The Bacterial Contaminants Associated with Kilishi Sold within Minna Metropolis, Nigeria.

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Abstract

The bacteriological quality of dried sliced beef (kilishi) sampled from different retail outlets in Minna metropolis was determined in this study. A total of Eighty (80) Kilishi samples were collected using sterile aluminium foil from eight different locations within Minna metropolis, Niger State. The samples were analyzed using Pour Plate Method while Gram staining and Biochemical tests were carried out to identify the various isolates. The analyses revealed bacterial counts ranging from $2.0x10^4$ to 3.6×10^5 CFU/g for all the samples. The organisms isolated include *Bacillus subtilis* 34(35.41%), *Staphylococcus aureus* 18(18.75%), *Klebsiella sp* 16(16.67%), *Escherichia coli* 12(12.50%), *Pseudomonas aeruginosa* 10(10.42%) *and Proteus sp* 6(6.25%). The diversity and the high viable count of bacterial isolates observed in this study are alarming because of the public health implication. The study underscores the need for the strict enforcement of regulations regarding maintenance of public health to forestall potential outbreak of diseases among consumers.

Keywords. Kilishi, Sterile, Contamination, Isolates, Bacterial, viable, outbreak, diversity.

Introduction

Dried sliced beef locally known as kilishi in Nigeria and some parts of Africa, is a popular meat product that is widely consumed in Northern Nigeria. Its preparation involves cutting raw meat into flat forms after

which it is spiced, roasted and sundried to add flavour and taste[1]. Because of its distinct and exotic flavour, kilishi is now widely consumed not only in Nigeria but also in some part of the sub Saharan region.

Food-borne illnesses resulting from the consumption of foods contaminated with pathogens and/or their toxins accounts for a significant cause of morbidity and mortality globally[2]. According to the European Food Safety Authority[3], over 320, 000 cases are reported in Europe yearly while admitting that the real number of cases which include unreported cases may actually be much higher. In developing nations, food-borne pathogens are reportedly one of the major causes of illnesses and deaths. In Nigeria, the incidence of food borne pathogens is quite alarming and represents a significant source of public health concern[4,5]. Additionally, the risk of emerging infection which may occur through the transfer of animal pathogens to man is also quite high[6].

Meat is a well known source of food borne pathogens mainly because of its composition which is particularly ideal for the growth of a wide range of bacteria[7]. The widespread acceptance and distribution of meat products all over the world makes its contamination with pathogenic microorganisms a public health threat of high significance[8]. Under unfavourable conditions such as high ambient temperature, high humidity and shortage of portable water prevalent in most parts of Nigeria, poor handling practices dispose meat product to microbial contamination and rapid deterioration[9].

A major source of concern, from a public health standpoint is the unhygienic conditions under which kilishi is processed and retailed in Nigeria and other West African countries which is capable of exposing consumers to food borne illnesses[10]. The need to encourage the adoption of regulatory measures that will assure consumer' s safety necessitates this inquiry in to the role of kilishi in the promotion of food borne illnesses in Minna, Nigeria.

Study Area

Dried sliced beef (kilishi) samples were obtained from Eight (8) kilishi spots in different locations within Minna metropolis, Niger State. The sampling locations include Bosso market, Stadium road, Mobil area, Kure market, Maikunkele market, Maitumbi market, Sabon gari area and Dutsen Kura.

Collection of Samples

A total of 80 kilishi samples comprising of ten samples per locations were collected randomly in sterile aluminium foil paper and transported immediately to the laboratory for analysis.

Analysis of Samples

One gram of each sample was aseptically minced into smaller pieces using a sterile surgical blade and picked using sterile forceps into 9ml of sterile normal saline. A four-fold serial dilution was made of each kilishi sample after which the viable count was done using the pour plate method[11]. An aliquot containing 1.0ml of each diluted kilishi sample was introduced into a sterile Petri dish after which 20ml of molten nutrient agar at 45^{o}C was poured into the diluted sample in the Petri dish. The plate was gently swirled to ensure the proper mixing of the sample and the media. The plate was allowed to solidify and then incubated at 37^{o}C for 24hours. Counting of the colonies was done and the colonies were recorded as the number of colony forming unit per gram (CFU/g).

Characterization and Identification of Isolates

Individual colonies in the primary cultures were subcultured into Nutrient Agar (NA) and MacConkey Agar (MCA) to obtain pure cultures. The pure cultures obtained were stored as slants and used for the characterization and identification of isolates. Microscopy and biochemical tests were carried for the characterization and isolation of isolates.

Results

The result revealed that all of the eighty samples screened had bacterial growth. The isolates include *Staphylococcus aureus, Bacillus subtilis, Klebsiella sp, Escherichia coli, Pseudomonas aeruginosa and Proteus sp*.

Table 1: Viable count of bacterial colonies in Kilishi Samples

Locations	Aerobic count (CFU/g)	Coliform count (CFU/g)
Kure Market	$7.0x10^4$-$2.1x10^5$	$7.0x10^4$-$2.0x10^5$
Maitumbi	$4.0x10^4$-$1.9x10^5$	$2.0x10^4$-$1.4x10^5$
Mobil Area	$4.0x10^4$-$3.6x10^5$	$9.0x10^4$-$2.1x10^5$
Dutsen Kura	$2.0x10^4$-$1.8x10^5$	$2.0x10^4$-$9.0x10^5$
Stadium Road	$1.8x10^4$-$1.4x10^5$	$2.1x10^4$-$1.1x10^5$
Bosso Road	$5.0x10^4$-$2.1x10^5$	$4.0x10^4$-$1.4x10^5$
Maikunkele	$4.0x10^4$-$1.7x10^5$	$2.0x10^4$-$1.9x10^5$
Sabon Gari	$6.0x10^4$-$1.9x10^5$	$4.0x10^4$-$1.7x10^5$

Table 2: Frequency of Occurrence of Individual Organisms Isolated from Sampling Locations

Location									
Organism Isolated	KM	BM	SG	MB	MK	MT	SR	DK	Total
Bacillus subtilis	3	4	3	8	5	4	3	4	34
Staphylococcus aureus	2	2	3	2	1	3	2	3	18
Escherichia coli	1	2	2	3	2	1	0	1	12
Pseudomonas aeruginosa	1	3	0	2	2	1	1	0	10
Klebsiella sp	3	4	1	3	2	0	2	1	16
Proteus sp	0	1	0	2	1	0	0	1	06
Total									96

KEY: KM- Kure Market, BM- Bosso Market, SG- Sabon Gari Street, MB- Mobil Junction, MK- Maikunkele Area, MT- Maitunbi Area, SR- Stadium Road, DK- Dutsen Kura.

Table 3: Percentage Occurrence of Bacteria Isolated from Kilishi

Organisms Isolated	Frequency of Occurrence	Percentage of Occurrence
Bacillus subtilis	34	35.41
Staplylococcus aureus	18	18.75
Escherichia coli	12	12.50
Pseudomona aeruginosa	10	10.42
Klebsiella sp	16	16.67
Proteus sp	06	6.25
Total	96	100

Discussion

In this study, it was observed that all the kilishi samples screened without exception were contaminated with one or more bacterial species which makes the samples potential sources of food-borne illness. This result is similar to the findings of an earlier study[10] in which all the samples collected within Ilorin metropolis were also found to be contaminated with different species of bacteria. This very high level of contamination observed in the sample may be due to a combination of factors such as the quality of the meat source, unhygienic handling of meat and the total or near absence of formal education among the retailers. There is the unhygienic practice of buying and using sick animals for kilishi because they are cheaper than healthy animals. Meat handling is also done in largely unhygienic manner thereby exposing the meat to contaminants from the surrounding environment. Kilishi sellers are mostly middle age illiterate men without any formal education or training in food preparation as a result of which the skills necessary and important for hygienic handling of foods, especially for a nutritious food like kilishi made from a substrate that is also an excellent culture medium for a wide variety of microorganisms is absent.

Out of the eight locations where samples were collected, Mobil area had the highest contamination of aerobic count ranging from $4.0x10^4$ CFU/g to $3.6x10^5$ CFU/g and coliform count ranging from $9.0x10^4$ CFU/g- $2.1x10^5$cfu/g while Stadium road had the least level of contamination with aerobic count ranging from $1.8x10^4$cfu/g - $1.4x10^5$cfu/g and coliform count ranging from $2.1x10^4$cfu/g - $1.1x10^5$cfu/g. Mobil area

unlike most of the other area sampled is a busy junction that hosts tremendous human and vehicular activities as a result of which high level of dust is continually generated. The kilishi which is always exposed both during drying and selling is therefore made vulnerable to contamination by the airborne organisms thus generated.

The organisms isolated include *Bacillus subtilis, Staphylococcus aureus, Escherichia coli, Pseudomonas aeruginosa, Klebsiella* sp and *Proteus* sp. This is similar to the findings of an earlier study[10] with the exception of *Proteus* sp. This fact that *Bacillus subtilis* had the highest frequency of occurrence may be as a result of the ubiquitous nature of the organism and its endospore forming ability, which enables it to survive harsh conditions. *S. aureus* is a normal flora of the human skin and nares and may have been transmitted from handlers to the product through unhygienic practices at any stage of the kilishi processing. The presence of E. coli suggests contamination of fecal origin. Although E. coli has been shown to exhibit a high desiccation tolerance, contamination of kilishi with these pathogens could be prior to or during the drying of the meat. The isolation of *Pseudomonas* and *Klebsiella* species from the kilishi samples screened is an indication of possible post production contamination as these organisms are incapable of withstanding the high temperature treatment associated with roasting and drying of the kilishi. However, gram negative aerobic rod shaped bacteria especially *Pseudomonas* sp has been reported as a dominant meat spoilage organism[12, 13]. Contamination of this product by *Proteus* sp may be due to soil or water contamination during the processing and/or sale of the kilishi.

Conclusion

This study revealed that kilishi sold in Minna metropolis is highly contaminated. All the kilishi samples screened were contaminated. The findings of this study have a serious implication for public health management since the consumption of kilishi cut across the entire social strata of the society. It therefore underscores the need for intervention measures such as enlightenment campaigns and strict enforcement regulatory provisions regarding sale of food products.

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