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Isolation, Characterization and Identification of Bacteria Associated with Suya Meat Sold in Owerri City, Imo State, Nigeria

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Abstract

This research was carried out to Isolate, Characterize and Identify Bacteria associated with Suya Meat sold in Owerri City, Imo State, Nigeria. Samples were collected in raps of aluminium foil and were then taken to the microbiology laboratory of the Department of Science Laboratory Technology of the Federal University of Technology Owerri, for analysis. The samples were homogenized, 5g of each Suya sample was measured and put in a sterile petri dish and 10 ml of distilled water was added to it and then filtered with filter paper into a conical flask, with a wire loop the samples were cultured on different media, McConkey agar, nutrient agar, man's Rogosa sharp (MRS) agar and Eosin methylene blue (EMB) and later incubated at 37^{o}C for 24 hours. Discrete bacteria colonies were observed and each colony was gram stained and examined microscopically. Biochemical tests were carried out to confirm the particular bacteria present. Results obtained showed that Bacteria isolated were *Staphylococcus aureus*, *Lactobacillus*, *Escherichia coli* and *Salmonella species.* Most of these microorganisms isolated are pathogenic and are toxic when in contact with suya meat and are capable of causing illness. *Staphylococcus aureus* produces air exotizcin when ingested by humans which causes intoxication manifesting with acute diarrhoea, vomiting and gastroenteritis. Proper handling of suya meat before selling is recommended.

Keywords: Beef Meat, Pathogenic Bacteria, Isolation, Hygiene

Introduction

The protein requirement of man may be obtained from either animal or plant sources (Aycicek et al., 2020). Meat is the common term used to describe the edible portion of animal tissues and any processed or manufactured products prepared from these tissues. Meats are often classified by the type of animal from which they are taken (Chidinma, 2020). Animal protein contains the essential amino acids tryptophan and lysine, which plants lack. Fat of meat also contains the essential fatty acid, linoleic acid that plants lack. For these reasons, animal proteins have a greater biological value than proteins of plant origin (Gwida et al., 2014). There are different types of meat from different types of animals such as pork meat (pig), mutton meat (sheep), and beef (cow) (Armitage, 2001). Meat is excellent in supplying quality protein, vitamins and minerals (Magwira et al., 2021). Thus, they are essential for the growth, repair and maintenance of the body cells which is for our everyday activities (Osuji et al., 2024). Consumption of meat can be traced back in history to the period when primitive man ate the raw flesh of animals (Egbebi & Seidu, 2011). But as he developed, he domesticated wild animals. Beef has been a major supply of meat in Nigeria as a result of extensive and semi-intensive cattle production.

According to Achi and Madubuike, (2007), Suya is a street vendor food which provides a source of inexpensive, convenient and often nutritious menus for cities, urban and rural areas; a major source of income for a vast number of people and creates opportunities for self-employment. Humans have hunted and killed animals for meat since prehistoric times (El-Sheikh & Amr, 2018). The advent of civilization allowed the domestication of animals such as chickens, sheep, rabbits, pigs and cattle. This eventually led to their use in meat production on an industrial scale

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with the aid of slaughterhouses. Meat is mainly composed of water, protein, and fat. It is edible raw but is normally eaten after it has been cooked and seasoned or processed in a variety of ways. Development in the production and consumption of"suya" in Nigeria has made it of great concern to study and know its microbial quality. This study is aimed at isolating and identifying bacteria associated with "suya" sold in Owerri Imo State Nigeria.

Materials and Methods

The study was carried out within the Owerri metropolis from May to July 2022. Owerri is the capital of Imo State in Nigeria, located in the heart of Igbo land. It is also the state's largest city, followed by Orlu and Okigwe. Owerri consists of three Local Government Areas including Owerri Municipal, Owerri North and Owerri West, it has an estimated population of about 1,401,873 as of 2016 (National Population Commission, 2016) and is approximately 100 square kilometres (40 sq miles) in area. Owerri is bordered by the Otamiri River to the east and the Nworie River to the south. Samples were collected in Douglas (Ama Hausa), MCC (Ama wire) and Eziobodo. A total of 60 samples were collected from 3 randomly selected Suya spots; within the Owerri metropolis, these spots were Douglas (Amahausa, MCC road, and Eziobodo of which 20 samples were taken from each location. The suya samples were wrapped in aluminium foil put in a sterile plastic plate to avoid contamination and brought to the laboratory for microbial examination.

The equipment used in this study includes a weighing balance, normal saline, an Anaerobic jar, a conical flask, filter paper, cotton wool, a measuring cylinder, a spatula, forceps, a swab stick, Aluminum foil, a wire loop, a Bunsen burner, petri dish, test tubes, microscope, Autoclave and incubator. Mann-Rogosa and Sharpe (MRS) media, hydrogen peroxide, Normal saline, sodium azide, Gram staining reagents, lactose, D-glucose, maltose, peptone water, urease, Simmons citrate, Kovac's reagents, oil immersion, and Boromoceresol purple.

This followed the method described by Skjerdal et al. (2021). Pieces of suya from each sample were removed and mashed in a sterile laboratory-type mortar and pestle into a paste. Ten percent of the stock solution was prepared by weighing 10g into 100ml of sterile buffered saline, properly shaking and sieving with filter paper into a conical flask. The Suya samples were inoculated in the different prepared media (Nutrient Agar, MackeyAgar, Eosin methylene blue (EMB) and Man's rogosa sharp (MRS) with a wire loop, covered with aluminium foil and incubated at 37°C for 24hrs to 48 hrs to determine the bacteria in it.

Microorganisms growing on solid surfaces tend to form colonies with distinctive morphologies. The variation in bacterial colony morphology can be observed with the naked eye (colony shape, appearance, colour, edge, elevation) and Gram Staining. This morphological variation includes shape, odour, and elevation surface on agar media. Specific biochemical tests were carried out to identify the isolates distinctively. The biochemical tests include gram staining, indole, catalase, coagulase, motility, citrate and carbohydrates fermentation, methyl red/voges-Proskauer of the method of Matos et al. (2006)

Characterization and identification of the colony isolates were achieved by initial morphological examination of the colonies in the plate (macroscopically) for colonial appearance, size, elevation, form, edge, consistency, colour, and odour, (Okonko et al.,2019). The isolates were characterized and identified based on their Biochemical Tests such as Catalase (Ca) Test, Indole (Idn) Test, Oxidase (Ox) Test, Motility (Mo) Test, Citrate (Cit) Test, Urease (Ur) Test, Methyl Red (MR) /Voges-Proskauer (Vp) Test, Carbohydrate, Fermentation Test were carried out using the method of Olodu et al. (2024).

Results

The mean number of bacteria isolates identified in Ama Hausa Douglas road Owerri(8.36cfu/s) was higher than MCC road (5.11 cfu/g) and the Eziobodo (3.28cfu/g) respectively at P=0.05 level.

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Table 1: bacteria isolates identified from the study location.

Locations	Number of bacteria (cfu/s)
MCC Road	5.11± 0.39
Ama Hausa	8.36± 0.58
Eziobodo	3.28 ± 0.21

Distribution of Isolated Bacteria in Various Locations

The distribution of isolates shown in Table 2 the most contaminated area was Ama Hausa with three bacteria spp. Identified.

Table 2: Distribution of bacteria isolates

Location	Isolates	Number of Isolate	Occurrence (%)
MCC	*S. aureus*	2.4	9.16
	L.bacillus	2.1	8.01
Ama Hausa	*E.coli*	4.5	17.18
	S.aureus	3.8	14.50
	Salmonellaspp.	5.3	19.85
Eziobodo	*S.aureus*	3.9	14.89
	E.coli	4.3	16.41

Morphological characteristics of isolates

The morphological characteristics of the bacteria isolate show the difference shapes, colour, their edges and opacity. Some are round, flat circular and rod in shape, while some are dark, Grey, white and milky in colour which can be seen with the naked eye or with the aid of a microscope.

Table 3: Morphological features of isolates

Location	Colony colour	Elevation	Edge	Opacity
MCC road	Milky	Flat	Entire	Opaque
	White	Round	Entire	Opaque
	Milky	Flat	Entire	Opaque
	White	Round	Entire	Opaque
Ama Hausa	Milky	Flat	Entire	Opaque
	Dark-purple (EMB)	Rod	Entire	Opaque
	Greyish white	Circular	Entire	Opaque
	Milky	Flat	Entire	Opaque
Eziobodo	Milky	Flat	Entire	Opaque
	Greyish white	Circular	Entire	Opaque

Grams reaction and Biochemical tests

Gram staining is a common technique used to differentiate gram-positive and gram-negative organisms. Biochemical tests are the tests used for the identification of bacteria species based on the differences in the biochemical activities of different bacteria.

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Table 4: Gram reaction and Biochemical tests

Samples	Cellular characteristics(Grams rxn)	Ca	Ox	MR	Ur	Vp	Cit	Ch	H_2s	Co	Ind	Mo	Suspected Microorganisms
A^1	+	+	-	-	+	+	+	-	-	+	-	+	*S. Aureus*
	+	-	-	-	-	-	-	-	-	-	-	-	*L.bacillus*
A^2	+	+	-	-	+	+	+	-	-	+	-	+	*S. Aureus*
	+	-	-	-	-	-	-	-	-	-	-	-	*L.bacillus*
B^1	+	+	-	-	+	+	+	-	-	+	-	+	*S. Aureus*
	-	+	-	+	-	-	-	+	-	-	+	+	*E. coli*
B2	+	+	-	-	+	+	+	-	-	+	-	+	*S. Aureus*
	-	+	-	+	-	-	-	+	+	-	-	+	*Salmonella spp*
C^1	+	+	-	-	+	+	+	-	-	+	-	+	*S. Aureus*
C^2	-	+	-	+	-	-	-	+	-	-	+	+	*E.coli*

Discussion

In this study, microbial counts were on the high thresholds although they were observed to be within microbiological acceptable limits recommended by regulatory bodies for vented meat and meat products. This corroborates the findings of Ike and Ogwuegbu (2020). According to Ike et al. (2017), retailed meat and meat products are normally sold in markets in unhygienic conditions, most often on open tables. The presence of bacteria in cow Suya meat could be a result of poor processing methods, poor hygiene practices, improper and unhygienic handling of meat products, bad sanitation operation and use of unclean utensils. Onuorah et al. (2021), stated that one of the major sources of contamination arises from the handlers during preparation and display of meat products for sale.

However, an important factor which significantly contributes to the great increase in the count is the location of the retail outlets which are motor parks or railway stations where "cow suya meat" can be easily contaminated by aerial spores or bacterial spores carried in the air and several other insects, such as flies, which are uncountable at such sites. Torok et al. (2018) state that insects such as flies can cause contamination by continuous contact with the product, as well also dust particles from heavily contaminated atmospheres around marketplaces and motor parks. When cattle are slaughtered and processed, Escherichia, coli bacteria in their intestines can *get* on the *meat without* thoroughly cooking the meat, especially ground beef. Ground beef should be cooked until it is no longer pink and the juices run clear. Contamination of meat products could arise from spice and the use of contaminated equipment/ utensils, and that spice may also be heavily loaded with microorganisms which served as a good source of contamination of meat products. Generally, the increased level of bacterial load observed in this study could also be a result of contamination arising from slaughter procedures (during bleeding, skinning and handling). Yousuf et al. (2008) stated that contamination occurs from various sources notably the present states of abattoirs, slaughterhouses, slaughter procedures and practices that may result in heavily contaminated meat.

It is important to note that, *Staphylococcus aureus, Proteus, and Escherichia. coli*are mostly isolated in “cow suya meat" samples thus confirming contamination resulting from spice, therefore, most bacteria isolated in "cow suya meat" are of public health importance causing food-borne diseases, food poisoning and food intoxication (e.g. *Staphyloenterotoxemia*), dysentery, travellers' diarrhoea, abdominal disorder and pains, sore throats, mild fever and lots more while other are of no public health significance but signifies poor hygiene during processing and handling of meat products (Onuorah et al., 2021).

Conclusion

The study was carried out to isolate and characterize bacteria associated with beef suya meat in Owerri Imo State. Some bacteria isolated and identified in the “cow suya” meat product is of public health importance thus their presence in such meat products continues to be considered a major cause of gastro-intestinal disorders, food poisoning and food-borne diseases. Hence there is a need for improved production hygiene and public health awareness.

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