

**Proximate Analysis and Mycological Assessment of Suya Sold in Kabuga, Kano State, Nigeria***¹Albert O. Fasogbon, ²Joseph O. Odewade, ³Lydia O. Odewade¹Department of Microbiology, Bayero University, Kano, Nigeria.²Department of Microbiology, Federal University, Dutsin-Ma, Katsina, Nigeria.³Department of Environmental Management, Bayero University, Kano, Nigeria.*Corresponding Author's email: odewadejoseph@yahoo.com

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Abstract

Suya is a cherished food delicacy which can serve as a source of infection as it can harbor pathogens of clinical importance. However, there is paucity of data on mycological assessment and proximate analysis of suya sold within Kabuga area, Kano State, Nigeria. Therefore, this study examined the proximate composition and mycological qualities of suya sold within this area in Kano State. A total of thirty-six samples were randomly collected from selected suya spots from the study area and microbiologically analyzed. Total fungal counts, identification of fungi and proximate analysis were NH determined using standard microbiological techniques. A total of fifty-one (51) fungal isolates were obtained from all the suya samples collected. The highest frequency of occurrence was shown by *Aspergillus* spp (16) while *Fusarium* spp had the lowest frequency of occurrence (10). Proximate analysis revealed the mean percentage of moisture as 24.00 - 48.00%, crude protein (24.64 - 46.32%), crude fiber (5.25 - 8.75%), fat (8.80 - 17.30%), carbohydrate (0.11 - 14.58%) and ash (1.05 - 2.45%) contents. This study showed the presence of *Penicillium*, *Aspergillus*, *Fusarium* and *Rhizopus* spp in suya samples examined and hence the need to improve on good hygiene practice by the suya vendors to control associated health risk.

Keywords: Proximate analysis, Mycological assessment, Fungi, Microbiological techniques, Vendors**1. Introduction**

The growing microbial contamination of food is of global public health significance as it results into various food borne diseases. It was reported that about 325, 000 people were hospitalized while 5,000 deaths occurred annually in the United States due to food-borne diseases [1]. In the United Kingdom, Gormely *et al.* [2] reported about 2429 food-borne outbreaks in England from 1992 to 2008. Reliable statistics on food borne diseases have not been reported in developing countries like Nigeria due to poor or non-existing reporting systems [3].

Traditionally, meat obtained from cow, chicken, cattle, pig and sheep may be processed into various meat products for human consumption [4]. In Nigeria, one such products is Suya.

Suya is a traditional meat product which can be prepared on sticks and spiced with ingredients such as vegetable oil, cake, peanut, salt, and maggi after which it is then roasted [5]. It is usually made by the Hausa people whose preoccupation is farming and rearing of animals; the latter serves as a source of income for the people. This resulted into the production of beef products such as Kundi, Kilishi, Balangu and Suya, which are commonly vended on the street. Suya is, however, the most popular as it is widely consumed by different people of different cultural backgrounds [5].

Suya can serve as a good medium that supports the growth of numerous microorganisms since it is prepared basically from animal meat [6]. Meat contains high percentage of moisture, nutrient and a pH close to neutral. Because of these, it is classified as a perishable food. Meat spoilage may affect the organoleptic properties of the meat as it may result to bad taste, oxidative rancidity, sliminess and discoloration. The kind and amount of spoilage microorganisms in meat depends upon the availability of nutrients, pH, temperature, oxygen, moisture content, generation interval of the spoilage microorganism and other growth factors under given environment upon storage [7]. The fact that sporadic cases of gastroenteritis and symptoms of food infection do occur after consuming meat / meat products is an indication that contaminated meat product can constitute a food safety risk [5].

The risks of taking foods that are unsafe have been well documented. According to a report in 2013, contaminated foods were estimated to cause 1.5 billion diarrhea cases and 1.5 million mortalities every year in children [8]. According to the researchers, suya is one of such contaminated foods hawked on the street causing high level of microbial contamination [9-11]. The contaminants may be environmental, chemical or physical contaminants [12, 13] found within suya samples which pose public health concerns to those who consume it.

Contamination by microbes (usually fungi) was reported to cause food spoilage and illness [14, 15]. Fungi are cosmopolitan as explained by Jonathan *et al.* [16]. Most filamentous fungi usually Deuteromycota fungi such as *Fusarium*, *Penicillium*, *Rhizopus*, *Mucor* and *Aspergillus* have been isolated from street vended foods and spices [14, 15, 17]. Fungi (usually molds) are implicated in food spoilage activity, which results in the reduction of organoleptic properties of food by introducing metabolites into the foods under favorable condition so as to prevent humans and other organisms from eating them [15]. Hence, making the food to be toxic due to the production of mycotoxin (a metabolite) such as aflatoxin mostly produced by *Aspergillus* species. Hence, the research was designed to assess the proximate composition and mycological quality of suya sold in Kabuga area, Kano State, Nigeria.

2. Materials and Methods

2.1 Study Area

The study was carried out in Kabuga, Gwale Local Government Area, Kano State, Nigeria. The area is located on latitude 12.00012°N and longitude 8.51672°E, having a population of about 362, 059 people.

2.2 Sample collection

Three suya samples were randomly collected from each suya spot location per week for four weeks making a total of 36 samples. The samples were collected from three locations, namely First gate, Gwarzo road and Janbulo area. All samples were collected using sterile hand gloves. The suya samples were collected and wrapped in aluminum foil and transported immediately to the laboratory of the Department of Microbiology, Bayero University, Kano, Kano State, Nigeria, and were analyzed microbiologically. The suya samples were collected between August and September, 2019.

2.3 Isolation and Enumeration of Fungal Count

The suya samples collected were smashed in a sterile laboratory mortar and pestle. Exactly one gram of smashed sample was weighed and aseptically introduced into 9 ml of sterile distilled water, and shaken after which a five-fold dilution was conducted in different test tubes. One millimeter of each of the dilution factor was pipetted and introduced into empty petri dishes. Sterile potato dextrose agar was poured into the sterile petri dishes which were left to solidify. The plates were then incubated at 25 °C for 3 – 5 days. The spores were enumerated on each agar plates for determination of total fungal counts. Distinct spores were observed morphologically and then sub-cultured onto pure freshly prepared potato dextrose agar to obtain pure cultures. The isolates were stored on potato dextrose agar slants at 4 °C for further identification.

2.4 Identification of Fungal Isolates

Pure fungal isolates were identified based on their colonial morphology and microscopic examination. The isolates were also characterized based on their size, shape, pigmentation, texture and septation of hyphae. The microscopic examination was achieved by using the lactophenol cotton blue stain method. This was done by carefully picking a portion of mycelial growth using sterile inoculating needle and spread evenly in a drop of the lacto-phenol cotton blue stain on a clean slide. The clean slide was covered gently with a cover slip to expel air bubbles and observed under microscope using x10 and x40 objective lenses. Identification was done by comparing the fungal morphological features with a fungal atlas [18].

2.5 Proximate composition analysis

The moisture, ash, protein and crude fiber contents were determined as described by AOAC [19].

2.5.1 Moisture content determination

The moisture content was determined following the method of AOAC [19], in which 2 g of the sample was dried in a hot air oven at 100 °C for 24 hr. The loss in weight represents the moisture content (Eq. 1)

$$\% \text{ Moisture} = \frac{W_1 - W_2}{W_1} \times 100 \quad (1)$$

Where, W_1 = Initial weight of the sample; W_2 = Weight of the dried sample

2.5.2 Ash Content Determination

The ash content was assessed by dry ashing method [19]. This was done by measuring 2 g of each of the samples into a porcelain crucible of known weight. The samples were burnt into ash in a muffle furnace at 550 °C for 12 hr. It was then transferred to a desiccator where it was allowed to cool. The weight of the ash was finally determined after weighing. The % ash content is given by Eq. 2:

$$\% \text{ Ash} = \frac{W_1 - W_2}{W_1} \times 100 \quad (2)$$

Where, W_1 = Initial weight of the sample; W_2 = Weight of the sample after ashing.

2.5.3 Determination of Protein Content

Kjeldahl method was employed to determine the crude protein content [19]. Exactly 2 g of the samples was introduced into a digestion flask, followed by 25 ml of concentrated sulphuric acid, 10 g of sodium sulphate and copper sulphate. The flask was put in the digestion block in a fume cupboard and heated until frothing stops producing a light blue coloration that was clear. The

mixture was left to cool after which it was diluted with distilled water until it reached the mark of a 25 ml volumetric flask. The mixture (10 ml) was poured into the distillation apparatus and 10 ml of 40% sodium hydroxide solution was added. Ammonia gas was given off after which boric acid (10 ml) was added to the mixture and observed for color change from green to purple. The percentage protein content was calculated using Eq. 3.

$$\% \text{ Crude protein} = \% N \times 6.25 \quad (3)$$

The nitrogen content of the sample can be calculated from Eq. 4 as

$$\%N = \frac{Tv \times Na \times 0.014 \times V_1}{GV_2} \times 100 \quad (4)$$

Where, Tv = Titre value of the acid (cm³); Na = Concentration or normality of the acid; V₁ = Volume of distilled water used for distilling the digest; V₂ = Volume of aliquot used for distillation.

2.5.4 Crude Fiber Determination

The crude fiber was determined by employing the method of AOAC [19]. Exactly 5 g of the suya sample was defatted with an organic solvent (petroleum ether) using Soxhlet extractor. The defatted sample was boiled in 150 ml of 1.25% H₂SO₄ solution for 30 min under reflux. The process was repeated three times under the same conditions. The sample was transferred to a crucible and dried using acetone.

The sample in the crucible was incubated at 550 °C for 3 h to burn off the carbonaceous content thus leaving ash as crude fiber in the crucible. The percentage crude fiber was determined using Eq. 5

$$\% \text{ Crude Fiber} = \frac{W_1 - W_2}{W} \times 100 \quad (5)$$

Table 3.2: Percentage frequency of fungal isolates from different sources in Kabuga

Fungi	F	G	J	Frequency	% Frequency
<i>Aspergillus</i> spp	4	5	7	16	31.4
<i>Fusarium</i> spp	2	3	5	10	19.6
<i>Penicillium</i> spp	3	4	5	12	23.5
<i>Rhizopus</i> spp	5	5	3	13	25.5
Total	15	17	20	51	100

Key: F = First Gate, G = Gwarzo Road, J = Janbulo Area

Table 3.3: Proximate Mean Values of Suya Samples from Different Sources

S/N	Source	Ash (%)	Moisture (%)	Crude Protein (%)	Fat (%)	Crude Fiber (%)	Carbohydrate (%)
1	F	35.00	40.57	13.08	5.25	3.25	2.45
2	G	24.00	46.32	8.80	5.25	14.58	1.05
3	J	48.00	24.64	17.30	8.75	0.11	1.20

Key: F = First Gate, G = Gwarzo Road, J = Janbulo Area

Where; W=Weight of the sample; W₁=Weight of sample; and crucible before ashing; W₂=Weight of crucible and ash.

2.6 Data Analysis

Data generated from isolation and proximate analysis of suya from different sites in this study was analyzed using descriptive statistics.

3. Results and Discussion

3.1 Results

Table 3.1 shows the total fungal count from different sources in Kabuga, Kano State, Nigeria. Samples from Janbulo area had the highest total fungal count of $2.2 \times 10^4 - 3.8 \times 10^4$ while the least total fungal count ($1.3 \times 10^4 - 2.5 \times 10^4$) were observed for the samples collected from First gate.

Table 3.1: Total Fungal Count from Different Sources in Kabuga

Source	No of Samples	Total fungal count (sfu/g)
First gate	12	$1.3 \times 10^4 - 2.5 \times 10^4$
Gwarzo Road	12	$1.0 \times 10^4 - 3.2 \times 10^4$
Janbulo area	12	$2.2 \times 10^4 - 3.8 \times 10^4$

A total of 51 species of fungi were isolated from all the suya samples collected. These include *Aspergillus* spp, *Fusarium* spp, *Penicillium* spp and *Rhizopus* spp. *Aspergillus* spp was the most prevalent fungus isolated from the samples exhibiting the highest occurrence of 31.4% while *Fusarium* spp was the least isolate with the percentage occurrence of 19.6% as shown in Table 3.2.

The proximate analysis revealed the percentage of ash, moisture, crude protein, fat, crude fiber and carbohydrate content (Table 3.3). The moisture and crude protein were high compared with other proximate/nutritional contents.

3.2 Discussion

The suya samples from Janbulo area had the highest range of fungi in spore forming unit per gram (sfu/g). Its total fungi fungal count ranged from 2.2×10^4 to 3.8×10^4 (Table 3.1). This agrees with the findings of Ike and Ogwuegbu [20] who also reported high range of fungi in a study conducted in Owerri metropolis, Imo State, Nigeria. In addition, these high fungal count as reported in this study are indicative of fungal contamination and this could enhance the spoilage of the meat. The study also revealed the presence of *Penicillium* spp, *Fusarium* spp, *Aspergillus* spp and *Rhizopus* spp in the suya samples from the study area (Table 3.2). This is consistent with the findings of Mohammed *et al.* [21] who isolated *Candida tropicalis*, *Mucor* spp, *Penicillium chrysogenum*, *Aspergillus flavus*, *Fusarium oxysporum*, *Aspergillus niger*, *Penicillium digitatum*, *Rhizopus stolonifer*, *Fusarium solanii*, *Aspergillus fumigatus* from suya vended in some areas in Minna, Niger State, Nigeria and that of Egbebi and Seidu [22] who isolated *Penicillium*, *Mucor*, *Rhizopus* and *Aspergillus* spp from suya sold in Ado-Ekiti and Akure, South Western part of Nigeria. The study by Hassan *et al.* [23] also revealed the presence of fungi such as *Aspergillus niger*, *Mucor*, *Aspergillus parasiticus*, *Penicillium*, *Trichoderma*, *Aspergillus flavus*, *Rhizopus* spp in ready-to-eat barbecue meat. Among the fungal species recovered from the suya samples, *Aspergillus* spp was the predominant isolates with the percentage frequency of 31.4%. *Aspergillus* have ability to produce mycotoxins (aflatoxins) which are detrimental to man, thus their occurrence in suya is very risky. The fungal contamination could be as a result of using contaminated hands by the handlers, contaminated pepper (suya spices), knives, papers/foil paper, utensils, water and serving tables-slabs [21]. This is similar to the reports of Bukar *et al.* [24] who said that the organisms responsible for contamination in suya might have originated from the spices, utensils, handler's hands and air.

According to the results of proximate analysis recorded in Table 3.3, the highest mean values of ash, moisture and crude protein were found to be 2.45, 48.00 and 46.32 in First gate, Janbulo area and Gwarzo road respectively. The fat, crude protein, crude fiber, ash, moisture and carbohydrate contents were highest with the mean values of 17.30, 46.32, 8.75, 2.45, 48.00 and 14.58, respectively in the suya from the three sample sites while the lowest proximate mean values obtained were 8.80, 24.64, 5.25, 1.05, 24.00 and 0.11, respectively (Table 3.3). The high fungal counts as observed in this study could be as a result of high protein content (46.32) which transforms into high nutrient and moisture content [25, 26]. The mean values obtained for moisture content (24 - 48%) was within the value (30.5-43.0%) obtained by Mohammed *et al.* [21]. The high moisture content of

meat was also reported by [26] and was directly related to water activity as it greatly encouraged fungal growth on meats.

4. Conclusion

This study revealed the presence of different species of fungi in the roasted meat (suya) which were *Fusarium*, *Rhizopus*, *Aspergillus* and *Penicillium* spp. The presence of such fungal species in the suya samples collected from various locations in Kabuga, Kano State, Nigeria could be as a result of poor hygiene. The poor hygiene practices exhibited by the vendors could cause severe health risk such as food poisoning which may result to loss of life if a good sanitation is not maintained towards suya production.

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