

Original Research Article

Bacteriological Quality of Vended Fresh Shrimps Harvested from Ndibe Beach, Afikpo North Local Government Area, Ebonyi State, Nigeria.Orji, J.O.¹, Nnachi, A.U.², Egwuatu, C.C.², Akujobi, C.N.², Iwuafor, A.A.³, Efunshile, A.M.⁴, Achi, E.C.⁵, Aghanya, I.N.²¹Department of Applied Microbiology, Faculty of Science, Ebonyi State University, Abakaliki, Nigeria²Department of Medical Microbiology and Parasitology, Faculty of Medicine Nnamdi Azikiwe University, Awka, Nigeria³Department of Medical Microbiology and Parasitology, Faculty of Medicine and Dentistry, College of Medical Sciences, University of Calabar, Calabar, Nigeria⁴Department of Medical Microbiology and Parasitology, Faculty of Medicine, Federal Teaching Hospital, Abakaliki, Nigeria⁵Accident and Emergency Unit, Federal Teaching Hospital, Abakaliki, Nigeria***Corresponding author**

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Abstract: This study determined the bacteriological quality of vended fresh shrimps harvested from Ndibe Beach, Afikpo North Local Government Area, Ebonyi State, Nigeria using standard microbiological and biochemical techniques. The total aerobic plate counts based on the body parts showed high microbial burden in the ranges of 2.0×10^7 to 2.84×10^7 cfu/g (head), 1.52×10^7 to 1.92×10^7 cfu/g (body) and 9.62×10^6 to 1.96×10^7 cfu/g (tail). The heads were the most contaminated with a mean bacterial load of $2.43 \pm 0.452 (x 10^7)$ cfu/g, while the tails were the least contaminated with $1.45 \pm 0.409 (x 10^7)$ cfu/g. The study also revealed the presence of four bacterial isolates including: *Klebsiella* species, *Escherichia coli*, *Staphylococcus aureus* and *Salmonella* species. Based on the body parts, *Klebsiella* spp. occurred highest in the head at the rate of 37%, followed by *Escherichia coli* (32%), *Staphylococcus aureus* (21%) and then *Salmonella* spp. (10%), while *Staphylococcus aureus* occurred highest (38%) in the body, followed by *Escherichia coli* (30%), *Klebsiella* spp. (27%) and then *Salmonella* spp. (5%). Also, *Staphylococcus aureus* occurred highest (38%) at the tail, followed by *Klebsiella* spp. (31%), *Escherichia coli* (31%) but *Salmonella* spp. was absent in the tail (0%). Overall, the results revealed that *Klebsiella* spp. had the highest percentage occurrence of 37(33%) while *Salmonella* had the least percentage occurrence of 8(7%). The study also showed that 62 (55%) of the isolated bacteria were from the head, 37 (33%) from the body and then 13 (12%) from the tail. The study has shown that fresh shrimps harvested from Ndibe Beach were highly contaminated with potential pathogenic bacteria and therefore unsafe for human consumption unless properly cooked. It is therefore recommended that both vendors and buyers should be well enlightened on the place of personal hygiene in the transmission of food borne diseases.

Keywords: Shrimps, bacteria, contamination, seafood, disease, vendors, Nigeria**INTRODUCTION**

Sea foods are important, healthy and rich sources of nutrients to man; this has earned them growing worldwide consumption and preference over red meat products. Shrimps, for instance, constitute a large group of crustaceans varying in sizes from microscopic to about 35 cm long [1] that have been considered the most important seafood traded worldwide [2]. They are widely distributed and their exports contribute largely to the total GDP of most countries such as China, India, Thailand, Bangladesh,

Taiwan and Vietnam [3-4]. More than 3.7 million tons were produced in 2010 with worth over 16 billion US dollars [5]. Shrimps are valuable sources of protein, xanthophylls carotenoid, selenium, heart-healthy vitamin B12 and a good, energy-promoting iron, phosphorus, niacin, anti-inflammatory omega-3-fatty acids, immune-supportive zinc, and bone-healthy copper and magnesium[6-9] Notwithstanding, the interplay of microorganisms that come in contact with them from harvesting through processing projects them as potential public health threats due to spoilage and

disease transmission. Baffone *et al.* [10] and Popovic *et al.* [11] noted that seafood products harvested from contaminated waters or which have been improperly preserved after harvesting are known to play an important role in infections. In Japan where raw sea food is very common, about 70% of food borne infections occurring in summer are mainly due to fish products [12]. In the fish processing chain, managing risks should be based on scientific knowledge of the microbiological hazards and the understanding of the primary production, processing and manufacturing technologies and handling during food preparation, storage and transport, retail and catering [13]. In Nigeria, there is lack of proper documentation of the contribution of shrimps to GDP; however, they are very expensive sea foods widely loved/consumed for their nutritional and savoury quality. However, the indiscriminate dumping of refuse into water bodies, unhygienic display of shrimps on open tables/floors, improper preservation techniques and lack of personal hygiene of vendors and buyers of shrimps call for public health concern over the bacteriological quality of vended shrimps and the potential for food-borne transmission. Therefore, this study determined the bacteriological quality of vended fresh shrimps harvested from Ndibe Beach, Afikpo North Local Government Area of Ebonyi State, Nigeria.

MATERIALS AND METHODS

Study Area

This study was carried out in Ndibe beach, Afikpo North in Ebonyi State, Nigeria. It is situated between latitude 5°53'N and longitude 7°56'E. Afikpo is the second largest city in Ebonyi State of Nigeria. It occupies an area of about 64 square miles (164km²) with an estimated population of about 156,611, according to the Nigerian 2006 Census. Afikpo is a hilly area despite occupying a region low in altitude, which rises 350 feet above sea level. It is a transitional area between open grassland and tropical forest and has an average annual rainfall of seventy-seven inches (198 cm). Ndibe beach (River) is located at Ndibe Autonomous community in Afikpo North Local Government Area of Ebonyi State.

Study Design/Microbiological Analysis

This was an observational study. Sixty (60) grams each of fresh shrimps were purchased from four different vendors within Ndibe beach market. The shrimps were immediately transported in a sterile plastic bag under ice to the Department of Applied Microbiology Laboratory, Ebonyi State University for identification and analysis. They were identified as *Palaemon serratus* by Mr. Uhuo Cosmos from the Department of Applied Biology of the same University. The shrimp samples from four different vendors were separated according to sizes and properly labelled. Each of the sizes was aseptically divided into three parts:

head, body, and tail using a sterile surgical blade. Then 10 g each of the parts were homogenized in 90 ml of 0.1% peptone water individually in sterile automatic blender and serially diluted. A 0.1 ml aliquot each of the dilutions of the different samples was pour-plated on Nutrient agar, *Salmonella-Shigella* agar, MacConkey agar and Mannitol Salt agar. The plates were incubated in triplicates at 37°C for 24 hours after which they were observed for growth and the colony counts taken and recorded as colony-forming unit per gram (cfu/g). The growths were further sub-cultured on NA, EMB, SSA and MSA and incubated at 37 °C for 24 hours after which the isolates were individually identified [14].

Identification of Isolates

All the suspected colonies were identified based on standard morphological/cultural characteristics (colour, shape, elevation, capacity, consistency and edge), gram reaction and biochemical tests (citrate, oxidase, indole, catalase, coagulase, methyl red, voges-proskauer tests) [15].

Statistical Analysis

ANOVA was used to determine the mean microbial load of the samples.

RESULTS

Investigation of the bacteriological quality of fresh shrimps harvested from Ndibe Beach based on the total aerobic plate counts on the body parts showed high microbial burden in the ranges of 2.0×10^7 to 2.84×10^7 cfu/g (head), 1.52×10^7 to 1.92×10^7 cfu/g (body) and 9.62×10^6 to 1.96×10^7 cfu/g (tail). The heads were the most contaminated with a mean bacterial load of $2.43 \pm 0.452 (x 10^7)$ cfu/g, while the tails were the least contaminated with $1.45 \pm 0.409 (x 10^7)$ cfu/g. Details of the results are shown in Table 1. Also, the study showed the presence of four bacterial genera including: *Klebsiella* species, *Escherichia coli*, *Staphylococcus aureus* and *Salmonella* species showing varied morphological and biochemical characteristics (Table 2).

Meanwhile, the results on the percentage occurrence of the bacterial isolates on the body parts showed that *Klebsiella* spp. occurred highest in the head at the rate of 37%, followed by *Escherichia coli* (32%), *Staphylococcus aureus* (21%) and then *Salmonella* spp. (10%), while *Staphylococcus aureus* occurred highest (38%) in the body, followed by *Escherichia coli* (30%), *Klebsiella* spp. (27%) and then *Salmonella* spp. (5%). Also, *Staphylococcus aureus* occurred highest (38%) at the tail, followed by *Klebsiella* spp. (31%), *Escherichia coli* (31%) but *Salmonella* spp. was absent in the tail (0%) (Figure 1).

Overall, the results revealed that *Klebsiella* spp. had the highest percentage occurrence of 37(33%)

while *Salmonella* had the least percentage occurrence of 8(7%) as shown in figure 2. The study also showed that 62 (55%) of the isolated bacteria were from the head,

37 (33%) from the body and then 13 (12%) from the tail (Figure 1).

Table-1: Total/Mean Aerobic Bacterial Counts (cfu/g) of Fresh Shrimps Harvested from Ndibe Beach.

Parts	Total Aerobic Bacterial Counts (cfu/g)				Mean (cfu/g) x 10 ⁷
	Vendor A	Vendor B	Vendor C	Vendor D	
Head	2.8 x 10 ⁷	2.84 x 10 ⁷	2.08 x 10 ⁷	2.0 x 10 ⁷	2.43±0.452
Body	1.92 x 10 ⁷	1.52 x 10 ⁷	1.68 x 10 ⁷	1.76 x 10 ⁷	1.72 ±0.167
Tail	1.96 x 10 ⁷	1.4 x 10 ⁷	1.48 x 10 ⁷	0.962 x 10 ⁷	1.45 ±0.409
Total	6.68 x 10 ⁷	5.76 x 10 ⁷	5.24 x 10 ⁷	4.722 x 10 ⁷	1.87±0.543

Table-2: Morphological and Biochemical Characteristics of Bacterial Isolates from Fresh Shrimps

S / N	Colonial Characteristics	Gram Reaction	Cell Morphology	Biochemical Test							Growth on MA	Growth on SSA	Suspected Bacterial Isolates
				Catalase	Coagulase	Oxidase	Indole	Mthyl Red	Voges-Proskauer	Citrate			
1	White smooth, flat slightly moist shiny surface and edge entire	-	Rods	+	-	-	+	+	-	+	+	-	<i>Escherichia coli</i>
2	Grey and mucoid	-	Rods	+	-	-	-	-	+	+	+	-	<i>Klebsiella spp.</i>
3	Light cream, irregular in shape, glossy in appearance with raised elevation and smooth edge	-	Rods	+	-	-	-	+	-	+	+	+	<i>Salmonella spp.</i>
4	Yellow, smooth, low convex edge, entire and shiny surface	+	Cocci in irregular clusters	+	+	-	-	-	-	-	-	-	<i>Staphylococcus aureus</i>

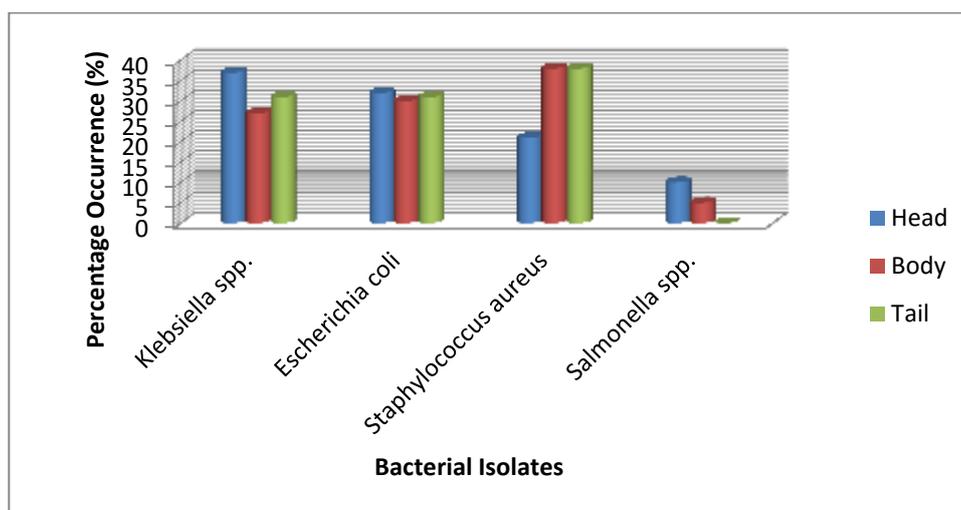


Fig-1: Percentage Distribution of Bacterial Isolates from Fresh Shrimps

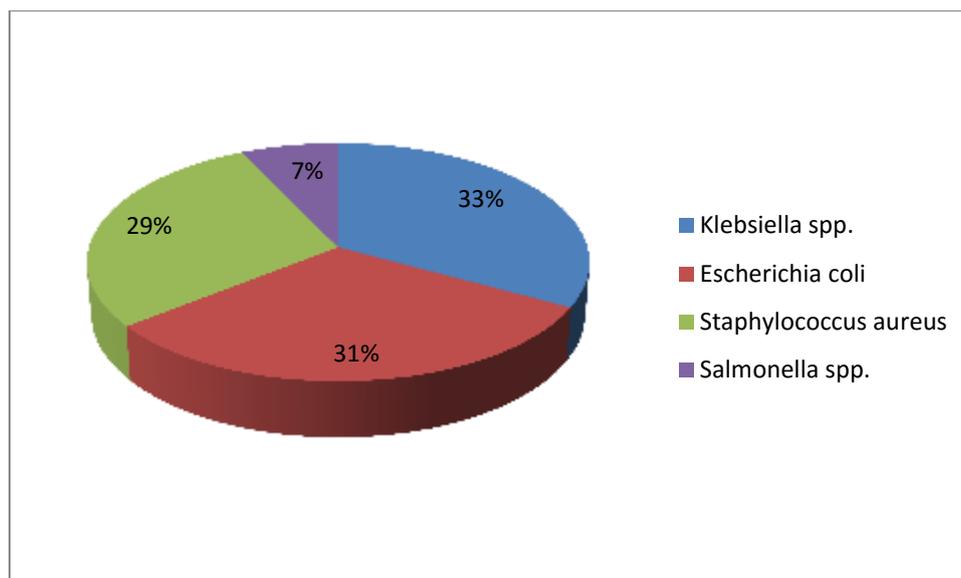


Fig-2: Overall Recovery Rate (%) of Each Bacterial Isolate

DISCUSSION

The role of sea foods in the transmission of food borne diseases is of global concern. This study on the bacteriological quality of fresh shrimps harvested from Ndibe Beach, Afikpo North Local Government Area, Ebonyi State, Nigeria revealed high microbial burden with overall mean aerobic bacterial counts of $1.87 \pm 0.543 (x 10^7)$ cfu/g. This high microbial burden corroborates the findings of Solomon *et al.* [9] in Rivers State, Nigeria. This is of great concern and could be explained by the unhygienic market environment where sea foods are generally sold in Nigeria as suggested by Ibe [16]. Most shrimps are displayed on dirty tables/containers and even on the bare floor in most places. Also, the vendors are mostly non literates and may not be enlightened about personal hygiene. Also, indiscriminate defaecation on land by both humans and animals could have contributed to this high microbial burden as these faeces are constantly swept into water bodies during rainfall. Due to the repeated touching of shrimps by buyers with bare hands, there could have been cross contamination by buyers who also bought contaminated products prior to the purchase of shrimps.

The study also revealed the presence of 112 bacterial isolates belonging to the following four (4) genera: *Klebsiella*, *Escherichia*, *Staphylococcus* and *Salmonella*. This is in line with the findings of Adesokan *et al.* [17] in Badagry Beach Lagos, who recovered similar bacterial isolates. However, the International Commission on Microbiological Specification of Foods (ICMSF) recommends zero presence of *Salmonella* spp. in raw crustacean. This is because faecal coliforms such as *E. coli* and *Salmonella* are indicators of faecal contamination, and their occurrence in seafood is associated with food poisoning. On the other hand, the presence of

staphylococci in food is associated with inadequate handling practices, causing food intoxication [18]. The high level of various isolates from shrimps may play major roles in causing sea food borne diseases and illness and this is why samples need to meet standards [19].

Staphylococcus aureus are mainly of human origin. The results on the prevalence of *Staphylococcus aureus* recorded in this study is in agreement with that reported by Whong *et al.* [20] and this may be due to the fact that both buyers and sellers constantly touch displayed shrimps with bare hands, thus introducing and increasing the population of *Staphylococcus aureus* in them. Ezeama [21] also noted that the frequency of occurrence of *Staphylococcus* in fresh shrimps may be associated with gross mishandling and contamination. Most illnesses from naturally contaminated seafood are associated with eating undercooked or raw shellfish [22]. *Staphylococcus aureus* enterotoxin A (SEA) is extremely potent with as little as 100 ng sufficient to cause intoxication [23].

The presence of *E. coli* in the sample investigated in this study can be attributed to the possibility of people defecating in the beach or even urinating while swimming which was also reported by Nester *et al.* [24] who stated that the presence of *E. coli* in all the samples they investigated may be suggestive of faecal contamination due to poor hygiene and sanitation. *E. coli* has been implicated in gastroenteritis leading to dysentery and bloody diarrhoea [25].

The presence of *Salmonella* spp. in this study is in disagreement with ICMSF [26] which stated that *Salmonella* and *Vibrio cholera* should not be found in sea food product. However, this result is in agreement

with the findings of Akinyema *et al.* [27] and Banerjee *et al.* [28] who previously reported *Salmonella* contamination in fresh shrimp. Zhang *et al.* [29] also documented the incidence of *Salmonella* in 730 samples aquaculture products from China. However aquaculture products including shrimps can become sources of *Salmonella* by exposure to contaminate water or through processing practices.

CONCLUSION

The study has shown that shrimps obtained from Ndibe Beach in Afikpo North Local Government Area of Ebonyi State, Nigeria were grossly contaminated by potential pathogenic bacteria at the time of this study. The contamination of the water body from faecal materials of animal and human origin, improper sanitation of the beach market environment, use of contaminated shrimps display materials as well as lack of or inadequate personal hygiene of both the vendors and the buyers are suggested as culprits to this high microbial burden observed in this study. Therefore, it is imperative that indiscriminate dumping of sewage/refuse into water bodies be strictly frowned at and discouraged, while use of clean display materials and good sanitation of the market environment encouraged. Vendors and buyers of shrimps should be enlightened on the health benefits of personal hygiene. Also, use of bare hands by intending buyers to touch displayed shrimps should be discouraged. Consumption of raw and/or undercooked shrimps should be stopped. The government should set up a monitoring and surveillance team that would constantly ensure the safety of shrimps sold in the state and the nation at large so as to curb the potential for food borne transmission of disease-causing microorganisms.

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