

# Microbiological quality and antibiotic sensitivity of potential pathogens isolated from meat product (Suya) sold in Rivers State University and its environs

Lucretia I. Barber • Patience C. Obinna-Echem\* • Sophia C. Amadi

Department of Food Science and Technology, Rivers State University, P.M.B 5080, Port Harcourt, Rivers State, Nigeria.

\*Corresponding author. E-mail: [patience.obinna-echem@ust.edu.ng](mailto:patience.obinna-echem@ust.edu.ng)

Accepted 22<sup>nd</sup> October, 2018

**Abstract.** The microbiological and chemical qualities of Suya, raw beef and chicken used in the suya production were assessed. *Coliform*, *Staphylococcus aureus*, *Lactobacilli* and *Escherichia coli* were isolated from the products using conventional microbiological method. The antibiotic sensitivity of isolated pathogens (*E. coli* and *S. aureus*) was evaluated. The suya had total volatile nitrogen content of 2.76 to 8.40 mg N/100 g and salt content 0.29 to 5.84%. *Lactobacilli* (6.46 Log<sub>10</sub> CFU/g) was detected in the raw chicken after 3 to 6 h and 24 h of purchases. In the raw beef, total coliform increased significantly ( $P \leq 0.05$ ) (6.08 to 6.32 Log<sub>10</sub> CFU/g) after 24 h while *Staphylococcus* and *Lactobacilli* decreased from 7.16 to 6.69 Log<sub>10</sub> CFU/g and 7.18 – 6.54 Log<sub>10</sub> CFU/g respectively. In the suya, total coliform varied from 8.54 – 6.05 Log<sub>10</sub> CFU/g and was below detection limit in some samples. *S. aureus*, *Lactobacillus* and *E. coli* ranged from 5.00 to 8.48, 4.00 to 8.48 and 4.00 to 8.52 Log<sub>10</sub> CFU/g, respectively. *E. coli* was sensitive to Ciproflaxain, Pefloxacin, Tarivid, Augmentin, Amoxicillin, Septrin, Gentamycin and Sparfloxacin but resistant to Chloramphenicol and Streptomycin. The inhibition zones varied from 2.67 ± 0.00 mm for Gentamycin to 10.00 ± 0.95 mm for Ciproflaxain. *S. aureus* was sensitive to Pefloxacin, Ciprofloxacin, Streptomycin, Septrin, Zinnacef, Gentamycin, Rocephin and Erythromycin but resistant to Ampiclox and Amoxicillin. The inhibition zones ranged between 1.85 ± 0.21 mm for Erythromycin and 11.70 ± 0.00 mm for Pefloxacin. Though the pathogens were sensitive to some antibiotics, proper processing, hygienic practices and adequate supply of portable water and good storage facilities will minimize contamination and ensure safe suya for consumption.

**Keywords:** Suya, meat product, microbiological quality, antibiotic sensitivity, pathogens.

## INTRODUCTION

Humans have hunted and killed animals for meat since prehistoric times. Meat can be defined as the flesh of an animal used as food and consumption of meat is as old as man. Most commonly consumed meat are specifically identified by the live animal from which they are obtain such as; beef–(meat from cattle), veal (meat from calf), pork (meat from pigs), mutton (meat from sheep) etc (Vilar *et al.*, 2000). Meat plays an important role in human diet by contributing both macro and micro nutrients that are required for growth and good health maintenance. Meat can be prepared by different methods such as cooking, boiling, frying, or roasting. It can also be

processed into different forms that are easier and convenient for consumers, these include canned beef, hot dogs, sausages, cured meat, 'suya', bacon etc. Traditionally processed meat products are consumed in different parts of the world (Vilar *et al.*, 2000).

Suya is popularly consumed spicy, barbecued, smoked or roasted meat product that originated from the Northern Nigeria, where rearing of cattle is an important preoccupation and a major source of livelihood for the people (Edema *et al.*, 2008). Suya can be prepared form ram, beef, or chicken and also from offals such as liver, kidney and gizzard. There are three main types of suya,

*Tsire, Kilishi and Balangu* (Omojola *et al.*, 2004). Its production process involves thinly slicing of raw boneless meat, staking onto skewers, marinating with various spices such as ginger, garlic, salt, pepper, black pepper etc. The spiced skewered beef is then coated with a mixture of groundnut paste, sprinkled with vegetable oil and barbecued over coal fire. Before serving or packaging it, its reheated for a given time, re-spiced with the addition of a little more spices and garnished with thinly sliced onion rings, cabbage and tomato ball (Abdullahi *et al.*, 2004; Jonathan *et al.*, 2016). Suya has become a popular street delicacy in several countries, particularly those in West Africa. It is consumed as a snack, and frequently served in hotels, bars, restaurants and in homes (Iyang *et al.*, 2005). Like any other meat product, it serves as an excellent supply of high quality protein, vitamins and mineral.

Suya processing is still at the artisanal stage and most processors are uneducated and operate under unhygienic condition with the increased risk of food borne infection. Suya spices are Nigeria Indigenous spices commonly used on roasted meat (barbecued meat) to give its unique desired taste. These spices help to add flavor and aroma to the roasted meat. It consists of red pepper (*Capsicum spp*), white pepper (*Piper guinensis*), ginger (*Zingiber officinate*), African nutmeg (*monodora myristica*), *xylopia aethiopica*, curry, salt, maggi seasoning, and monosodium glutamate. Studies have shown that spices used in suya preparation may contain high population of bacteria and fungi which may be viable even at the time of marketing (Olayiwola *et al.*, 2013). According to FOA (1998), if spices are not kept properly, it may be contaminated by microorganisms and may pose health hazard to humans. There are reports of sporadic cases of gastroenteritis and symptoms of food borne infection after consumption of suya which make this product (suya) a food safety risk (Odule and Akinyaiju, 2003; Iyang *et al.*, 2005). If these infections are not properly treated, it can result in the development of antibiotic resistance organisms (FAO/WHO, 2003; 1998). Several studies on meat product including suya sold in different parts of the country (Bori metropolis, Maiduguri, Ozoro Delta State, Ekpoma Edo, Enugu State) have revealed potential pathogens such as *Bacillus cereus*, *B. subtilis*, *Enterobacter aerogenes*, *Escherichia coli*, *Micrococcus luteus*, *Staphylococcus aureus*, *S. epidermidis*, *Proteus species*, *Serratia species*, *Salmonellae*, *Shigella*, yeast and aflatoxigenic molds (*Aspergillus flavus* and *Aspergillus parasiticus* (Edema *et al.*, 2008; Lawrence *et al.*, 2016; Eke *et al.*, 2013; Orogu and Oshilim, 2016; Ogbonna *et al.*, 2012, Chukwura and Mojekwu, 2002). In the study conducted by Lawrence *et al.* (2016) many of the isolated pathogens were resistant to most antibiotic used in their study and this posed a high health risk to humans.

This study aimed at assessment of the microbiological quality and antibiotic sensitivity of isolated potential

pathogens in suya sold in Rivers State University and its environs.

## MATERIALS AND METHODS

### Sample collection

Raw beef and raw chicken, and beef and chicken suya used for this study were purchased from the Staff Club in Rivers State University, Nkpulu-Oroworuku, Port Harcourt, Nigeria. Beef suya samples were also bought from five (5) different locations outside the University. These locations were: Eagle Island by the University backgate I, Azikiwe by the University backgate II, University main gate by Chinda Estate, University main gate by Npkolu Junction and Iloabuchi. The samples were bought wrapped in their usual packaging materials (foil for samples from the university and used newspapers for samples from outside the university). The samples were each placed in a well labelled secondary carry bag and transported in a cooler containing ice block to the Microbiology laboratory in the Department of Food Science and Technology, Rivers State University Port Harcourt. Microbiological analysis was carried out after 3 to 6 h of purchase and after 24 h of storage in the refrigerator.

### Microbial media and reagents

MacConkey, Mann-Rogosa and Sharpe (MRS), Eosin methylene blue (EMB) and Nutrient agars and Peptone water were obtained from TM Media, Titan Biotech Ltd, Rajasthan, India. Other reagents used were of analytical grade in the Department of Food Science and Technology, Rivers State University, Port Harcourt, Rivers State, Nigeria.

### Suya preparation

To be able to ascertain the critical control points (CCP) in the suya processing, the processing method at the Staff club of Rivers State University, Port Harcourt (locations F) was monitored. The raw meat products were delivered to the processor. It was washed under running tap water. Processing table was washed with water. The meat was thinly sliced and skewered in thin sticks. The skewered meat was marinated with spices, sprinkled with vegetable oil and barbecued over coal fire.

### Microbial analysis

#### Enumeration of microorganisms

Samples (25 g) of the raw beef and raw chicken, and the

**Table 1.** The loaded antibiotics, their abbreviations and concentration in the Gram –ve and the Gram +ve discs.

Gram negative disc			Gram positive disc		
Antibiotics	Abbreviation	Concentration (µg)	Antibiotics	Abbreviation	Concentration (µg)
Septrin	SXT	30	Pefloxacin	PEF	10
Chloramphenicol	CH	30	Gentamycin	GN	10
Sparfloxacin	SP	10	Ampiclox	APX	30
Ciproflaxain	CPX	10	Zinnacef	Z	20
Amoxacillin	AM	30	Amoxacillin	AM	30
Augmentin	AU	30	Rocephin	R	25
Gentamycin	CN	10	Ciprofloxacin	CPX	10
Pefloxacin	PEF	30	Streptomycin	S	30
Tarivid	OFX	10	Septrin	SXT	30
Streptomycin	S	30	Erythromycin	E	10

suya were homogenized with 225 ml of sterile peptone water in a laboratory blender, stomacher 400 circulator (Seward Ltd., West Sussex, UK) for 1 min and serially diluted to  $10^{-5}$  in the same diluent following the procedure described by Harrigan, (1998). One hundred micro litres aliquots of the dilutions were spread-plated on appropriate microbial media for each organism. Total *coliform*, *Lactobacilli* and *Escherichia coli* were enumerated on MacConkey, Mann-Rogosa and Sharpe (MRS) and Eosin methylene blue (EMB) agars respectively, incubated at 37°C for 24 to 48 h for total coliform and lactobacillus. The EMB plates were incubated at 44°C for 24 to 48 h. *Staphylococcus* was enumerated on Manitol salt agar incubated at 37°C for 24 to 48 h.

### Isolation and purification of bacteria

Representative colonies from various enumeration plates were subcultured by streak plating onto their respective fresh agar plates and were incubated at 37°C for 24 to 48 h for total coliform and *Lactobacillus*. The EMB plates were incubated at 44°C for 24 to 48 h. *Staphylococcus* was subcultured on Manitol salt agar and was incubated at 37°C for 24 to 48 h. At the end of the incubation period, plates were examined to ensure that they contained pure cultures.

### Characterization of colony

Isolated organisms from the different plates were characterized by their colony morphology (colony shape, appearance, colour, edge, elevation) and Gram Staining. Gram staining was carried out according to the method of Harrigan, (1998).

### Biochemical tests

Catalase and coagulase tests were carried out as described by Cheesebrough (2000). Briefly, a colony of

the subcultured bacterial was inoculate into few drops of hydrogen peroxide solution on a slide and observed for immediate active bubbling for positive catalase test. For coagulase test, a colony of the subcultured organism was emulsified with a drop of distill water on the end of a slide to make a thick suspension. With a wire loop, plasma was added and mixed gently. The slide was examined for clumping or clotting of the bacteria within 10 s.

### Antibiotic sensitivity test

The antibiotic sensitivity of the isolated pathogens was conducted using a gram positive disc (Maxi Disc High profile +ve, Maxicare Medical Laboratory, Lagos Nigeria) for *S. aureus* and a gram negative disc (Maxi Disc High profile –ve, Maxicare Medical Laboratory, Lagos Nigeria) for *E. coli*. The antibiotics loaded are shown in Table 1. The inoculum was prepared according to the standard method by Harrigan (1998). The bacteria were cultivated on Nutrient agar incubated at 37°C for 24 h. A distinct colony from the agar plate culture was inoculated into 10 ml of Peptone water and incubated at 37°C without agitation for 18 to 20 h such that 1 ml of inoculum produced 8 Log<sub>10</sub> CFU/ml. One thousand micro litres of the inoculum was spread plated onto nutrient agar plates. The discs were then placed on the plates and were incubated for 24 h.

### Chemical test

#### Total volatile nitrogen

Total volatile nitrogen was carried out on the samples using standard AOAC (2005) methods. Five grams (5 g) of each sample were weighed and blended into digestion flask (heating flask) containing 100 ml of distill water. 0.6 g of magnesium oxide (MgO) was added to the flakes. 25 ml of boric acid was introduced into a conical flask which acts as a receiving flask with 3 drops of methyl red

indicator. The two flasks were mounted in the distillation unit for 5 min. The content in the receiving flask was titrated to the end by a weak acid (0.1 N HCl). The total volatile nitrogen was determined as:

$$\text{TVN (mg N/100 g)} = [(\text{Sample titre} - \text{blank titer}) \times \text{Molarity of acid} \times 14] / \text{Sample weight}$$

### **Salt content**

Salt content was carried out on the samples using standard AOAC (2005) methods. Two grams (2 g) of each of the samples were weighed and ashed at 550°C for 2 h. The ashed samples were dissolved in 100 ml of distilled water. Two (2) drops of potassium dichromate was added to 10ml of the solution in a conical flask. The solution was titrated against 0.1 N silver nitrate (AgNO<sub>3</sub>). The salt content was expressed as:

$$\% \text{ Salt} = (T/V) \times sf \times d \times 100$$

Where:

T = Titre value

sf = Salt factor 0.005844

V = Volume of sample used

d = Dilution factor

### **Statistical analysis**

Data collected were subjected to analysis of variance (ANOVA): general linear model (GLM) using IBM SPSS (Release 2015) software for windows version 23.0 (Armonk, Ny: IBM Corp). Statistical differences were established at ( $P \leq 0.05$ ).

## **RESULTS AND DISCUSSION**

### **The critical control points (CCP) in the suya processing**

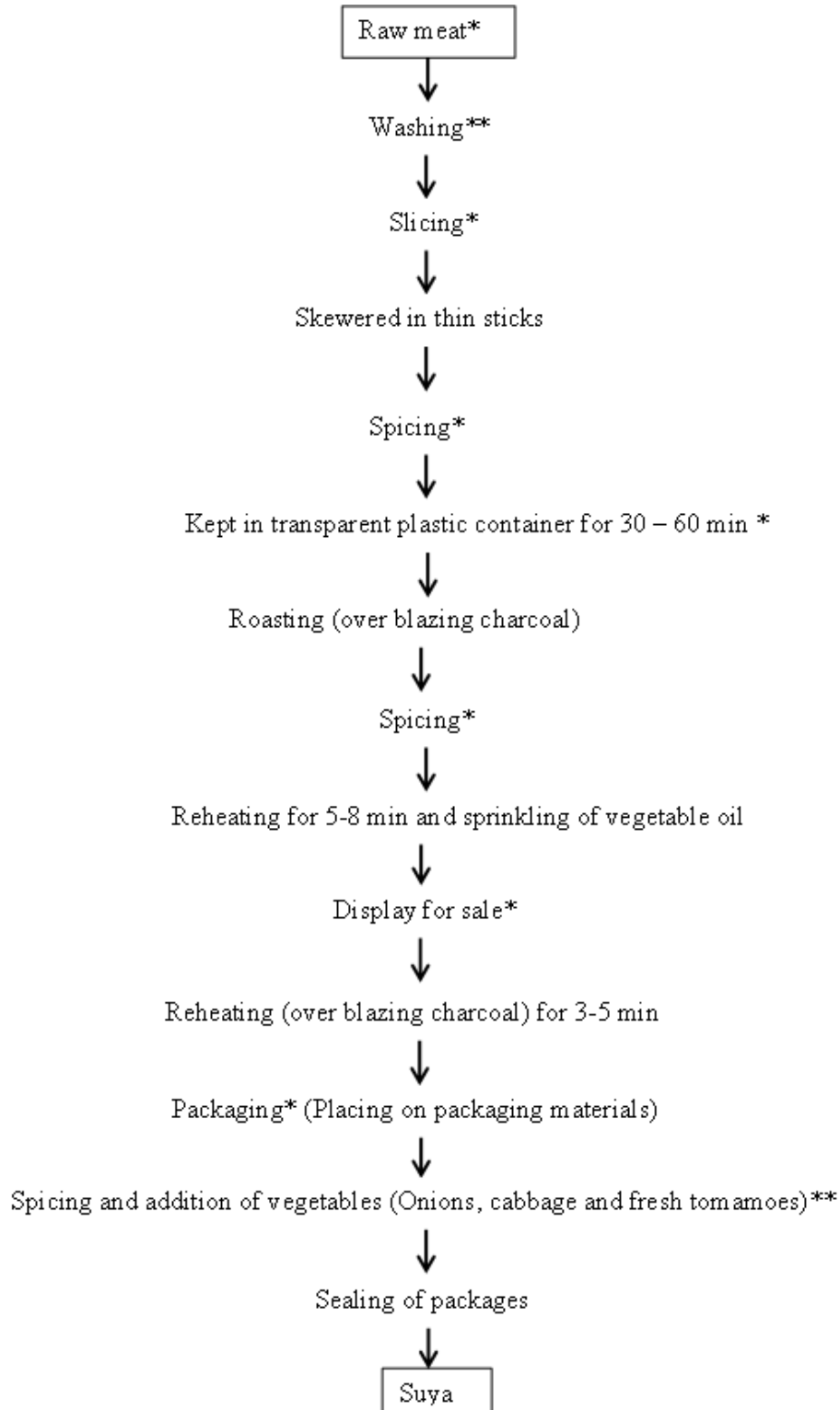
The monitoring of the suya production method at the Staff club of Rivers State University, Port Harcourt revealed the steps shown in Figure 1. The identified critical control points (CCPs) were indicated with asterisks. This is in line with the method described by Edema *et al.* (2008) but some additional steps were observed: the washing, storage before roasting, the sprinkling of vegetable oil during reheating and the addition of vegetables during the last spicing stage. The main points of contamination of the product were observed to be from the raw meat, washing, slicing, spicing, storage, garnishing with vegetables, and packaging. The raw beef and chicken after purchases were held in the refrigerator for a short while before use, while others were stored away in the freezers. Although, inconsistent power supply posed some challenge. The wooden table used can harbour

microbial contaminants in the scratches from the cutting and slicing process and can serve as a source of contamination to the product. To check this, there is steady water supply and the table was washed twice every day, before the beginning of processing and at the end of processing. Washing alone may not get rid of any bacteria in the chinks created during cutting and slicing of the beef. It was observed that the chopping board that is sometimes used for raw chicken was used during the packaging of the processed suya, although, it is the packaging material (aluminum foil) that comes in contact with the board that has been washed after the raw material processing. It was suggested that a separate chopping board be provided for use during the packaging of the processed product. The mixed suya spices were stored in an air tight container with lids. It is opened when needed and lids replaced after use and kept away in the refrigerator. This is an important practice to avoid contamination. More on the recommended control measures in suya production was reported by Edema *et al.* (2008)

### **The total volatile nitrogen and the salt content**

The total volatile nitrogen and the salt content of the suya samples varied significantly ( $P \leq 0.05$ ) as shown in Table 2.

Total volatile nitrogen ranged from 2.76 for sample B to 8.40 mg N/100 g in sample E while the salt content varied from 0.29 in sample B to 5.84% in sample C. Total volatile nitrogen content is an important indicator of meat's freshness. TVN compounds in chicken include mainly ammonia, trimethylamine (TMA) and dimethylamine (DMA) and the levels of TVN compounds increase with spoilage by either microbiological or enzymatic degradation (Urmila *et al.*, 2015). In meat, total volatile nitrogen consists mainly of ammonia with trace amount of trimethylamine. Ammonia is commonly considered as a chemical indicator of microbial meat spoilage, due to the deamination of amino acid by psychrophiles under aerobic condition. The safe permissible limit of TVN recommended by EOS (2005) in red meat should not exceed 20 mg/100 g (Hemmat *et al.*, 2013). The low total volatile nitrogen content of the suya product was an indicative of good quality of the meat and could account for their shelf life under good refrigeration storage before use for the raw product or sales for the suya. The total salt content found in the sample can be attributed to the spices and salt used during the product preparation. In addition to flavor and taste of food, salt is important in lowering the water activity of the meat product. Water activity is the amount of water available for microbial growth and other chemical activities. Hence salt plays important role in inhibiting microbial growth. Health Canada recommends that the maximum recommended amount of sodium per day for adults is 2,300 mg (CMC, 2004). The salt content of the suya



**Figure 1.** The diagram for suya preparation indicating critical control points (CCPs) as observed at the Staff club of Rivers State University, Port Harcourt (\* indicate CCPs).

samples (0.29 to 5.84% which is equivalent to 2.9 to 58.4 mg/g) is below the maximum recommended content and would not pose any health risk.

#### Microbial analysis

The morphological and biochemical (catalase and coagulase reaction) characteristics of the isolated

**Table 2.** Total volatile nitrogen and salt content of the suya.

Location	Sample ID	TVN (mg N/100g)	Salt (%)
Backgate I (Eagle Island)	A	4.98 ± 0.08 <sup>d</sup>	1.75 ± 0.17 <sup>bc</sup>
Backgate II (Azikiwe)	B	2.76 ± 0.21 <sup>e</sup>	0.29 ± 0.07 <sup>c</sup>
Maingate I (Chinda Estate)	C	6.15 ± 0.40 <sup>c</sup>	2.34 ± 0.21 <sup>b</sup>
Maingate II (Npkolu)	D	6.00 ± 0.28 <sup>c</sup>	5.84 ± 0.08 <sup>a</sup>
Iloabuchi	E	8.40 ± 0.30 <sup>ab</sup>	2.33 ± 0.07 <sup>b</sup>
Staff club beef	*Fb	6.99 ± 0.15 <sup>b</sup>	1.90 ± 0.18 <sup>bc</sup>
Staff club chicken	*Fc	5.01 ± 0.50 <sup>d</sup>	2.00 ± 0.15 <sup>b</sup>

Values with same superscript in the same column are not significantly different ( $P \leq 0.05$ ).

\*Fb and Fc represents beef and chicken suya from the Staff club in Rivers state University, Port Harcourt.

**Table 3.** Morphological and biochemical characteristics of microorganisms Isolated from the suya.

Isolates	Color on media	Gram reaction	Shape	Arrangement	Coagulase	Catalase	Probable identity
<i>Coliform</i>	Pink on MCA	-	Rod	Clustered	-	-	Pink and yellow
<i>Staphylococcus aureus</i>	Yellow on MSA	-/+	Round	Clustered	+	+	Golden yellow
<i>Lactobacillus</i> spp.	Creamy on MRS	+	Rod	Irregular	-	-	Creamy
<i>Escherichia coli</i>	Purple on EMB plate	-	Snort rod	Separated	-	-	Dark purple

MCA - MacConkey Agar

MSA - Manitol salt Agar

MRS – de Mann-Rogosa and Sharpe Agar

EMB - Eosin methylene blue agars

(-) and (+) are negative and positive reactions respectively

organisms isolated from the raw and processed beef and chicken and their preliminary identification are presented in Table 3. The isolated and identified bacteria were *Coliform*, *Staphylococcus aureus*, *Lactobacillus* spp. and *Escherichia coli*. *Staphylococcus aureus* and *Escherichia coli* are potential pathogens, *Staphylococcus*, *Samonella* spp. and *Escherichia coli* have been isolated in similar meat product (Egbebe and Seedu, 2011; Hassan *et al.*, 2014). *Coliform* is indicative of both microbiological quality and safety issues. The isolation of these organisms from the meat product is an indication of contamination at the various critical control points (CCPs) as shown in Figure 1. The growth of *S. aureus* indicated contamination of raw materials, poor hygiene practices by the suya processors, dirty environment, and contaminated processing utensils like knives and tables, in accordance with the report by Field (2002) and Edema *et al.* (2008). Presence of coliform indicated contaminated water and poor hygiene of suya handlers. *E. coli* may be as a result of faecal contamination or the use of contaminated water during washing of raw meat which was also reported by Odey *et al.* (2013). *Escherichia coli* is a Gram negative facultative anaerobic rod belonging to the family Enterobacteriaceae. It is a natural component of human gut flora its presence in food will imply contamination of faecal origin and a risk that other enteric pathogens (Jay, 2000). The

consumption of these products can lead to food poisoning which will pose a public health challenge.

Table 4 showed the microbial load of raw beef and raw chicken from the staff club in Rivers State University (location F), after 3 to 6 h of purchase and after 24 h of storage in the refrigerator.

Microorganisms in the raw chicken were below detection limit, except for *Lactobacillus* count of 6.46 Log<sub>10</sub> CFU/g after 3 to 6 h of purchases. This was also reduced below detection limit after 24 h of storage in the refrigerator. The microbial load that was below detection limit in the raw chicken could be attributed to the cold temperature storage as they were purchased in frozen condition from the suppliers and there was no resuscitation (that is, growth in enrichment medium) before the isolation and enumeration. The storage for 24 h in the refrigerator was to mimic what was obtainable in the suya locations. For the raw beef, there were significant ( $P \leq 0.05$ ) differences in the microbial counts after 3 to 6 h and after 24 h except for *E. coli*. There was significant ( $P \leq 0.05$ ) increase in the Total coliform count from 6.08 to 6.32 Log<sub>10</sub> CFU/g after 24 h. *S. aureus* and *Lactobacilli* decreased significantly ( $P \leq 0.05$ ) from 7.16 to 6.69 Log<sub>10</sub> CFU/g and 7.18 to 6.54 Log<sub>10</sub> CFU/g respectively. The microbial load of the raw materials can influence the microbial quality of the final product.

Shown in Table 5 are the microbial growth on beef and

**Table 4.** Microbial count (Log<sub>10</sub> CFU/g) of the raw beef and chicken before processing.

Sample	Time (h)	Total coliform	<i>Staphylococcus</i>	<i>Lactobacillus</i>	<i>Escherichia coli</i>
Raw beef	3-6	6.08 ± 0.05 <sup>b</sup>	7.16 ± 0.05 <sup>a</sup>	7.18 ± 0.04 <sup>a</sup>	6.38 ± 0.05 <sup>a</sup>
	24	6.32 ± 0.05 <sup>a</sup>	6.69 ± 0.03 <sup>b</sup>	6.54 ± 0.05 <sup>b</sup>	6.38 ± 0.05 <sup>a</sup>
Raw chicken	3-6	-	-	-	-
	24	-	-	-	-

Values with the same superscript in the same column do not differ significantly ( $P \leq 0.05$ ). N = 3±SD  
 Samples were from the staff club in Rivers State University, Port Harcourt (location F)

chicken suya from the various locations within and around the University. The microbial count varied significantly ( $P \leq 0.05$ ) for the different locations. Total coliform varied from 6.05 to 8.54 Log<sub>10</sub> CFU/g and was below detection limit in sample C and E. *S. aureus*, *Lactobacilli* and *E. coli* counts were in the ranges of 5.00 to 8.48, 4.00 to 8.48 and 4.00 to 8.52 Log<sub>10</sub> CFU/g, respectively. *Lactobacilli* and *E. coli* were below detection limit in sample Fb and C respectively. The counts from the location F which is within the university was significantly ( $P \leq 0.05$ ) the least except for *S. aureus* in sample C. This is an indication of the high level of hygiene practices by the processors and probably due to, the clean environment within the university, adequate supply and use of portable water and adequate storage facilities. However, the microbial load of the suya from Location F (Table 5) in comparison with the raw materials (Table 5) revealed the contamination of the chicken soya as the organisms were detected in the chicken soya and the raw chicken had none detectable levels. There were no significant ( $P \leq 0.05$ ) differences in the count between the raw beef and the beef suya for coliforms and *S. aureus* while *Lactobacilli* and *E. coli* were significantly ( $P \leq 0.05$ ) reduced. This is also an indicative of contamination of the suya product, although, the heat treatment would have led to decrease in the microbial count after processing.

### Antibiotic sensitivity of isolated organisms

Figure 2 showed the antibiotics sensitivity of the isolated *E. coli* on a gram negative disc. The gram negative disc had the following antibiotics: Septrin, Chloramphenicol, Sparfloxacin, Ciproflaxain, Amoxacillin, Augmentin, Gentamycin, Pefloxacin, Tarivid and Streptomycin. *E. coli* was resistant to Chloramphenicol and Streptomycin. The inhibition zones varied from 2.67 ± 0.00 mm for Gentamycin to 10.00 ± 0.95 mm for Ciproflaxain.

The antibiotics sensitivity of the isolated *S. aureus* on a gram positive disc is shown in Figure 3. The antibiotics contained in the gram positive disc were: Pefloxacin, Gentamycin, Ampiclox, Zinnacef, Amoxacillin, Rocephin, Ciprofloxacin, Streptomycin, Septrin and Erythromycin. *S. aureus* was resistant to Ampiclox and Amoxacillin. The

inhibition zones ranged between 1.85 ± 0.21 mm for Erythromycin and 11.70 ± 0.00 mm for Pefloxacin.

Ciprofloxacin and pefloxacin had significantly ( $P \leq 0.05$ ) the highest inhibition zones for both pathogens. The sensitivity of *E. coli* to ciproflaxain has been reported by Nutanbala *et al.* (2011). Ciprofloxacin and pefloxacin are fluoroquinolone antibiotics that have been reported to have excellent activities against gram negative bacteria such as *E. coli* and gram positive bacteria such as *S. aureus*. Tarivid and Sparfloxacin also belong to the fluoroquinolone antibiotics, but the inhibition zone of Sparfloxacin against *E. coli* (2.67 ± 0.94 mm) was significantly ( $P \leq 0.05$ ) lower than Ciprofloxacin (10.00 ± 0.95 mm), Tarivid (8.50 mm) and pefloxacin (8.17 ± 0.23 mm). Though, sparfloxacin is known to contain compounds with intermediate anti-anaerobic activity, there are also concerns about the increasing resistance of *E. coli* to fluoroquinolone antibiotics (Cohen *et al.*, 2017). The sensitivity of *S. aureus* to fluoroquinolone antibiotics (ciprofloxacin and pefloxacin) was reported by Sani *et al.* (2012). The fluoroquinolone based antibiotics act by inhibiting bacterial DNA gyrase responsible for DNA replication and transportation (Moore, 2015). This group of antibiotics had significantly ( $P \leq 0.05$ ) the highest inhibition zones as compared to the penicillin-class of antibiotics such as Amoxicillin and Rocephin that act against the bacterial cell wall synthesis. This confirms the report of Vanhoof *et al.* (1986) that ciprofloxacin and pefloxacin belonged to the most active group of drugs, generally most significantly more active than compounds such as ampicillin, chloramphenicol, co-trimoxazole and tetracycline that are very often used in the treatment of diarrheal diseases.

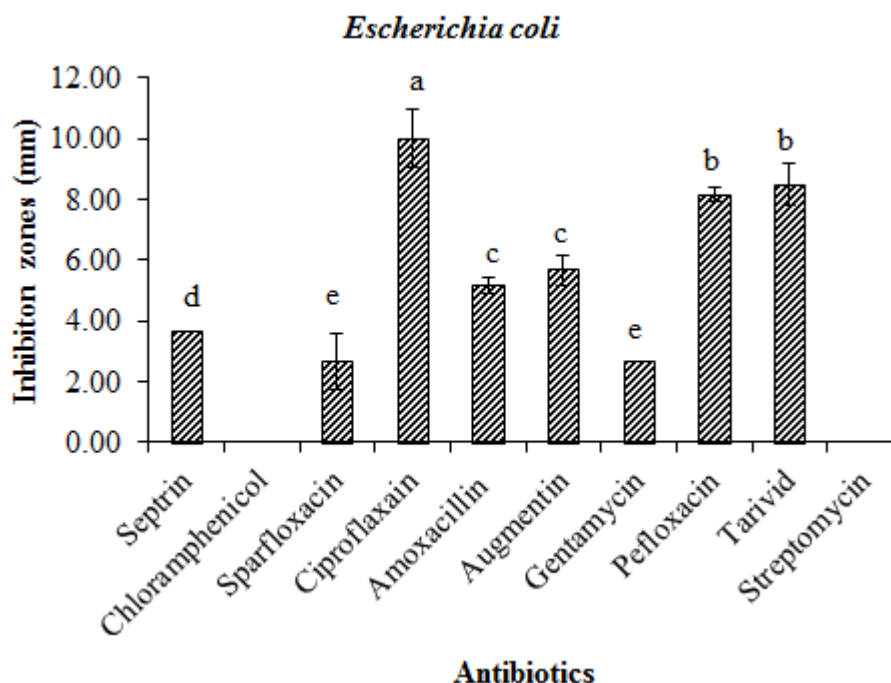
Amoxicillin, Ampiclox and Augmentin are penicillin-based antibiotics that binds to and inactivates penicillin-binding proteins (PBPs) located on the inner membrane of the bacterial cell wall. Inactivation of PBPs interferes with the cross-linkage of peptidoglycan chains necessary for bacterial cell wall strength and rigidity. This interrupts bacterial cell wall synthesis and results in the weakening of the bacterial cell wall and causes cell lysis (Niwa *et al.*, 2016). Recophin is a third generation cephalosporin antibiotic that acts like the penicillin-based antibiotics. It disrupts the synthesis of the peptidoglycan layer of bacterial cell walls through competitive inhibition on

**Table 5.** Microbial count (Log<sub>10</sub> CFU/g) of the beef and chicken suya from different locations within and around Rivers State University.

Location	Sample	Total coliform	<i>Staphylococcus</i>	<i>Lactobacillus</i>	<i>Escherichia coli</i>
Backgate I (Eagle Island)	A	7.36 ± 0.05 <sup>b</sup>	7.30 ± 0.05 <sup>c</sup>	8.48 ± 0.02 <sup>a</sup>	6.30 ± 0.05 <sup>c</sup>
Backgate II (Azikiwe)	B	8.54 ± 0.05 <sup>a</sup>	8.40 ± 0.03 <sup>a</sup>	7.18 ± 0.05 <sup>b</sup>	6.49 ± 0.02 <sup>b</sup>
Maingate I (Chinda Estate)	C	-	5.00 ± 0.05 <sup>e</sup>	5.30 ± 0.05 <sup>d</sup>	-
Maingate II (Npkolu)	D	7.38 ± 0.05 <sup>b</sup>	8.48 ± 0.02 <sup>a</sup>	6.85 ± 0.03 <sup>c</sup>	5.18 ± 0.03 <sup>e</sup>
Iloabuchi	E	-	7.48 ± 0.05 <sup>b</sup>	7.23 ± 0.05 <sup>b</sup>	8.52 ± 0.05 <sup>a</sup>
Staff club beef	*Fb	6.05 ± 0.05 <sup>d</sup>	7.26 ± 0.05 <sup>c</sup>	-	5.98 ± 0.05 <sup>d</sup>
Staff club chicken	*Fc	6.26 ± 0.05 <sup>c</sup>	6.52 ± 0.05 <sup>d</sup>	4.00 ± 0.05 <sup>e</sup>	4.00 ± 0.05 <sup>f</sup>

Values with the same superscript in the same column do not differ significantly ( $P \geq 0.05$ ). N = 3±SD

\*Fb and Fc represents beef and chicken suya from the Staff club in Rivers state University, Port Harcourt.

**Figure 2.** Antibiotics zones of inhibition (mm) against *E. coli* isolated from suya obtained from Rivers State University.

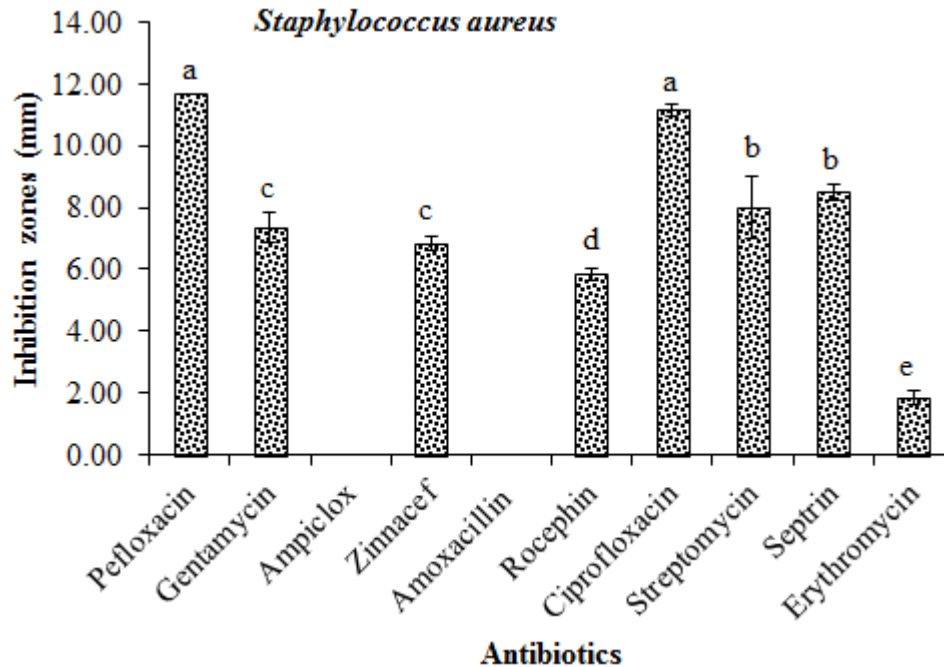
penicillin-binding proteins (Moore, 2015). *S. aureus* was sensitive to Rocephin but resistant to Amoxicillin and Ampiclox. This is in line with the reported by Sanni *et al.* (2012). *E. coli* however, was sensitive to Amoxicillin and Augmentin. Augmentin contains amoxicillin and clavulanic potassium. Although, Augmentin had an inhibition zone of  $5.67 \pm 0.47$  mm against *E. coli*, it was not significantly ( $P \leq 0.05$ ) different from that of Amoxicillin ( $5.17 \pm 0.24$  mm). The clavulanic potassium content of Augmentin is a beta-lactamase blocker. Beta-lactamase inhibitors are not antibiotics but rather block the enzymes that contribute to antibiotic resistance (Duda 2018).

*S. aureus* was sensitive to Zinnacef and Erythromycin. Zinnacef contains cefuroxime that inhibits cell wall

synthesis through the inhibition of  $\beta$ -lactamas (Page, 2012). Erythromycin had the significantly ( $P \leq 0.05$ ) least inhibition zone ( $1.85 \pm 0.21$  mm) against *S. aureus*, It is a macrolides based antibiotics that is bacteriostatic as it reversibly binds to the 50s ribosomal subunit inhibiting protein synthesis (Moore, 2015).

Gentamicin and streptomycin belong to the aminoglycoside based antibiotics (Hardman *et al.*, 2017). They bind irreversibly to the 16S rRNA subunit of the 30S ribosome and inhibit bacterial protein synthesis (Armstrong *et al.*, 2102). *S. aureus* was sensitive to both aminoglycoside antibiotics:  $8.00 \pm 0.99$  mm for streptomycin and  $7.35 \pm 0.49$  mm for gentamicin but Streptomycin had no inhibitory effect on *E. coli*. The inhibition zone of Gentamicin against *E. coli* ( $2.67 \pm 0.00$





**Figure 3.** Antibiotics inhibition zones (mm) against the *S. aureus* isolated from suya obtained from Rivers State University.

mm) was significantly ( $P \leq 0.05$ ) the least amongst others in the gram negative disc. The resistance of gram negative bacteria to antibiotics is usually attributed to the induction, mutation or by acquisition of R-plasmids. *E. coli* was resistant to Chloramphenicol an amphenicol based antibiotics that interferes with bacterial protein synthesis. Farzana *et al.* (2009) also reported the resistance of *E. coli* to chloramphenicol. Bacteria that are resistant to chloramphenicol are reported to produce chloramphenicol acetyltransferase (CAT) (Powell and Livermor, 1988). The resistance of bacteria to antibiotics can also be due to the inability of the antibiotics to reach their site of action (El-maali *et al.*, 1993).

## CONCLUSION

The microbial load of the samples revealed improper suya processing methods, poor hygienic condition of processors and the environment. The isolated bacterial were *Coliform*, *Staphylococcus aureus*, *Lactobacilli* and *Escherichia coli*. Streptomycin and Chloramphenicol had no inhibitory effect on *E. coli*. *S. aureus* was resistant to Amoxicillin and Ampiclox. The fluoroquinolone based antibiotics: ciprofloxacin, Pefloxacin and Tarivid had significantly ( $P \leq 0.05$ ) the highest inhibition zones against both pathogens. Though the bacterial were sensitive to most of the antibiotics, proper processing, hygienic practices and adequate supply of portable water and good storage facilities will minimize contamination and ensure safe suya for consumption.

## ACKNOWLEDGEMENT

The authors appreciate the technical assistance of Miss Ernest Gbbari Nwidebom of the Food Microbiology Laboratory Unit of the Department of Food Science and Technology, Rivers State University, Port Harcourt.

## REFERENCES

- Abdullahi IO, Umoh VJ, Ameh JB, Galadima M (2004). Hazards Associated with Kilishi Preparation in Zaria, Nigeria. *Niger. J. Microbiol.* 18(1-2):339-345.
- AOAC (2005). *Official Methods of Analysis*. Association of Official Analytical Chemists, Washing D.C Association of official analytic chemists.
- Armstrong ES, Kostrub CF, Cass RT, Moser HE, Serio AW, Miller GH (2012). Aminoglycosides, in: T.J. Dougherty, M.J. Pucci (Eds.), *Antibiotic Discovery and Development*, 1, Springer, New York, pp. 229-269.
- Cheesebrough M (2000). *District laboratory practice in topical countries part 2*. Cambridge University press.
- Chukwura EI, Mojekwu CN (2002). Prevalence of Microbial Contaminants of suya meat sold in Akwa Urban. *J. Trop. Microbiol.* 11:89-91.
- CMC (2004) Canadian Meat Council Fact Sheet on Sodium in Processed Meat Products. 305 - 955 Green Valley Crescent, Ottawa ON K2C 3V4. [www.cmc-cvc.com](http://www.cmc-cvc.com).
- Cohen J, Powderly WG, Opal SM (2017). *Infectious diseases*. 4ed. Elsevier Ltd. ISBN 978-0-7020-6285-8. <https://doi.org/10.1016/C2013-1-00044-3>.
- Duda KRN (2018). Choosing the right antibiotic for an infection. Verywellhealth. <https://www.verywellhealth.com/difference-between-amoxicillin-and-augmentin-4115052>.
- Edema MO, Osoho AT, Diala CI (2008). Evaluation of Microbial Hazards Associated with the Processing Suya (A grilled meat product). *Sci. Res. Essay.* 3(12):621-622.

- Egbebe AO, Seedu KJ (2011).** Microbiological Evaluation of Suya (dried smoked meat) sold in Ado and Akure, South West Nigeria. *Eur. J. Exp. Biol.* 1(4):1-5.
- Egyptian Organization for Standardization and Quality Control "EOS" (2005).** Egyptian Organization for Standardization and Quality Control. No. 1522 for frozen meat.
- Eke SO, Irabor JI, Okoye M, Aitufe OF, Ekoh SN (2013).** The microbial status of commercial 'Suya' meat products in Ekpoma, Edo, Nigeria. *Int. J. Com. Res.* 2(1):18-21.
- EI-maali NA, Ali AMM, Ghandour MA (1993).** Electrochemical reduction and oxidation of two cephalosporin antibiotics: Ceftriaxone (rocephin) and cefoperazone (cefobid) *Electroanalysis* 5(7):599-604.
- FAO/WHO (Food and Agriculture Organization) (2003).** Assuring Food Safety and Quality; Guidelines for Strengthening National Food Control System. *Food Nutr.* p. 76.
- Food and Agriculture Organisation of United Nations, FAO (1998).** Street Foods: Technical Meeting Report (Food and Nutrition Paper). Food and Agriculture Organisation of United Nations. Rome, Italy. ISBN13: 9789250039596.
- Farzana K, Akhtar S, Jabeen F (2009).** Prevalence and antibiotic resistance of bacteria in two ethnic milk based products. *Pak. J. Bot.* 41(2):935-943.
- Field RA, (2002).** Enteric Foodborne Illnesses. *Adv. Food Res.* 27:28-35.
- Hardman B, Hunt DJ, Mojaab D, Naor A (2017).** Sub-lethal gentamicin treatment of *Escherichia coli* UB1005 induces the release of soluble factors that reduce susceptibility to T7 bacteriophage infection. *J. Exp. Microbiol. Immunol.* 21:128-133.
- Harrigan WF (1998).** Laboratory methods in Food Microbiology, (edn) Academic press Limited, London.
- Hassan IA, Emun HO, Adekunle EO (2014).** Microbial quality of ready to eat barbecue meat (Suya) sold on the streets of Lagos State. *Int. J. Adv. Pharm. Biol. Chem.* 3(4):973-982.
- Hemmat MI, Reham AA, Omima AS, El Shafay MS (2013).** Quality of beef and edible offal at abattoir level. *Benha Vet. Med. J.* 25(2):254-263.
- Iyang CU, Inyor MA, Uma EN (2005).** Bacteriological quality of a Smoked Meat Product (Suya). *Niger. Food J.* 23:239-242.
- Jay MJ (2000).** *Modern food microbiology.* 6ed. Aspen publishers, Inc Gaithersburg, Maryland.
- Jonathan SG, Adeniyi MA, Asemoloye MD (2016).** Fungi biodeterioration aflatoxin contamination and nutrient value of Suya spices. *Trade Sci. Chem.* 9(4):279.
- Moore D (2015).** Antibiotic Classification and Mechanism. <https://www.orthobullets.com/basic-science/9059/antibiotic-drugs>.
- Niwa T, Morimoto M, Hirai T, Hata T, Hayashi M, Imagawa (2016).** Effect of penicillin-based antibiotics, amoxicillin, ampicillin, and piperacillin, on drug-metabolizing activities of human hepatic cytochromes P450. *J. Toxicol. Sci.* 41(1):143-6. doi: 10.2131/jts.41.143.
- Nutanbala NG, Hiren RT, AlpshPur PG, Tejas KP, Tripathi CB (2011).** Antibiotic sensitivity profile of bacterial pathogens in postoperative wound infections at a tertiary care hospital in Gujarat, India. *J. Pharmacol. Pharmacoth.* 2(3):158-164.
- Odey MO, Mboso EO, Ujon UP, Johnson JT, Gauje B, Ategwu MA (2013).** Microflora Analysis of Selected Meat and Meat Products from Calabar Cross Rivers State, Nigeria. *Arch. Appl. Sci. Res.* 5(3):50-56.
- Odusole KA, Akinyanju OO (2003).** Red Suya syndrome-acute Intravascular Administration and control. *Cons. Saf. Bull.* 2(2):20-24.
- Ogbonna IO, Danladi MS, Akinmusir O, Odu CE (2012).** Microbial Safety and Proximate Composition of Suya Stored from Maiduguri, Northern Nigeria. *Int. J. Food Saf.* 14:11-16.
- Olayiwola IO, Oganah BC, Oguntona CRB, Popoola AR, Sanni SA, Wobo SO (2013).** Status of aflatoxin and anti-nutritional contents of standardized maize-based dishes/snacks consumed in Nigeria. *Dis. J. Agric. Food Sci.* 1(5):93-96.
- Omojola AB, Kassim OR, Adewum MK, Ogunsola OO, Adiyemo GO, Deshiyan AB (2004).** Evaluation of the effect of variation in ingredient composition on the eating quality of Suya. *Afr. J. lives stock Ext.* 3:28-38.
- Orogu JO, Oshilim AO, (2017).** Comparative study of bacteriological analysis in hawked Suya meat and Suya meat on a Barbeque stand. *J. Microbiol. Res.* 3(1):5-8. DOI: [http://dx.doi.org/10.18685/EJMBr\(3\)1\\_EJSR-16-025](http://dx.doi.org/10.18685/EJMBr(3)1_EJSR-16-025).
- Page MGP (2012).** Beta-Lactam Antibiotics. In: Dougherty T., Pucci M. (eds) *Antibiotic Discovery and Development.* Springer, Boston, MA ISBN 978-1-4614-1399-8.
- Sani RA, Garba SA, Oyewole OA, Ibrahim A (2012).** Antibiotic Resistance Profile of Gram Positive Bacteria Isolated from Wound Infections in Minna, Bida, Kontagora and Suleja Area of Niger State. *J. Health Sci.* 2(3):19-22.
- Urmila K, Li H, Chen Q, Hui Z, Zhao J (2015).** Quantifying of total volatile basic nitrogen (TVB-N) content in chicken using a colorimetric sensor array and nonlinear regression tool. *Anal. Meth.* 7(13):5682-5688.
- Vanhoof R, Hubrechts JM, Roebben E, Nyssen H J, Nulens E, Leger J, De Schepper N (1986).** The Comparative activity of Peffloxacin, Enoxacin, Ciprofloxacin and 13 other antimicrobial agents against Enteropathogenic Microorganisms. *Infection* 14(6):294-298.
- Vilar I, Gracia ML, Fontar B, Tornadgo ME, Carballo J (2000).** A survey on the microbiological changes during the manufacture of dry-cured lacon, a Spanish traditional meat product. *J. Appl. Microbiol.* 89:1018-1028.