

Bacteriological quality and antibiotic sensitivity pattern of the isolates from *suya* spice sold in Port Harcourt, Nigeria

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Abstract: *Suya* is a spiced, barbecued, smoked or roasted meat product, prepared basically from meat of animals. The occurrence of microorganisms that are potentially pathogenic in spices used in *suya* preparation is regarded as main cause of gastrointestinal disturbances resulting from the consumption of *suya* in Nigeria. The essence of this study was to assess the bacteriological quality of *suya* spices sold at different *suya* spice depot in Port-Harcourt, Rivers State, Nigeria and to compare it with the International Commission on Microbiological Specifications for Foods (ICMSF). One hundred and fifty (150) samples of *suya* spice were sampled from three different *suya* spice depot located at Rumuokoro, Woji, and Trans – Amadi all in Port-Harcourt, Rivers State. Standard and established methods was used for bacteriological analyses. The Total plate counts (log cfu/g) for the samples varied from 3.45 to 6.16. *Staphylococcus* sp was present at high levels in all samples ranging from 3.69 to 6.02 that may indicate an inappropriate hygienic quality of samples. Isolates were equally identified using Analytical Profile Index. Unsatisfactory qualities were found to be 28.67% of the samples due to the presence of *E. coli*. In all samples examined, only 12% of *Salmonella* was detected from the sample collected from Woji. The Antibiotic sensitivity pattern was carried out to ascertain the best antibiotic suitable for a particular microorganism. Result from this study that some organisms have multiple resistance to antibiotics; this is of public health importance. Isolates were sensitive to some of the antibiotics tested, with *Kocuria kistina* and *Paenibacillus polymyxin* been the most sensitive to gram- negative disc, while *Citrobacter* is most resistant for gram negative disc. Whereas for Antibiotic gram positive disc *salmonella* is most sensitive and is most resistant is *Paenibacillus polymyxin*. Considering the results obtained, the samples analyzed contain a high level of total viable count of 7.90 (log cfu/g) as against 5.70 to 6.70 of ICMSF. Only eight samples (5.33%) had acceptable levels for all microbial factors according to EU Commission Recommendation of (plate count: 5.70 to 6.70 (log CFU/g) *staphylococcus*: 2 (log CFU/g) *E.coli*: Not detected, *salmonella*: Not detected. The isolation of these potential pathogens from these spice samples analyzed is of public health significance. The need to provide control mechanisms and establish best practice to improve the quality and safety of spices, means more studies are needed.

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1. Introduction

Suya is a ready to eat spicy, barbecued, smoked or roasted meat. Its origin can be traced to the Hausa people of northern Nigeria, Sub-Saharan Africa, where their main occupation is rearing of cattle and growing of cash crops. Thus, it is an important preoccupation and a major source of livelihood for the people. This led to the production of different types of beef products such as *Kundi*, *Kilishi*, *Balangu* and *Suya*, which are very popular protein-rich foods. However *Suya* is now the most popular as it is consumed in other parts of the country (Inyang *et al.*, 2005; Ogbonna *et al.*, 2012).

Today, *Suya* vendors have become prominent with their grill stands becoming very busy from about midday until late at night in town and cities. It is gradually making its way into elite circles where it has become a delicacy served at parties and other social

events. *Suya* is prepared basically from boneless meat of animals (Abdullahi *et al.*, 2004; Ogbonna *et al.*, 2012). The thinly sliced meat is marinated in various spices which include peanut cake, salt, ginger and other flavorings, and then barbecued (Egbebi and Seidu, 2011). *Suya* is usually served with further helpings of dried pepper mixed with spices, and sliced onions. There is no standard recipe for the production of the complex mixture of spices and additives which make up the *Suya* marinade (called *Yaji*) and the spice mix served with it (Ugwuja *et al.*, 2009). The ingredients used for *Suya* spice vary according to personal and regional preferences for taste, which comprises of garlic, clove, ginger, chili pepper, negro pepper, salt, peanut cake as well as food additives such as Monosodium glutamate and Maggi Cube in some cases (Ugwuja *et al.*, 2009). Addition of spice is one of the most important stages during the production of

suya because it is a critical control point, it is equally added to the *suya* after processing (Shamsuddeen and Ameh, 2008).

The safety of food is a major concern not only to the consumers but also the producers as well. Despite the high degree of awareness of food preservation methods there is increasing occurrence of disease outbreaks caused by pathogenic and spoilage microorganisms in foods (Meng and Doyle, 1998). Spices are natural products that can be obtained from parts of certain plants such as the roots, rhizomes, bulbs, bark, leaves, stems, flowers, fruits and seeds. They are valued for their distinctive flavors, colors and aromas, and are among the most versatile and widely used ingredients in food preparation, processing and preservation worldwide (McKee, 1995).

Spices are cultivated and collected in tropical areas using traditional methods, which means they are exposed to contaminants from the soil and air, before being well dried to prevent possible microbial growth, as well as during harvesting, handling and packing (Kneifel and Berger, 1994).

Over the years the use of spices have increased tremendously, they have been applied in several foods especially protein foods. Spices by nature are prepared in combination of herbs (condiments) and the method of preparation lack good hygiene practice. Study of the spice by some researchers' shows that they harbour microorganisms and some are spore formers which can give rise to food poisoning outbreak with public health significance. This research study investigates the health implication as the spice is added to the meat before and after it has been processed.

Thus, the aim of this study is to assess the bacteriological quality of *suya* spice, using cultural and Analytical Profile Index (API) method and to compare it with the International Commission on Microbiological Specifications for Foods (ICMSF).

2. Materials And Methods

2.1. Source of Samples: Samples of *suya* spice mixture were purchased from Three locations, all situated in Rivers State. The mixture is composed of ginger, garlic, West African black pepper, hot pepper and groundnut. A total of one hundred and fifty (150) samples were collected with each location having 50 samples. Samples were taken to the laboratory immediately and analysed within two hours.

2.2 Sample Preparation and Serial Dilution: The sample preparation was carried out according to the method described by Ogbonna et al. (2012) and Okonko et al. (2013). In this method, 25g of sample (spice) was weighed and homogenized by blending in 225ml peptone water using blender at 15,000-20,000 rpm. This was labelled as 1:10 dilution which is also

the stock or the homogenate. This was further serially diluted to 1:10⁻⁵.

2.3. Total Aerobic Plate Count: This was carried out according to the method of Abdullahi *et al.* (2004). Total bacterial count were enumerated on nutrient agar plates by spread plate method using 0.1 ml of 10-fold dilution (10⁻² to 10⁻⁵) of bacterial suspensions. All inoculated plates were incubated at 35±2 °C for 48 hours at 35°C. The bacterial colonies on plates were counted and randomly picked and purified by sub-culturing unto fresh agar plates using the streak plate technique. Isolated colonies that appear on plates are then transferred into nutrient agar slants, properly labelled and stored as stock cultures. The bacterial isolates were identified based on their morphology, Gram reaction and biochemical characterization.

2.4. Isolation and enumeration of *E. coli*: Eosine Methylene Blue Agar (EMBA) (Himedia Laboratories Pot Ltd, India) was used for the isolation and enumeration of *E. coli*. EMBA plates prepared following autoclaving were inoculated with and incubated at 37°C for 24 hours after which typical colonies with greenish metallic sheen were subjected to biochemical tests for *E. coli*. This was followed by further biochemical characterization for confirmation as described by Cowan and Steel (1965) and Cheesbrough (2004). Colonies were counted and expressed as CFU/g.

2.5. Isolation and enumeration of *Salmonella* and *Shigella sp.*: Desoxycholate Citrate Agar (DCA) (Park Scientific Limited, Moulton Park, Northampton) was used for the isolation and enumeration of *Salmonella* and *Shigella spp.* 1 ml quantity of homogenized spice saline mixture was each inoculated into 9ml of pre-enrichment broths (tetrathionate and selenite cysteine) and incubated at 37°C as described by Cowan and Steel (1965) and Cheesbrough (2004). DCA plates were inoculated with 0.1ml of the pre-enrichment broth 24h growth and incubated at 37°C overnight. Typical colonies with black centres were identified as *Salmonella spp* on DCA as described by Cowan and Steel (1965) and Cheesbrough (2004). Pinkish colonies were identified presumptively as *Shigella* on DCA and subjected to further biochemical testing as described by Cowan and Steel (1965) and Cheesbrough (2004).

2.6. Enumeration and Detection of *Staphylococcus aureus*: This was carried out according to Abdullahi *et al.* (2004). A quantity (0.25 ml) of inoculum from 10⁻³, 10⁻⁴, and 10⁻⁵ dilutions were transferred into duplicate Petri dishes which were considered accordingly. This was followed by pouring aseptically about 20- 25 ml of molten Mannitol Salt Agar (MSA). The plates were incubated at 37 °C for 24 hrs. Plates containing 30-300 black colonies were selected and the colonies counted. The average was taken and the number obtained was

multiplied by four and then by the inverse of the dilution factor. This gave the number of colony forming units per gram of a sample (CFU/g). Plates of Mannitol Salt Agar were inoculated and incubated at 35°C for 24 hrs. Following incubation, mannitol fermenting organisms which showed a yellow zone surrounding their growth were isolated onto agar slants for biochemical tests.

2.7. Identification of isolates from *Suya*: Pure cultures of the isolates were subjected to different biochemical test: Oxidase, Catalase, Indole Production, Methyl Red Vogues Proskauer (MRVP), and Sugar fermentation. Identification of isolates were done using API 50 CH/CHB/ 20E kits.

2.8. Statistical Analysis: Data obtain from proximate analysis and mean of microbial load were analyzed using Single factor ANOVA.

3. Results

The results of bacteriological analysis from the one hundred and fifty samples of *suya* spice collected from three study areas, Rumuokoro, Trans- Amadi and

Woji ranged from 5.46 to 6.43, 5.72 to 5.90 and 5.84 to 6.47 (log CFU/g) respectively. Table 1-6 shows mean bacterial load/count (TVC) of *Suya* spice collected from three different site in Rivers State, Nigeria. Trans-Amadi has the lowest of 5.72 to 5.90 CFU/g and Woji recorded the highest of 5.84 to 6.47CFU/g of bacterial load. The study showed that there was no significant difference in the mean microbial load of the three study site, as the P-value were all greater than 0.05 (Table 1-6).

Table 1: Mean Bacterial Load/Count (TVC) Of *Suya* Spice Collected From Three Different Site In Rivers State, Nigeria

Sites	Mean Bacterial Count (log CFU/g)		
	Trans-Amadi	Rumuokoro	Woji
Site One	5.90	5.78	6.47
Site Two	5.72	6.61	5.84
Site Three	5.67	6.43	6.08

Table 3: Analysis on microbial load from samples collected from Trans-Amadi

Microorganism	Number of Samples	Number of samples contaminate	Acceptable Levels (log cfu/g)	Number of satisfactory samples	Number of unsatisfactory samples
Plate count	50	50	5.70 to 6.70	72% (36)	28% (14)
<i>Staphylococcus</i>	50	43	2	14%(7)	86%(43)
<i>E. coli</i>	50	25	Not detected	50%(25)	50%(25)
<i>Salmonella</i>	50	0	Not detected	100%(50)	0

Table 4: Analysis on microbial load from samples collected from Rumuokoro

Microorganism	Number of Samples	Number of samples contaminated	Acceptable Levels (log cfu/g)	Number of satisfactory samples	Number of unsatisfactory samples
Plate count	50	50	5.70 to 6.70	80% (40)	20% (10)
<i>Staphylococcus</i>	50	21	2	58% (29)	42% (24)
<i>E. coli</i>	50	3	Not detected	94% (47)	6% (3)
<i>Salmonella</i>	50	0	Not detected	100% (50)	0

Table 5: Analysis on microbial load from samples collected from Woji

Microorganism	Number of Samples	Number of samples contaminated	Acceptable Levels (log cfu/g)	Number of satisfactory samples	Number of unsatisfactory samples
Plate count	50	50	5.70 to 6.70	84% (42)	16% (8)
<i>Staphylococcus</i>	50	21	2	58% (29)	42% (21)
<i>E. coli</i>	50	15	Not detected	70% (35)	30% (15)
<i>Salmonella</i>	50	6	Not detected	88% (44)	12% (6)

Table 6: Analysis on microbial load from samples collected from Analysis results the three sites cumulative

Microorganism	Number of samples	Number of samples contaminated	Acceptable Levels (log cfu/g)	Number of satisfactory samples	Number of unsatisfactory samples
Plate count	150	150	5.70 to 6.70	79% (118)	21.33% (32)
<i>Staphylococcus</i>	150	85	2	47.33% (71)	52.67% (79)
<i>E. coli</i>	150	43	Not detected	71.33%(107)	28.67% (43)
<i>Salmonella</i>	150	6	Not detected	96% (144)	4% (6)

Figure 1-4 shows the percentage of unsatisfactory samples as a result of contamination by locations. The plate count (TVC), Staphylococcus counts and *E. coli* counts shows that Trans-Amadi had the highest unsatisfactory samples, followed by Rumuokoro and Woji had the least total viable counts (Figure 1-3), except for Salmonella count which was present in samples from Woji (Figure 4). However, samples from Rumuokoro had the lowest *E. coli* counts (Figure 3) while samples from Rumuokoro and Woji locations had equal proportion of Staphylococcus counts (Figure 2).

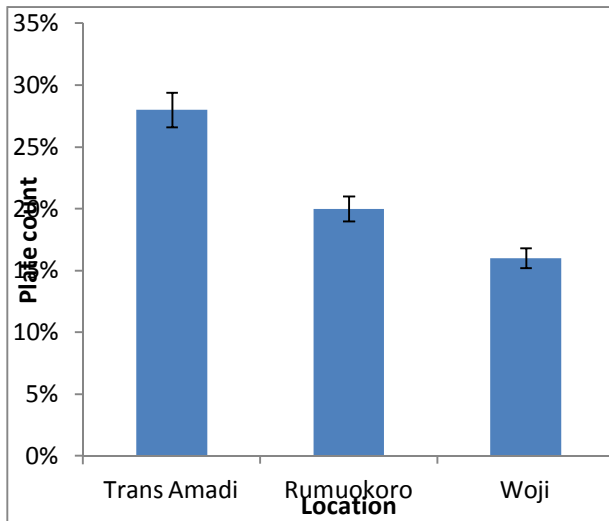


Figure 1: Percentage of Unsatisfactory samples as a result of contamination by plate count (TVC). Error bars represent the degree of accuracy from the mean.

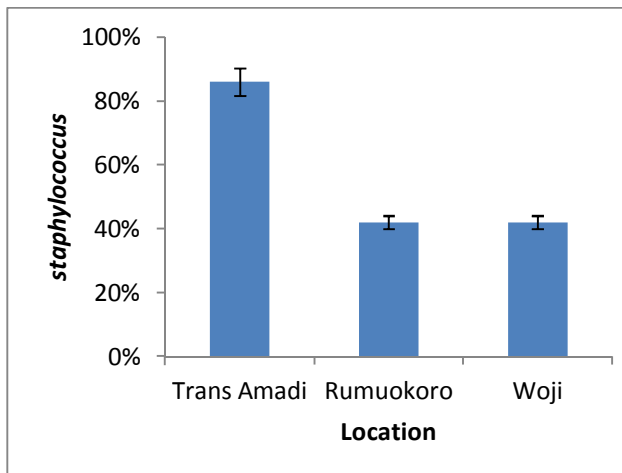


Figure 2: Percentage of Unsatisfactory samples as a result of contamination by *Staphylococcus*. Error bars represent the degree of accuracy from the mean.

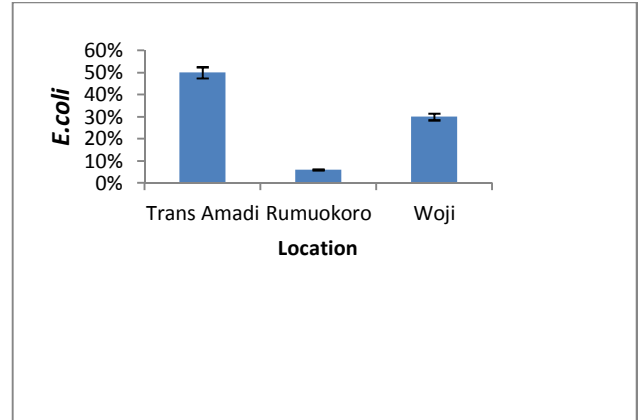


Figure 3: Percentage of Unsatisfactory samples as a result of contamination by *E. coli*. Error bars represent the degree of accuracy from the mean.

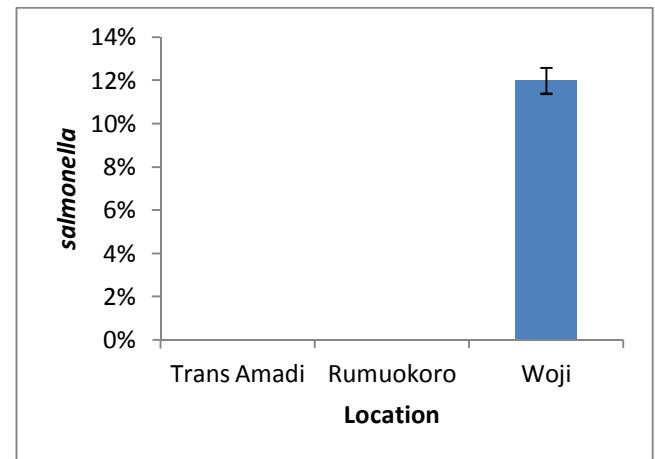


Figure 4: Percentage of Unsatisfactory samples as a result of contamination by *salmonella*. Error bars represent the degree of accuracy from the mean.

Table 7: Identification of Bacteria Using Analytical Profile Index (API)

S/N	Organisms Identified By Analytical Profile Index (API)	% Identification
1.	<i>Kocuria kristinae</i>	99.9
2.	<i>Bacillus circulans</i>	54.1
3.	<i>Paenibacillus</i>	98.9
4.	<i>Enterobacter cloacae</i>	95.0
5.	<i>Enterobacter amnigenus</i>	96.6
6.	<i>Bacillus subtilis</i>	66.5
7.	<i>Corynebacterium accolens</i>	98.1
8.	<i>Bacillus cereus</i>	99.8
9.	<i>Bacillus megaterium 2</i>	99.9
10.	<i>Paenibacillus macerans</i>	94.0
11.	<i>Staphylococcus</i>	98.7
12.	<i>Salmonella</i>	96.8

Table 7 shows identification of bacteria using Analytical_Profile Index (API). The organisms identified were *Kocuria kristinae*, *Bacillus circulans*, *Paenibacillus*, *Enterobacter cloacae*, *Enterobacter amnigenus*, *Bacillus subtilis*, *Corynebacterium accolens*, *Bacillus cereus*, *Bacillus megaterium 2*, *Paenibacillus macerans*, *Staphylococcus* and *Salmonella* (Table 7).

Identification of isolates made by different API KITS: API STAPH V.40, API 50 CHB V 3.0, API CORYNE V2.0, API 20 E V4.0.

Figure 5-7 shows the percentages of occurrence of the identified isolates. It showed that *Staphylococcus* sp.

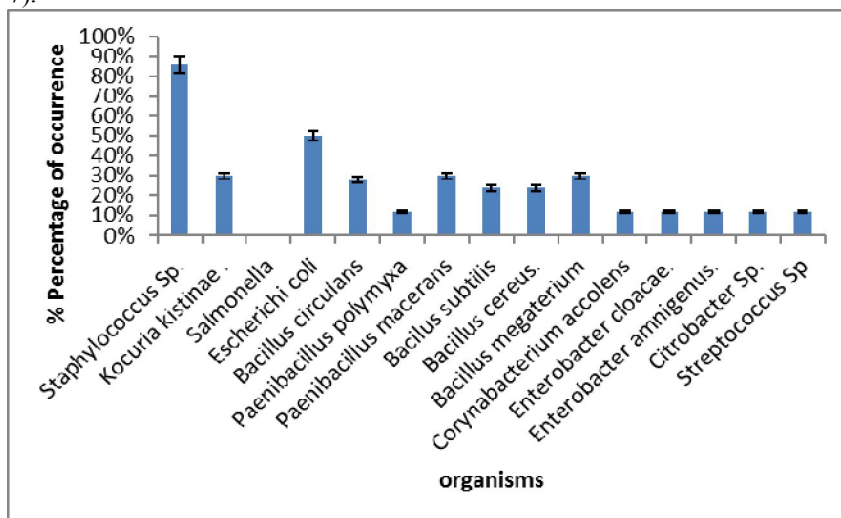


Figure 5: Percentage Occurrence of Bacteria In The Sample From Trans-Amadi. Error bars represent the degree of accuracy from the mean

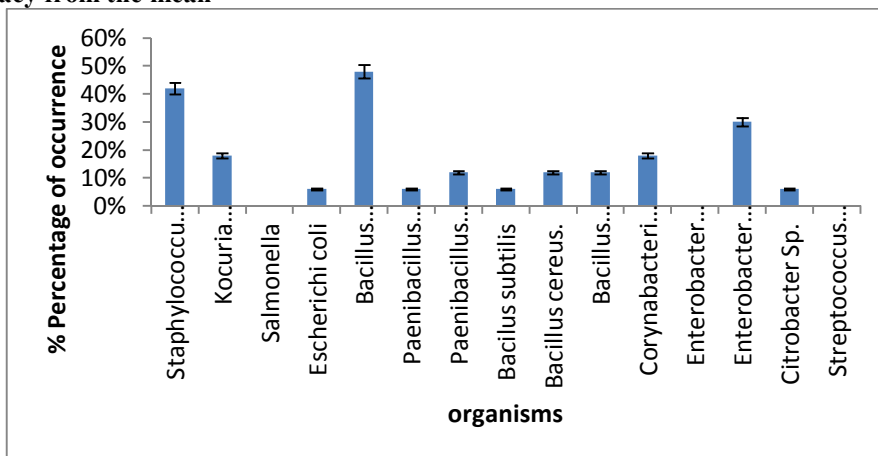


Figure 6: Percentage Occurrence of Bacteria in the Sample from Rumuokoro Error bars represent the degree of accuracy from the mean.

Table 7-8 shows antibiotic sensitivity pattern of identified isolates. It shows that all the organisms exhibited inhibition zones during testing for both gram -ve . Some organisms like *Kocuria kristinae*, *Paenibacillus polymyxa* organism are more sensitive, as they exhibited high sensitivity to all gram -ve disc while *Citrobacter* sp and *E.coli* showed most resistance to Septrin, Chlorophenical, Amoxalin, Augmentin, Gentamycin, Tarivid and Streptomycin of the antibiotics, but however, they are still sensitive to

Sparfloxacin and Pefloxacin. *Salmonella* showed to be most sensitive using gram +ve disc than any other organism, as they sensitive to Ciproflo, Norfloxacin, Gentamycin, Amoxil, Streptomycin, Rifampicin, Erythromycin, Chloramphenicol, Ampiclox and Levofloxacin. *Enterobacter cloacae* and *Enterobacter amnigenus* showed the most resistance, though they are sensitive to antibiotic like Rifampicin and Norfloxacin (Table 7-8).

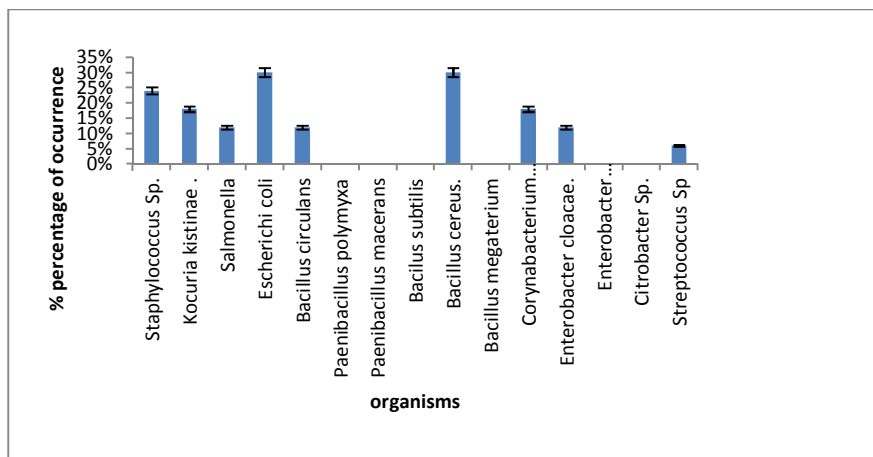


Figure 7: Percentage of Occurrence of Bacteria in the *suya* spice sample from Woji. Error bars represent the degree of accuracy from the mean.

Table 7: Antibiotic sensitivity pattern of the identified isolates (GRAM -VE DISC)

ISOLATES	SP 30µg	AM 30µg	AU 30µg	CN 10µg	PEF 30µg	OFX 10µg	S 30µg	SXT 30µg	CH 30µg	CPX 10µg
1. <i>Staphylococcus Sp.</i>	-	-	-	+	+	+	+	+	+	+
2. <i>Kocuria kistinae</i>	+	+	+	+	+	+	+	+	+	+
3. <i>Salmonella</i>	+	-	-	+	+	+	+	-	+	+
4. <i>Escheriachi coli</i>	+	-	-	-	+	-	-	-	-	-
5. <i>Bacillus circulans</i>	+	-	-	-	+	-	-	-	-	+
6. <i>Paenibacillus polymyxa</i>	+	+	+	+	+	+	+	+	+	+
7. <i>Paenibacillus macerans</i>	+	+	+	+	+	+	+	+	+	-
8. <i>Bacilus subtilis</i>	+	-	-	-	+	-	-	-	-	+
9. <i>Bacillus cereus</i>	+	-	-	-	+	-	-	-	-	+
10. <i>Bacillus megaterium</i>	+	-	-	-	+	+	-	-	-	+
11. <i>Corynebacterium accolens</i>	-	-	-	-	+	-	+	-	-	-
12. <i>Enterobacter cloacae</i>	+	-	-	-	+	-	-	-	-	+
13. <i>Enterobacter amnigenus</i>	+	-	-	-	+	-	-	-	-	+
14. <i>Citrobacter Sp.</i>	-	-	-	-	+	-	-	-	-	-
15. <i>Streptococcus Sp.</i>	+	+	+	+	+	-	+	+	+	+

Resistant (-), Sensitive (+), SEP- Septrin, CH- Chlorphenicol, SP Sparfloxacin, CPX- Ciprofloxacin, AM- Amoxicillin, AU-Augmentin, CN-Gentamycin, PEF-Pefloxacin, OFX –Tarivid and S- Streptomycin.

Table 8: Antibiotic sensitivity pattern of the identified isolates (GRAM +VE DISC)

ISOLATES	CH 30µg	CPX 10µg	E 30µg	LEV 20µg	RD 20µg	NB 10 µg	CN 10µg	APX 20 µg	AML 20 µg	S 30µg
<i>Staphylococcus Sp.</i>	+	+	+	-	+	+	-	-	+	+
<i>Kocuria kistinae</i>	+	+	+	+	+	-	-	-	-	+
<i>Salmonella</i>	+	+	+	+	+	+	+	+	+	+
<i>Escheriachi coli</i>	-	-	-	+	-	-	-	-	-	-
<i>Bacillus circulans</i>	-	-	+	+	+	-	+	+	-	-
<i>Paenibacillus polymyxa</i>	-	-	+	-	-	-	-	-	-	-
<i>Paenibacillus macerans</i>	-	-	+	+	-	-	-	+	-	-
<i>Bacilus subtilis</i>	-	-	+	+	+	-	+	+	-	-
<i>Bacillus cereus</i>	-	-	+	+	+	-	+	+	-	-
<i>Bacillus megaterium</i>	-	-	+	+	+	-	+	+	-	-
<i>Corynebacterium accolens</i>	+	-	+	+	+	-	+	+	-	-
<i>Enterobacter cloacae</i>	-	-	-	-	+	+	-	-	-	+
<i>Enterobacter amnigenus</i>	-	-	-	-	+	+	-	-	-	+
<i>Citrobacter Sp.</i>	-	-	+	+	+	-	-	-	+	-
<i>Streptococcus Sp</i>	+	+	+	+	+	+	+	+	+	-

Resistant (-), Sensitive (+).CPX- Ciproflox, NB- Norfloxacin, CN- Getamycin, AML-Amoxyl, S- Streptomcin, RD- Rifampicin, E- Erythromycin, CH- Chloramphenicol, APX – Ampclox, LEV- Levofloxacin

4. Discussion

Suya is extensively used in Nigeria cooking, babecue etc, either in the prepared meal or ready to eat meal. *Suya* spice is very good seasoning due flavor and aroma it add to the meal, from the study it is good especially in minerals. It was equally observed from this study that some organisms have multiple resistance to antibiotics, this is of public health importance. The wholesomeness and safety of the consumer must therefore be given high priority. The essence of this study was to assess the bacteriological quality of *suya* spices sold at different *suya* spice depot in Port-Harcourt, Rivers State, Nigeria and to compare it with the International Commission on Microbiological Specifications for Foods (ICMSF).

The results of bacteriological analysis from the one hundred and fifty samples of *suya* spice collected from three study areas, Rumuokoro, Trans- Amadi and Woji ranged from 5.46 to 6.43, 5.72 to 5.90 and 5.84 to 6.47 (log CFU/g) respectively. Trans-Amadi has the lowest of 5.72 to 5.90 and Woji recorded the highest of 5.84 to 6.47 of Bacterial load, this is in line with the work of (Debs-Louka *et al.*, 2013) whose bacteria load ranges from 5.57 to 8.85 (log CFU/g). The high bacterial count found in the samples reflect poor handling, inappropriate drying conditions, poor storage and lack of good manufacturing practice (Gillespie *et al.*, 2000; Okonko *et al.*, 2013). Statistical analysis showed that there was no significant difference in the mean microbial load of the three study site, as the P-value were all greater than 0.05.

The International Commission on Microbiological Specifications for Foods (ICMSF) specifications require that bacterial load should not be more than ($5 \times 10^5 - 10^6$), while *Salmonella* like all pathogens, should be absent in 25 g of spice. As for the other contaminants, standards fluctuate among the different references. For instance, the European Spice Association [ESA (2004)] and the International Commission on Microbiological Specifications for Foods (ICMSF 2011) set up the absolute maximum of *E.coli* must be less than 3 log cfug⁻¹, whereas the Canadian guidelines established by the ministry of Agriculture of Quebec requires this count to be less than 2 log cfug⁻¹ and *Staphylococcus aureus* 2 log cfug⁻¹ and other bacteria requirements should be agreed between the buyer and the seller (Muggeridge *et al.*, 2001).

All *Suya* spice samples were contaminated with Bacteria. The average count was 5.91 log CFU g⁻¹. Plate counts greater than 6 log CFU g⁻¹ were approximately 21.33% of the samples whilst 78.67% were between 3 log CFU g⁻¹ and 6 log CFU g⁻¹, and are acceptable. Generally, in spices, plate count equal to or more than 6 log cfug⁻¹ is not acceptable based on

the international commission on microbiological specification for food (ICMSF, 2005).

The high plate count found in the *suya* spice samples may reflect poor handling, poor storage, exposure to open air, inappropriate drying conditions or just a general lack of hygiene during processing (Okonko *et al.*, 2013), this is in line with the work of Salari *et al.* (2012) whose bacteria count ranges from 3 to 10.6 (CFU/g). Based on recommendation 2004/24/EC and European Spice Association (ESA) specifications, 118 out of 150 samples were satisfactory, i.e. were of an acceptable microbiological quality, but 32 (21.33%) were unsatisfactory due to high levels of plate count contamination.

High incidences and numbers of plate count could be considered part of the normal flora or exposure to the environment pre- and post-harvesting. It has been reported that microbial counts vary according to the region, year of production, harvest month, storage conditions prior to drying and poor hygiene during processing. Thus, the observed plate counts are likely to reflect the original bio-load of the raw material during harvesting and transportation (Farkas *et al.*, 2000) and poor handling during processing. These findings conform to existing literature (King *et al.*, 1981; Banerjee and Sarkar, 2003), which reported high microbial counts on dried spices of export quality due to preparation methods and handling. *Staphylococcus aureus* and *Bacillus cereus* have been known to be implicated in food borne illness. *Staphylococcus aureus* have been reported to be normal flora in human and animals, their presence in foods are indicative of excessive human handling (Odu and Okonko, 2012; Okonko *et al.*, 2013).

The presence of aerobic spore formers like *Bacillus cereus* and *Bacillus subtilis* is due to poor handling, lack of storage facility and lack of good manufacturing practice. This was equally reported by (Debs-Louka *et al.*, 2013) on the assessment of microbiological quality of spice and herbs sold in Lebanon. The presence of spore-forming bacteria like *Bacillus cereus* is of public health importance as these spices are added to the ready –to- eat *suya* after heating, when the heat generated is not enough to kill the spores, yet it may provide them the energy required to initiate spore germination.

Escherichia coli are one of the dominant microorganisms present in animal and human faeces. The assumption of this bacterium as an indicator organism is based on the concept that its detection in *suya* spice samples indirectly provides evidence that the sample has been contaminated with fecal material and that pathogenic organisms may potentially be present (Feng, 2001).

E. coli, was observed to be present at all the study sites with Trans – Amadi town recording the highest of 50%, Woji (30%) while Rumuokoro recorded the lowest of 6% of unsatisfactory quality. The presence of indicators of faecal contamination, such as *E. coli*, has equally been reported by other studies have reported *E. coli* in a wide range of spices (Garcia *et al.*, 2001; Banerjee and Sarkar, 2003; Sagoo *et al.*, 2009). This findings has public health implications with respect to safety, especially since at several occasions the spices are not subject to any further heat treatment.

The presence of *Staphylococci aureus* may result from soil or other sources associated with unsanitary conditions in the production, mainly handlers (Gillespie *et al.*, 2000; Richardson and Stevens, 2003; Marriott, and Gravani 2006). Frequency of occurrence of bacterial isolate in relation to the three study areas shows *Staphylococcus aureus* was found generally to be the highest, with Trans-Amadi recording 86%, Rumuokoro 42% and Woji 24%. In all, 76 samples out of the total of 150 samples *suya* spice has *S. aureus*. This is due to poor personal hygiene as well as inappropriate manufacturing practice. *S. aureus* are found in human nose, throats, skin and 50-60% of normal people are carriers' thus high occurrence.

Salmonella was not detected in any of the samples from Trans-amadi and Rumuokoro, only six samples (12%) of the fifty samples from Woji location recorded the presence of *Salmonella*, which is 4% of the three sites accumulative. It is most likely that the presence of *Salmonella* spp. count (4%) at only one site (Woji) is due to handling from the vendor, while 96% of the satisfactory samples can be attributed to the nature of spice products. This is contrary to the work of Salari *et al.* (2012), which recorded the absent of *Salmonella* on the study carried out on assessment of the microbiological quality and mycotoxin contamination of Iranian red pepper spice. Our results are inconsistent with others researchers, which suggest *Salmonella* spp. is uncommon in spices (Julseth and Deibel, 1974; Garcia *et al.*, 2001; Abou Donia, 2008). However, it is consistent with Banerjee and Sarkar (2003) who found two positive samples in their study.

This study evaluated the antimicrobial activity organism isolated from of *suya* spice, result obtained shows that all the organisms exhibited inhibition zones during testing for both gram -ve. Some organisms like *Kocuria kistinae*, *Paenibacillus polymyxa* organism are more sensitive, as they exhibited high sensitivity to all gram –ve disc while *Citrobacter* sp and *E.coli* showed most resistance to Septrin, Chlorophenical, Amoxalin, Augmentin, Gentamycin, Tarivid and Streptomycin of the antibiotics, but however, they are still sensitive to some antibiotic like Sparfloxacin and Pefloxacin. *Salmonella* showed to be most sensitive

using gram +ve disc than any other organism, as they sensitive to Ciproflox, Norfloxacin, Gentamycin, Amoxil, Streptomycin, Rifampicin, Erythromycin, Chloramphenicol, Ampiclox and Levofloxacin. While *Enterobacter cloacae* and *Enterobacter amnigemus* showed the most resistance, though they are sensitive to antibiotic like Rifampicin and Norfloxacin. This showed that all the organisms involved could be treated by antibiotics

5. Conclusion

Suya spice samples analyzed in this study contained a high number and wide range of microorganisms with only two (5.33%) fit-for-purpose/safe for human consumption. These results suggest sanitary conditions at different stages in production must be improved to reduce the potential hazard to human health. However, it is difficult to select a single microbial index for the determination of quality because spices are used as ingredients in a variety of products prepared in different ways. The need to provide control mechanisms and establish best practice and to improve the quality and safety of spices, means more studies are needed, particularly to determine effective methods of decontamination but also safe processing methods pre- and post-harvest, and better transportation and storage.

References

1. Abdullahi, I. O., Umoh, V. J., Ameh, J. B. and Galadima, M. (2004). Hazards Associated with *Kilishi* Preparation in Zaria, Nigeria. *Nigerian Journal Microbiology*. 18 (1-2): 339 – 345.
2. Abou Donia, M. A. 2008. Microbiological Quality and Aflatoxinogenesis of Egyptian Spices and Medicinal Plants. *Global Vet.*, 2(4): 175-181.
3. Banerjee, M. and Sarkar, P.K. (2003). Microbiological quality of some retail spices in India. *Food Research International* 36: 469-474.
4. Cheesbrough M. 2004. District laboratory practice in tropical countries. Part 2. p. 62-70 Cambridge University Press, Great Britain.
5. Cowan ST and Steel KJ. 1965. Manual for the identification of medical bacteria. p. 149 – 165. Cambridge University Press, Great Britain.
6. Debs-Louka E, Joelle E. and Fouad D. (2013). Assessment of the Microbiological Quality and Safety of Common Spices and Herbs Sold in Lebanon. *Journal of Food Nutrition*. 2:4.
7. Egbebi, A. O and Seidu, K. T. (2011). Microbiological Evaluation of *Suya* (dried smoked meat) sold in Ado and Akure, South West Nigeria. *European Journal of Experimental biology* 4 (1): 1-5.

8. European Commission (EC). 2004. Commission Recommendation of 19 December 2003 Concerning a Coordinated Programme for the Official Control of Food Stuffs for 2004 (2004/24/EC). *Off. Journal European Union*, 6: 29–37.
9. European Spice Association (2004) European Spice Association Quality Minima Document. Bonn, Germany.
10. Farag, S.E.A and Abo-Zeid, M.(1997). Degradation of the natural mutagenic compound satrole in spices by cooking and irradiation. 41:359-361.
11. Farkas, J., (2000). *Spices and Herbs, in the Microbiological Safety and Quality of Foods*. (Eds.): Lund, B. M., Baird-Parker, T.C. and Gould, G. W. Aspen Publication, 1:35-40.
12. Feng P (2001) *Escherichia coli: Guide to Foodborne Pathogens* John Wiley and Sons, Inc., New York. 2:20-23.
13. Garcia, E.C., Cabrera, C., Lorenzo, M.L., and Lopez, M.C. (2000), Chromium levels in spices and aromatic herbs, *The Science Total Environment*. 247, page 51-56.
14. Gillespie, I., Little, C. and Mitchell, R. (2000). Microbiological Examination of Cold Ready-to-eat Sliced Meats from Catering Establishments in the United Kingdom. *Journal of Applied Microbiology*, 88: 467-474.
15. International Commission on Microbiological Specification for foods (ICMSF, 1978). *Microorganisms in foods. 2. Sampling for microbiological analysis. Principles and specific applications*, University of Toronto press. Pp:2.
16. International Commission on Microbiological Specifications for Foods (ICMSF, 2011) *Microorganisms in foods 8. Use of Data for Assessing Process Control and Product Acceptance*. (1st edn), Springer, India. Pp 12-36.
17. Inyang C. U., Igyor, M. A and Uma, E. N. (2005). Bacterial Quality of a Smoked Meat product ('Suya'). *Nigeria Food Journal*. 23(3): 239-242.
18. Julseth, R. M. and Deibel, R. H. 1974. Microbial Profile of Selected Spices and Herbs at Import. *J. Food Tech.*, 37(8): 414-419.
19. King, A. D., Hocking, A. D. and Pitt, J. I. (1981). The Mycoflora of Some Australian Foods. *Food Tec. Aust.*, 33: 55-60.
20. Kneifel, W. and Berger, E. (1994). Microbial criteria of random samples of spices and herbs retailed on the Australian market. *Journal Food Protein* 57: 893-901.
21. Marriott, N.G., Gravani, R.B. (2006) Principles of food sanitation. Springer, India. 5: 221-225
22. McKee, L. H. (1995). Microbial Contamination of Spices and Herbs: A Review. *L.W. T.*, 28: 1–11.
23. Meng, J. and Doyle, M.P. (1998). Emerging and evolving microbial foodborne pathogens. *Bulletin De L Intitut Pasteur*, 96 (3): 151-163.
24. Muggeridge, M. and Clay, M. (2001). Quality Specifications for Herbs and Spices. In: "Handbook of Herbs and Spices". Woodhead Publishing Ltd, Cambridge, page 13–22.
25. Odu, N.N. and Okonko, I.O. (2012). Bacteriology quality of traditionally processed peanut butter sold in Port Harcourt metropolis, Rivers State, Nigeria. *Researcher* 4(6):15-21.
26. Ogbonna, I. O., Danladi, M. S., Akimusire, O. and Odu, C. M. (2012). Microbiological Safety and Proximate Composition of suya stored at Ambient Temperature for six hours from Maiduguri, Northern Nigeria. *International Journal of Food Safety*. 14 (2): 11-16.
27. Okonko IO, Odu NN and Igboh IE. 2013. Microbiological Analysis of Kilishi Sold In Port Harcourt, Nigeria. *New York Science Journal*, 6(7):37-43.
28. Richardson, I. R. and Stevens, A. M. 2003. Microbiological Examination of Ready-to eat Stuffing from Retail Premises in the North-east of England. The 'Get Stuffed' Survey. *J. Appl. Microbiol.*, 94: 733-737.
29. Sagoo, S.K., Little, C.L., Greenwood, M., Mithani, V. and Grant, K.A. (2009) Assessment of the microbiological safety of dried spices and herbs from production and retail premises in the United Kingdom. *Food Microbiology* 26: 39-43.
30. Salari B, Habibi-Najafi, Boroushaki MT, Mortazavi SA, Fathi Najafi M. 2012. Assessment of the microbiological quality and mycotoxin contamination of Iranian red pepper spice. *J. Agr. Sci. Tech. (2012) Vol. 14: 1511-1521*.
31. Shamsuddeen, U. and Ameh, J. B. (2008): Survey on the possible critical control points in kilishi (a traditional dried and grilled meat snack) produced in kano. *International Journal of Bioscience*. 3(2): 34-38.
32. Ugwuja, E.I., Ugwu, N.C. and Nwibo, A.N. (2009). Dietary supplement containing mixture of raw curry, garlic and ginger. *Internet Journal on Nutrition Wellness*, 5(2), ISSN:1937-8297.