OPEN ACCESS Freely available online

Journal of Food Microbiolgy, Saftey & Hygiene

Research Article

Microbiological Quality of Kilishi Sold in Nasarawa, Nasarawa State

Ada AI[1*], Isa Saadatu[2], Ada AO[3]

[1]*Department of Training and Development, Terraskills Learning Systems Limited, Abuja, Nigeria;*

[2]*Department of Agriculture, Agricultural Research Council of Nigeria (ARCN), Abuja, Nigeria;*

[3]*Department of Microbiology, School of Applied Sciences, Federal Polytechnic, Nasarawa, Nasarawa State, Nigeria*

ABSTRACT

This study was carried out to determine the occurrence of bacteria and fungi in already processed and ready-to-eat Kilishi sold in Nasarawa, Nasarawa State. The bacterial and fungal population was isolated, characterized and identified. Bacterial Isolates were identified based on their cultural and biochemical characteristics, whereas fungi isolates were identified based on their cultural and morphological characteristics. The bacteria isolated were *Escherichia coli, Salmonella spp., Staphylococcus aureus* and *Staphylococcus epidermidis.* Whereas fungi isolated were identified to include *Aspergillus niger, Aspergillus flavus, Aspergillus fumigatus* and *Candida albicans.* The aerobic bacteria count ranged from 3.4×10^4 cfu/g to 9.1×10^4 cfu/g in the three points studied. The total coliform bacterial counts on the other hand ranged from 3.2×10^2 cfu/g to 9.3×10^2 cfu/g. This study concluded that Kilishi was heavily contaminated with microorganisms with high microbiological counts, indicating that the meat product was not adequately processed and may have been done in unsanitary conditions. The presence of toxigenic microorganisms indicates that the meat product is unsafe for human consumption.

Keywords: Kilishi; Public health

INTRODUCTION

Meat contributes significantly to the diets of developing countries because it is a rich source of high-quality protein, minerals, essential vitamins, fats, and carbohydrates [1]. Meat is highly perishable because it contains an abundance of nutrients that promote the growth and proliferation of microorganisms [2]. Some of these microorganisms could render it unfit for human consumption. This may be one of the reasons why man has developed a number of meat preservation techniques over the years that can keep meat stable and extend its shelf life while maintaining adequate nutritional value and flavor [1,2].

Because of the nutritional quality of meat and its mass appeal, regions of the world where meat is consumed have a variety of meat products, the majority of which are ready to eat and sold on street corners and in shopping malls. Bacon, kebabs, sausages, corned beef, beef jerky, and canned meat-based sauces are examples of processed meat products [3]. However, different regions have processed meats that are unique to their culture. Suya and Kilishi are two locally processed meat products in Nigeria. Their popularity varies by region of the country.

Kilishi is a form of jerky made traditionally from defatted groundnut paste and spiced meat [4]. In Northern Nigeria, it is a popular meat product, made by partially sun-drying thin sheets of high-quality beef, then adding some ingredients before a second sun-drying and partial roasting period. The final product can be kept for months without visible spoilage and change of taste. However, studies have shown that even with the increase in shelf life, Kilishi is still subject to bacterial spoilage due to contamination during production [5]. The non-mechanized means of producing Kilishi particularly in a country with high ambient temperature, low humidity, shortage of potable water as well as poor handling practices, meat and meat products can be exposed to microbial contamination and therefore deteriorate rapidly [6].

Correspondence to: Ada AI, Department of Training and Development, Terraskills Learning Systems Limited, Abuja, Nigeria, Tel: 8100895523; E-mail: adainnocent@terraskills.com

Received: 05-Dec-2022, Manuscript No. JFMSH-22-25159; **Editor assigned:** 07-Dec-2022, PreQC No. JFMSH-22-25159 (PQ); **Reviewed:** 21-Dec-2022, QCNo.JFMSH-22-25159; **Revised:** 28-Dec-2022, Manuscript No. JFMSH-22-25159 (R); **Published:** 04-Jan-2022, DOI: 10.35248/2476-2059.22.7.187.

Citation: Ada AI, Saadatu I, Ada AO (2022) Microbiological Quality of Kilishi Sold in Nasarawa, Nasarawa State. J Food Microbiol Saf Hyg. 7:187

According to research on the sources of bacterial contamination in kilishi, the majority of contaminations are encountered during the various stages of slaughtering from inherent intestine microbiota, the prevailing environment, transportation, and handling [7,8]. Even after production, displaying kilishi in open markets and in trade basins can expose the product to environmental contamination.

Most ready-to-eat meat products, particularly suya and Kilishi, are frequently displayed in unsanitary conditions and thus contaminated by various microorganisms [9]. Kilishi production in Nigeria is not supervised or regulated, so the edibility, safety, and general quality vary from one producer to the next due to a lack of standardized methods of preparation that would ensure consistent product quality [10].

Despite increased attention from public authorities and food operators to food hygiene and food safety, consumption of foods contaminated with pathogenic microorganisms or their toxins remains one of the leading causes of disease hospitalization and economic loss [11]. As a result, the uncontrolled and unsupervised preparation of Kilishi may pose public health risks. Therefore, this study was carried out to determine the microbial quality of Kilishi sold within Nasarawa, Nasarawa State.

MATERIALS AND METHODS

Study area

This study was carried out in Nasarawa, Nasarawa State, Nigeria. Nasarawa State is situated between latitude 700 40' 0" N and 900 40' 0" N, and longitude 700 0' 0" E and 900 30' 0" E.

Samples collection: Freshly prepared dried roasted sliced meat samples (100 g each) were collected from three well known selling points (Sample A, B and C). The samples were aseptically placed in a clean, sterile aluminum foil immediately after collection and subsequently transported to the laboratory. Sampling was conducted according to availability and hygienic practices of processors and the environment.

Sample preparation and serial dilution

The sample (25 g) was weighed and homogenized by blending in 225 ml of buffered peptone water (HiMedia, India) at 15,000-20,000 rpm. This was further diluted serially up to 1:107[12].

Total aerobic bacterial count

The total aerobic mesophilic bacterial count was carried out according to [13] where 1 ml of Aliquot from 104 and 106 dilutions were transferred into duplicate Petri dishes. This was followed by pouring aseptically about 15 ml of molten agar. The culture was then homogenized by swirling the plates and later allowed to solidify. Inoculated plates were then incubated at 37°C for 24 h.

Isolation and identification of bacteria

Aliquots (0.1 ml) of each dilution of 104 and 106 was transferred in duplicates into Mannitol Salt Agar (HiMedia, India), MacConkey Agar (HiMedia, India), Eosin Methylene Blue Agar (HiMedia, India) and Desoxycholate Citrate Agar (Oxoid, England) and then spread uniformly using a sterile bent glass rod. The plates were then incubated aerobically at 37°C for 18 to 24 h. Discrete colonies were aseptically subcultured into fresh agar plates and a pure culture of each isolate was obtained. Pure isolates of resulting growth were stored for further characterization.

The isolates were characterized based on microscopic appearance, colonial morphology and Gram staining reactions as well as appropriate biochemical tests such as Catalase test [14,15], Coagulase test [16], Citrate utilization test [5], Motility test [16] and methyl red test.

Isolation of yeast and molds

Sabouraud Dextrose Agar (Hi-Media, India) containing Chloramphenicol (50 mg) was used for the isolation of fungi. The media was prepared following manufacturer's instructions. Inoculation was done using pour plate method and the inoculated plates were thereafter incubated at 28°C for 5 days. Emerging colonies were then counted, calculated and expressed as cfu/g and further identified through microscopy [14].

The Fungi were identified using the Lactophenol cotton blue technique. Aliquot of 100 μg of Lactophenol cotton blue was dropped on a frosted end precleaned glass slide. After which a wire loop was used to pick the colony and teased on the lactophenol drop. A cover slip was placed on the Lactophenol cotton blue and examined under×40 objective lens to check for the structure of the organism [15].

RESULTS AND DISCUSSION

This study was carried out to determine the microbial contamination of Kilishi sold within Nasarawa. Microbial contamination of food has become a public health concern because of the possibilities of causing harm to consumers as well as affects the livelihood of those that produce it. Due to its beneficial chemical composition, meat is a suitable medium for the development of all microorganisms [17]. The high protein content of meat which transforms into high nutrient and moisture content makes meat prone to bacterial invasion and fungal growth [7,8].

Total aerobic bacterial count of freshly prepared Kilishi samples

The result of the current study showed that there was a significant level of aerobic bacterial as well as coliform bacterial contamination in Kilishi sold within Nasarawa. The aerobic bacteria count ranged from 3.4×10^4 cfu/g to 9.1×10^4 cfu/g in the three points studied. The total coliform bacterial counts on the other hand ranged from 3.2×10^2 cfu/g to 9.3×10^2 cfu/g. This bacterial load is higher than the load recorded by Jabaka et

al. (2021) who reported a bacterial load ranging from 8.0×10^2 cfu/g to 3.5×10^3 cfu/g.

The high aerobic bacteria count in the Kilishi studied could be attributed to lack of good hygienic practice and the harsh dusty environmental condition characterized by heavy wind along with dust. This is also in line with the attribution of [5]. Similar reports have also been made. They attributed the high bacteria count to poor processing and unhygienic practices.

Although both aerobic bacterial counts and coliform counts were significant enough to question the safety of consumption, the aerobic bacterial counts were higher and signify poor production hygiene and handling. The presence of such high counts of coliform bacteria is also a clear indication that the Kilishi in this study were handled poorly and exposed to faecal contamination therefore, are unsafe for human consumption.

The presence of high numbers of coliform bacteria could be attributed to the contamination of meat through the process of slaughtering.

It is worthy to note that during production of Kilishi, meat samples are sliced directly from butchered meat without proper washing. This process allows the contaminated meat to go through the processes of slicing, and the eventual drying of the meat (Table 1).

Table 1: Total aerobic bacterial count of freshly prepared Kilishi samples.

Sample	Aerobic bacterial counts (cfu/g)			Total coliform count (cfu/g)		
	Point A	Point B	Point C	Point A	Point B	Point C
1	3.4×104	5.6×104	3.1×104	3.9×102	4.2×102	4.6×102
2	5.1×104	8.9 × 104	4.2×104	7.2×102	3.2×102	7.8×102
3	4.9×104	3.6 × 104	9.1×104	3.7×102	7.1×102	8.4×102
4	3.6×104	8.1 × 104	7.4×104	7.0×102	6.2×102	3.3×102
5	4.4×104	3.2 × 104	8.3×104	3.3×102	3.9×102	9.3×102

Effect of storage on the aerobic bacterial load in Kilishi

The effect of storage duration on the bacterial load of Kilishi was determined starting from the fresh samples and then to the same sample stored for 5 days, 10 days, 15 days and 20 days. The result showed a marked increase in the Kilishi stored for a period of 20 days.This shows that storage of Kilishi allowed for the growt h of bacteria. The total aerobic bacterial count showed that the co unts significantly increased from the tenth (10^{th}) day of storage.

Although Kilishi has been reported to have low levels of moisture [18], which have been reported to be the reason for the prolonged storage of Kilishi, the result of this study showed that Kilishi could not only support growth of microorganism due to its high nutrient but is prone to microbial spoilage due to the increasing number of bacterial counts observed after 10–20 days in storage. The increased microbial load observed after a period of storage is as a result of the presence of high microbial load in the freshly prepared Kilishi.

Although the moisture content of the Kilishi studied was not determined in the current study, the ability of bacteria to proliferate easily could also be that the Kilishi studied were not properly dried to adequately remove moisture which is important for microbial growth [6]. It has been stated by other studies that the quality of Kilishi varies from one producer to the other and from one batch to another from the same producer [18] (Table 2).

Table 2: Effect of storage on the aerobic bacterial load in Kilishi.

Sample	Storage duration (*cfu/g)				
	Freshly prepared	5 days old	10 days old	15 days old	20 days old
1	3.0×104	7.2×104	9.8×106	1.12×107	9.1×107
2	6.1×104	9.1×104	1.01×107	8.3×107	1.12×108
3	5.9×104	8.7×104	1.00×107	7.5×107	7.9×107
4	6.4×104	7.3×104	9.6×106	8.4×107	9.6×107
5	5.3×104	6.9×104	1.14×107	9.8×107	1.34×108
Mean	5.3×104	7.8×104	1.02×107	9.0×107	1.02×108

Identification of isolated bacteria

Based on the characteristics of microbes isolated in the current study, bacteria isolated were identified to include *Escherichia coli, Salmonella spp., Staphylococcus aureus* and *Staphylococcus epidermidis.* Whereas fungi isolated were identified to include *Aspergillus niger, Aspergillus flavus, Aspergillus fumigatus* and *Candida albicans.*

The result of this study agrees with the finding of the study of Tijani and Jumare (2014) who reported the presence of *Aspergillus fungi* and the presence of *Staphylococcus aureus* and *Salmonella typhi.* The microorganisms isolated in the current study also agree with the report of [19].

The presence of Escherichia coli indicates faecal contamination of the samples. The faecal contamination may have arisen from the faeces of the cattle during slaughtering. The presence of fungi such as *Aspergillus niger*, *Aspergillus flavus*, *Aspergillus fumigatus* (Table 4) is an indication of environmental contamination because of the ubiquitous nature of these fungi. The presence of *Aspergillus flavus* in Kilishi is a major concern because the fungus is capable of producing aflatoxins which are a group of highly toxic fungi metabolites [20] (Table 3 and 4).

All microbes isolated have been reported to be pathogenic except *Staphylococcus epidermidis* which is a commensal but could be an opportunistic pathogen especially in patients receiving medical devices causing approximately 20% of all orthopedic device-related infections [21-25].

Table 3: Biochemical Characteristics of isolated bacteria.

Test	**Characteristics of isolated bacteria**			
	Isolate 1	**Isolate 2**	**Isolate 3**	**Isolate 4**
Gram reaction	neg. rods	pos. cocci	neg. rods	pos. cocci
Indole	+	-	-	-
Methyl red	+	-	+	-
Citrate	-	-	-	-
Catalase	-	+	+	+
Motility	+	-	+	-
Coagulase	-	+	-	-
Presumed bacteria	*E. coli*	*S. aureus*	*Salmonella spp*	*S. epidermidis*

Key: + =positive, - =Negative

Table 4: Characteristics of fungi isolated.

Cultural features	**Microscopic features**	**Suspected fungi**
Fast growing colonies. Colonies were flat and covered with dense layer of black conidial heads.	septate hyphae with conidiophore with the apex forming globose vesicle, bearing conidial head.	*Aspergillus niger*
Fast growing colonies. Green surface with yellow reverse side	Chains of round conidia with compact conidia head. Phialides uniseriate, concentrated on the upper surface of the vesicle.	*Aspergillus flavus*
Fast growing colonies. Grey to green surface with yellow reverse side.	uniseriate vesicle with smooth and globose conidial	*Aspergillus fumigatus*
Small oval colony with creamy appearance.	Unicellular budding pseudo-hyphae	*Candida albicans*

CONCLUSION

According to the findings of this study, Kilishi was heavily contaminated with microorganisms with high microbiological counts, indicating that the meat product was not adequately processed and was done in unsanitary settings. The presence of organisms such *as E. coli, Salmonella spp., Staphylococcus aureus, Staphylococcus epidermidis, Aspergillus niger, Aspergillus flavus, Aspergillus fumigatus,* and *Candida albicans* indicates that the meat product has been tainted in the various preparation and distribution chains.

RECOMMENDATIONS

It is therefore recommended that;

- A systematic method of production is designed in such a way that Kilishi producers are educated on the various critical control points in the system.
- Kilishi is a meat product and like other meat products it is subject to deterioration, therefore, the batch number, production date and expiry or best before date should be appended in every product sent to the market.
- Since the Kilishi is sold in open markets and in open stalls, it is possible that contamination will be increased therefore, this practice should be discouraged.
- The method of production should also be upgraded to a more mechanized form that will ensure hygienic preparations.

REFERENCES

1. Mgbemere VN, Akpapunam MA, Igene JO. Effect of Groundnut Flour Substitution on Yield, Quality and Storage Stability of Kilishi - a Nigerian Indigenous Dried Meat Product. African J Food, Agri, Nutr Dev. 2011;11(2):4718-4738.
2. Wu B, Dahlberg K, Gao X, Smith J, Bailin J. Rapid Measurement of Meat Spoilage using Fluorescence Spectroscopy. Proceedings of the SPIE. 2017;(7):20-27.
3. Pearson AM, Tauber FW. Processed Meats. Springer Science and Business Media. Science and Engineering. 2012;427.
4. Olusola OO, Okubanjo AO, Omojola AB. Nutritive and Organoleptic Characteristics of Kilishi as Affected by Meat Type and Ingredient Formulation. J Animal Produc Adv. 2012;2(5):221-232.
5. Jabaka RD, Ododife Q, Daniel AD, Nuhu UD, Doro EJ. Assessment of Bacteria and Parasite Contamination of Dried Sliced Beef (Kilishi) Sold within Birnin Kebbi Metropolis, Kebbi State, Northern Nigeria. South Asian J Res Microbiol. 2021;8(3): 58-66.
6. Okonko IO, Odu NN, Igboh IE. Microbiological Analysis of Kilishi Sold in Port Harcourt, Nigeria. New York Sci J. 2013;6(7): 37-43.
7. Ike CC, Akortha EE. Microbial diversity associated with different fresh meats sold in Aba metropolis, Abia State, Nigeria. Int J Res Develop Org. 2017;3(5):108-121.
8. Ike CC, Akortha EE. Microbial diversity associated with different fresh meats sold in Aba metropolis, Abia State, Nigeria. International Journal of Research and Development Organization (IJRDO). J Biol Sci. 2017;3(5):108-121
9. Hassan EL, Umar FI, Yahaya SF, Ali M. Microbial Quality Assessment of Processed Meat Product (Tsire) Sold Within Wudil Town, Wudil Local Government Area, Kano State, Nigeria. Modern Appl Pharm Pharmacol. 2018;2(2):1-7.
10. Igene JO, Farouk MM, Akanbi C. Preliminary Studies on the quality and drying Characteristics of "suya". Nigeria Food J. 2009;7:29-38.
11. Dahiru AT, Maigari AK. Bacteriological Quality Assessment of Kilishi Produced in Kunchi Local Government Area, Kano State, Nigeria. J Microbiol Res. 2019;4(1):12-18.
12. Daminabo V, Isun R, Agarry OO. Isolation of enterococci from dried beef crackers (kilishi) and its antibiogram. African J Parasitol Res. 2017;4(1):191-196.
13. Abdullahi IO, Umoh VJ, Galadima M. Hazards Associated with Kilishi Preparations in Zaria. Nigerian J Microbiol. 2004;18(1-2): 339-345.
14. Cheesbrough M. District laboratory practice in tropical countries, part 2. Cambridge university press; 2005.
15. Cheesbrough M. District laboratory practice in tropical countries, part 2. Cambridge University Press, Cambridge, UK. 2006;137-150.
16. Oluntiola PO, Famurewa O, Sontag HG. Introduction to general microbiology: A practical approach. 2nd Edition, Bolabay Publication, Ikeja. Nigeria; 2000.
17. Shamsuddeen U, Ameh JB. Survey on the possible critical control points in kilishi (a traditional dried and grilled meat snack) produced in kano. Int J Biosci. 2008;3(2):34-38.
18. Inusa SK, Said IS. Evaluation of the Chemical and Microbiological Properties of Kilishi Sold in Kano Metropolis. J Dryland Agri. 2017;3(1):59-69.
19. Kasimu S, Ibrahim AS, Naziru S. Mycological and mycotoxicological quality assessment of dried meat (Kilishi) sold in Kebbi state, Nigeria. World J Adv Res Review. 2020;08(01):006-011.
20. Tirmenstein MA, Mangipudy R. Alfatoxin. Encyclopedia of Toxicology. 2014;104-106.
21. Apata ES, Osidibo OO, Apata OC, Okubanjo AO. Effects of different solar drying methods on quality attributes of dried meat product (Kilishi). J food res. 2013;2(1):80.
22. Felegan CR, Akoja SO, Oyarekua MA. Mocrobiological assessment of suya (sliced roasted beef) in Ado-Ekiti metropolis, Ekiti state Nigeria. MOJ Biol Med. 2017;2(3):266-269.
23. Isah OA, Okubanjo AO. Effects of various additives on proximate composition and acceptability of kilishi made from semi tendinosus of white Fulani cattle. The Pacific J Sci Technol. 2012;13(1):506-511.
24. Sabaté Brescó M, Harris LG, Thompson K, Stanic B, Morgenstern M, O'Mahony L, Richards RG, Moriarty TF. Pathogenic mechanisms and host interactions in Staphylococcus epidermidis device-related infection. Frontiers microbiol. 2017;8:1401.
25. Tijani AO, Jumare S. Microbiological quality assessment of samples sold in Kaura Namoda. Int Conf Earth, Environ Life Sci. 2014;23-24.