and Agricultural Organization (FAO, 1979), standard limit for aerobic mesophilic bacterial count should be less than 10^5 cfu/g/ml. The high bacterial count observed in this study might be attributed to factors such as the environment, which include exposure of the foods to air, soil; type of water used in processing of the food; post production operations and personal hygiene of the food handlers (Kawo and Abdulmumin, 2009; Aboloma, 2008; Wada – kura et al., 2009; Sagoo et al., 2003) . Exposure of the foods to air or dust at the point of sale is likely to increase the counts of the bacteria as virtually most of the bacteria are carried in aerosols by dust and air (Food and Drug Administration, 2009). Foods such as vegetables could have been contaminated from the farm (Parry and Pawsey, 1984) and as observed in this study, water used for the irrigation of vegetables examined in this study was highly polluted. High counts in the drinks and other foods could be attributed to poor handling practices during production, as handlers sometime dip their hands into the containers while making the drinks and in the case of solid foods such as fura, bread and fried groundnut, etc, handlers' hands play a role in contaminating them with high counts, as a lot post processing handling takes place during their production. For rice and beans dish, the plate containing the food is an important vehicle of transmission as all the plates for the day sometimes are washed in one change of water throughout the day.

The occurrence of *E. coli, S. typhi* and *Vibrio cholerae* in all the foods examined is presented in Table 2. Of the total of 60 samples examined, 28 (46.6%) were positive

for *E. coli,* out of which the fully and minimally processed foods had 14 (25.0%) each. Boiled rice and beans dish and lettuce had the highest percentage of 5 (100%), followed by fried groundnut with 4 (80.0%) and tomato, cabbage, fura, bread, carrot and Kunun zaki each with 2 (40.0%). Awara was negative for *E. coli.* The high occurrence rate of 46.6% of *E. coli* recorded in this study is contrary to 2.0% reported by Mosupye and von Holy (1999) in ready – to – eat street vended foods in Johannesburg. The result is still higher than the 24.7% for ready – to – eat salad reported by Udo et al. (2009) in Calabar, Nigeria. Oyeyi and Lum – Nwi (2008) reported *E. coli* incidence of 27.7% in street vended foods in Bayero University campuses, Kano, Nigeria. The high occurrence of *E. coli* in the present study might be attributed to the diverse types of foods studied in addition to the source locations, an open market and vegetables sourced from heavily polluted irrigation site. The contamination of virtually all the food samples with *E. coli* might be attributed to use of recent feacally contaminated water in the food preparations or from the farms in the case of vegetables and also to the activities of food handlers. In a report by Bukar et al. (2009), 5 (10.0%) out of 50 food handlers in three small – scale food industries in Kano metropolis investigated carried *E. coli* on their hands. This percentage could easily cross – contaminate a whole production batch unnoticed. Ironically, most food handlers do not practice good personal hygiene and do not follow good manufacturing practices, which could reduce the occurrence of such bacteria in foods (Wada-kura et al., 2008; Bukar et al., 2009; Kawo and Abdulmumin, 2009).

V. cholerae recorded an occurrence of 15 (25.0%) out of 60 food samples examined. Fully processed foods had 0 (0%) occurrence of *V. cholerae* as compared to

Table 2: Occurrence of *E. coli, Salmonella typhi* and *Vibrio cholerae* in the fully and minimally processed ready – to – eat foods in Kano metropolis

S/N	Food types	Number of samples examined	No of *E. coli*	No. of *Salmonella typhi*	No. of *Vibrio cholerae*
Fully processed foods					
1	Awara	5	-	-	-
2	Fried Groundnut	5	04(80)	-	-
3	Boiled Rice and Beans dish	5	05(100)	-	-
4	Fura	5	02(40)	-	-
5	Bread	5	02(40)	-	-
6	Zobo drink	5	01(20)	01(20)	-
	Sub –total	**30**	**14(23.3)**	**01(1.6)**	**0(0)**
Minimally processed foods					
1	Tomato	5	02(40)	02(40)	02(40)
2	Cabbage	5	02(40)	-	03(60)
3	Ginger drink	5	01(20)	-	-
5	Lettuce	5	05(100)	02(40)	05(100)
6	Carrot	5	02(40)	01(20)	05(100)
7	Kunun zaki drink	5	02(40)	-	-
	Sub - total	**30**	**14(23.3)**	**05(8.3)**	**15(25.0)**
	Grand total	**60**	**28(46.6)**	**06(10.0)**	**15(25.0)**

Key: - = negative

Values in parenthesis ( ) are percentages

minimally processed with 15 (25.0%). Lettuce and carrot recorded highest number of positive samples with 5 (100.0%) each, followed by cabbage with 3 (60.0%) and tomato 2 (40.0%). The other non – vegetable foods examined recorded negative for *V. cholerae* (Table 2). It could be observed from the result of this study that *V. cholerae* was only isolated from minimally processed foods, specifically vegetable samples. This could be attributed to the fact that feacally contaminated water has been incriminated in the transmission of the bacteria (Nester et al., 2004). And the bacteria survives only for few days on moist environments, such as surface of fruits and vegetable (Nester et al., 2004). The water used for the irrigation of vegetables in this study was observed to be contaminated with feacal matter.

Salmonella typhi recorded the least number of positive samples with 06(10.0%) out of the 60 samples examined (Table 2). *S. typhi* had 5 (8.3%) in minimally processed foods with 1 (1.6%) occurrence in the fully processed foods. Chomvarin *et al.* (2006) reported an incidence of 4.3% out of 145 ready – to – eat food samples examined. Lettuce and tomato had the highest number of 2(40.0%) each, with zobo drink and carrot each having 1(20.0%) positive sample. Awara, fried groundnut, cabbage, ginger drink, boiled rice and beans, fura, bread, kunun zaki all recorded negative for *S. typhi*. The occurrence of *S. typhi* in foods such as tomato, lettuce, carrot and zobo drink might be attributed to the water used in the preparation of such foods, as *S. typhi* has been reported to be transmitted via water and *Salmonella* carriers as food handlers (De wit, 1985), eventhough *S. typhi* has now been frequently isolated from other sources, such as sewers and feacally contaminated waters (Famurewa

and Moro, 1989; Uzeh. and Agbonlahor, 2001).

The occurrence of enteropathogenic bacteria such as *E. coli, V. cholerae* and *S. typhi* on the foods examined renders them as vehicles for food – borne infections. *E. coli* if consumed in foods is liable to causing peritonitis, mastitis, septicemia and gastrointestinal infections some-time becoming fatal (New World Encyclopedia, 2009). *V. cholerae,* on the other hand, is a well-known causative agent of cholera with symptoms of vomiting and diarrhea and could be fatal, while *S. typhi* is the causative agent of typhoid fever, an enteric fever that can also be fatal (Nester et al., 2004).

In Nigeria, an important consideration is the fact that most food handlers do not practice good personal hygiene and good manufacturing practices because of lack of continuous awareness and education by the relevant authorities concerned with food safety regulations and also the consumers' carefree attitude and gross poverty that makes people to sometimes eat whatever comes their way, whether hygienically safe or not.

Post – production contamination of ready – to – eat foods by environmental and human sources is a factor that tends to be overlooked here in Nigeria, but needs attention from researchers, authorities and consumers (Umoh, 1989; Oyeyi and Lum – Nwi, 2008).

Conclusion

The results of the present study revealed the occurrence of enteropathogenic bacteria such as *E. coli, V. cholerae* and *S. typhi* in fully and minimally processed ready – to – eat foods. This renders the quality of the foods examined

inadequate. Relevant authorities should educate food handlers on good personal hygiene and good manufacturing practices, which are sure ways of reducing the likelihood of the foods serving as vehicles for food-borne diseases/illnesses.

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