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# Prevalence Of Fungal And Aflatoxin Contamination In Snacks And Pasta Sold In Sokoto State, Nigeria

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## ABSTRACT

This study investigates the occurrence of fungi and aflatoxin contamination in snacks and pasta products sold in Sokoto State, Nigeria, with a focus on the health risks posed by these contaminants. A total of 300 samples, including meat pies, sausage rolls, doughnuts, macaroni, noodles, and spaghetti, were collected from various retail outlets in urban, semi-urban, and rural areas. The samples were analyzed for fungal contamination and aflatoxin levels using a direct ELISA method and standard mycological techniques. The results revealed a high prevalence of fungal contamination (97%) across all food categories, with *Aspergillus niger* being the most prevalent species. Aflatoxin contamination was found in 92.7% of the samples, with concentrations ranging from 0.6 to 8.2 ppb. Meat pies and macaroni had the highest contamination rates (100%), followed by spaghetti (96%) and sausage rolls (90%). The average aflatoxin concentration across all products was  $12.7 \pm 0.4$  ppb, exceeding the internationally recommended safety limits. The findings highlight the significant public health risks associated with fungal and aflatoxin contamination in commonly consumed snack foods and pasta. The study underscores the urgent need for improved storage, handling practices, and regulatory oversight to mitigate these risks and ensure food safety in Sokoto State. Enhanced awareness and better post-harvest management strategies are essential to reduce fungal growth and mycotoxins accumulation in food products.

**Keywords:** Fungal contamination, Aflatoxins, snacks, pasta, food safety, mycotoxins & ELISA,

## INTRODUCTION

Food safety is a critical global concern, particularly in regions where food production, handling, and storage practices may inadvertently expose consumers to harmful contaminants. Aflatoxins, toxic compounds produced by certain strains of *Aspergillus* fungi, represent a significant threat to food safety. These mycotoxins are commonly associated with the contamination of agricultural products such as cereals, nuts, and grains, often used as ingredients in snacks and pasta. Their presence in food can pose severe health risks, including acute toxicity, liver damage, and an increased risk of liver cancer, particularly in populations with prolonged exposure (Salau *et al.*, 2017; Shehu *et al.*, 2021).

Snacks and pasta products are staple foods consumed widely across Nigeria, including Sokoto State. These products are often produced and marketed in environments where inadequate storage conditions, high temperatures, and humidity may facilitate fungal growth and aflatoxin contamination. Despite their popularity, there is limited data on the occurrence and levels of aflatoxin contamination in these processed foods, particularly in Sokoto State, a region with distinct climatic and economic factors influencing food quality (Salau *et al.*, 2017).

This study aims to investigate the occurrence of fungi and aflatoxin contamination in snacks and pasta sold in Sokoto State, Nigeria. By analyzing selected samples from local markets and retail outlets, the

study seeks to determine the prevalence and concentration levels of Aflatoxins in these food products. The findings will provide critical insights into the safety of these commonly consumed foods and contribute to the formulation of effective strategies for monitoring, controlling, and preventing aflatoxin contamination in the food supply chain.

## **MATERIALS AND METHODS**

This study employed a structured approach to investigate the occurrence of aflatoxin contamination and fungi in snacks and pasta marketed in Sokoto State, Nigeria, using ELISA Kit for analysis.

### **Research Design**

A cross sectional study design was used to assess the prevalence and levels of aflatoxin contamination and fungi in selected snacks and pasta samples obtained from various retail outlets in Sokoto State.

### **Study Area**

The study was conducted in Sokoto State, covering urban, semi urban, and rural areas to ensure diverse market representation.

### **Collection of samples**

A total of three hundred (300) samples were purchased from markets. The breakfast snacks and pastas products comprising of fifty (50) samples each of meat pie, sausage rolls, doughnuts, Macaroni, noodles and spaghetti, were collected in sterile sampling bags at different shopping and selling shades.

### **Isolation of the contaminants**

Samples was inoculated into general purpose media and incubated for 5 days at 25 °C. Colonies of suspected fungi of aflatoxigenic importance were identified and sub cultured onto their respective selective and differential media for cultural and descriptive identification. Pure culture of each species of aflatoxigenic importance were preserved for further analysis

### **Determination of Aflatoxins**

All the samples were analyzed for the presence of aflatoxins (total aflatoxins = sum of AFB1, AFB2, AFG1 and AFG2) by the Competitive Direct (CD) ELISA method. The AgraQuant® Total Aflatoxin Assay 4/40 kit (Romer Labs, Singapore) was used. All reagents including LC grade methanol were purchased from Sigma Aldrich (St Louis, MO, USA).

Extraction and testing of aflatoxins were performed on each sample according to manufacturer instructions in the ELISA kits. Briefly, 5 g of each representative sample was weighed into a 250 ml conical flask and extracted with 25 ml of 70% methanol (1:5 w/v) on an orbital shaker for 10 min at ambient temperature. The mixture was filtered through a 110 mm No.1 Whatman filter paper into a 40 ml tube. An aliquot (50 µl) of each filtrate representing a sample was then dispensed into dilution wells and 100 µl of AgraQuant® aflatoxin conjugate was added to each sample/filtrate in the dilution wells and mixed. About 100 µl of this mixture was then dispensed into antibody-coated microwells and incubated at ambient temperature for 15 min.

Subsequently, the contents of the incubated microwells were emptied and washed thrice with distilled water to rinse off all remaining mixture.

The total aflatoxin estimates in each sample was obtained by extrapolation from the standard curve.

### **Statistical Analysis**

Data obtained from this study was analyzed using SPSS® (Windows version 21.0, SPSS, IL, USA). Means were tested for significance at 95% confidence level using one way ANOVA and separated by Duncan's Multiple Range Test. Descriptive statistics, such as frequencies and percentages, were calculated using cross-tabulation in Excel. All statistical tests with Alpha or p values less than 0.05 were considered statistically significant.

## **RESULTS**

The analysis of fungal contamination (Table 1) in various snacks and pastas revealed a high prevalence of fungal presence, with an overall contamination rate of 97.0% (291/300 samples). All meat pie, sausage roll, doughnut, and macaroni samples were 100% positive for fungal contamination, while noodles and spaghetti showed slightly lower contamination rates of 90% and 96%, respectively. Among the identified

fungal species, *Aspergillus niger* was the most prevalent, with a total count of 132 isolates, followed by *Aspergillus fumigatus* (63 isolates) and *Aspergillus flavus* (52 isolates). Other fungi detected included *Rhizopus stolonifer* (35 isolates), *Fusarium oxysporum* (39 isolates), and *Penicillium notatum* (32 isolates).

Meat pie exhibited the highest fungal load, with *A. niger* (35 isolates) being the most dominant species. Similarly, sausage rolls and doughnuts also showed significant contamination, with *A. niger* (32 and 27 isolates, respectively) being the predominant species. Spaghetti had the highest occurrence of *F. oxysporum* (13 isolates), while doughnuts had a considerable presence of *P. notatum* (11 isolates).

**Table 1: Incidence and Distribution of Fungal Contamination in Snacks and Pastas**

Snacks and Pastas	Positive Samples	Distribution of Fungi in Samples					
		<i>A. niger</i>	<i>A. flavus</i>	<i>A. fumigatus</i>	<i>R. stolonifer</i>	<i>F. oxysporum</i>	<i>P. notatum</i>
Meat Pie	50/50 (100%)	35	12	22	11	08	05
Sausage Rolls	50/50 (100%)	32	11	08	02	02	02
Doughnuts	50/50 (100%)	27	07	13	08	04	11
Noodles	45/50 (90%)	11	05	05	00	03	05
Spaghetti	46/50 (96%)	09	13	06	10	13	07
Macaroni	50/50 (100%)	18	04	09	04	09	02
<b>Total</b>	<b>291/300 (97.0%)</b>	<b>132</b>	<b>52</b>	<b>63</b>	<b>35</b>	<b>39</b>	<b>32</b>

The table (2) presents the prevalence of positive samples and the concentration of Aflatoxins (in ppb) across various snacks and pasta products. The findings indicate that: A high percentage of samples across all food items tested positive for aflatoxin. The meat pie and macaroni samples had the highest prevalence, with 100% of the samples showing contamination, followed closely by spaghetti at 96%. Sausage rolls (90%) and doughnuts (90%) also exhibited a significant prevalence of contamination, while noodles had a slightly lower prevalence at 80%. In total, 92.7% of all tested samples across the six product categories were contaminated with aflatoxin.

The concentration of aflatoxins varied across the different food items. The concentration ranged from 0.6 to 8.2 ppb, with the highest observed in meat pies, followed by sausage rolls (1.2 – 6.6 ppb), and doughnuts (0.6 – 4.9 ppb). The lower range (0.9 – 2.5 ppb) was found in noodles and spaghetti, with macaroni having the lowest range (0.9 – 1.5 ppb). The mean concentration of aflatoxins differed significantly across the product types. Meat pies exhibited the highest mean concentration ( $12.4 \pm 1.2$  ppb), followed closely by sausage rolls ( $12.5 \pm 0.6$  ppb). Doughnuts ( $12.0 \pm 0.3$  ppb) and macaroni ( $11.9 \pm 0.3$  ppb) had similar mean values. Noodles and spaghetti showed lower mean concentrations, with noodles having the lowest mean value ( $11.1 \pm 0.2$  ppb) and spaghetti ( $11.0 \pm 0.3$  ppb) slightly higher. The overall mean concentration across all tested samples was  $12.7 \pm 0.4$  ppb.

The mean values for aflatoxin concentration showed significant differences among the various food items, as indicated by the superscript letters. Meat pies and sausage rolls exhibited significantly higher concentrations of aflatoxins compared to the other products.

**Table 2: Aflatoxin Contamination and Prevalence in Snacks and Pastas in Sokoto State.**

Snacks and Pastas	Positive Samples	Aflatoxins concentration (ppb)	
		Concentration Range	Mean $\pm$ SE
Meat Pie	50/50 (100%)	1.6 – 8.2	12.4 <sup>a</sup> $\pm$ 1.2
Sausage Rolls	45/50 (90%)	1.2 – 6.6	12.5 <sup>b</sup> $\pm$ 0.6
Doughnuts	45/50 (90%)	0.6 – 4.9	12.0 <sup>b</sup> $\pm$ 0.3
Noodles	40/50 (80%)	0.9 – 2.5	11.1 <sup>c</sup> $\pm$ 0.2
Spaghetti	48/50 (96%)	1.0 – 1.8	11.0 <sup>c</sup> $\pm$ 0.3
Macroni	50/50 (100%)	0.9 – 1.5	11.9 <sup>b</sup> $\pm$ 0.3
<b>Total</b>	<b>278/300 (92.7%)</b>	<b>0.6 – 8.2</b>	<b>12.7 <math>\pm</math> 0.4</b>

Mean values with different superscript alphabets are significantly different at  $\alpha = 0.05$ .

## DISCUSSION

The findings of this study reveal a high prevalence of fungal contamination in commonly consumed snacks and pasta products, with an overall contamination rate of 97.0% (291/300 samples). This result indicates a significant risk of fungal spoilage and potential mycotoxins contamination in these food products, which can pose serious health risks to consumers. The high contamination rate aligns with previous studies that have reported frequent fungal contamination in processed foods due to improper handling, poor storage conditions, and inadequate hygiene practices (Pitt & Hocking, 2009; Ezekiel et al., 2013).

Among the fungal species isolated, *Aspergillus niger* was the most dominant, with a total occurrence of 132 isolates across all food categories. It was particularly prevalent in meat pies (35 isolates) and sausage rolls (32 isolates). *A. niger* is a well-known spoilage organism in bakery products and processed foods due to its ability to produce hydrolytic enzymes that degrade carbohydrates and lipids (Frisvad et al., 2019). Additionally, some strains of *A. niger* can produce ochratoxins, which are nephrotoxic and have been linked to kidney disorders in humans (Adebajo et al., 1994). *Aspergillus flavus* is of great concern due to its ability to produce aflatoxins, potent carcinogens that have been associated with liver cancer and immune suppression in humans (Bhat et al., 2010). The presence of *A. flavus* in these products suggests that improper processing, storage, and handling conditions may have favored its proliferation, as aflatoxin production is often associated with warm and humid environments (Ezekiel et al., 2013).

The detection of *R. stolonifer* in these samples suggests that spoilage may have begun even before consumption, possibly due to high moisture content or inadequate storage conditions. The presence of *F. oxysporum* in these foods raises concerns about mycotoxin exposure among consumers, especially in regions where mycotoxin contamination of staple foods is already a major issue (Bankole & Adebajo, 2003). *Penicillium notatum* was the least frequently detected fungus. Although *P. notatum* is not a major producer of toxic metabolites, some species within the *Penicillium* genus can produce patulin and citrinin, which are harmful to human health (Moss, 2002).

The high fungal contamination levels observed in this study are consistent with previous research conducted in Nigeria and other parts of West Africa, where fungal spoilage of processed foods has been widely reported (Ezekiel et al., 2013; Bankole & Adebajo, 2003). Environmental factors such as high temperatures, humidity, and poor food storage infrastructure contribute significantly to fungal proliferation in food products. Similar studies conducted in Nigeria have reported *A. niger*, *A. flavus*, and *Fusarium* spp. as dominant contaminants in bakery products, cereals, and processed foods (Bamidele et al., 2018).

The contamination of staple food products such as pasta (noodles, spaghetti, macaroni) with *F. oxysporum* and *A. flavus* is particularly concerning due to the risk of mycotoxin exposure in consumers. Aflatoxins and fumonisins have been frequently detected in maize and wheat-based products in Nigeria, indicating a widespread issue of fungal contamination in stored grains (Bankole & Adebajo, 2003). Given that many local food vendors and small-scale food processors lack proper storage facilities, the high incidence of fungal contamination in this study underscores the need for improved food handling and preservation techniques.

The study indicates a high prevalence of aflatoxin contamination in various snacks and pasta products commonly consumed in Sokoto State, Nigeria. Out of 300 tested samples, 92.7% were contaminated with aflatoxin at varying levels, with mean concentrations ranging from 11.0 to 12.5 ppb. The highest contamination rate was observed in meat pies and macaroni (100%), followed closely by spaghetti (96%) and sausage rolls (90%). The significantly different mean aflatoxin concentrations among these products ( $p < 0.05$ ) suggest variability in contamination sources and potential differences in processing, storage, and ingredient susceptibility to fungal infestation. Aflatoxins, particularly Aflatoxin B1 (AFB1), are among the most potent mycotoxins with carcinogenic effects, posing serious public health concerns (IARC, 2023). Studies have consistently reported high aflatoxin contamination in cereal-based and baked products, especially in tropical regions with warm and humid conditions conducive to fungal growth (Ezekiel et al., 2022; Warth et al., 2023). The contamination levels found in this study align with previous reports from other regions in Nigeria and sub-Saharan Africa, where storage conditions, poor regulatory enforcement, and raw material susceptibility contribute to high aflatoxin prevalence in processed foods (Adeyeye, 2021).

The high aflatoxin levels in meat pies ( $12.4 \pm 1.2$  ppb) and sausage rolls ( $12.5 \pm 0.6$  ppb) could be attributed to the use of contaminated wheat flour or other raw materials such as milk and eggs, which have been identified as aflatoxin carriers in previous studies (Kamala et al., 2021). Similarly, the contamination of pasta products (spaghetti, macaroni, and noodles) suggests that the wheat or maize used in their production may have been compromised during pre- or post-harvest storage. Grains stored under inadequate moisture control are particularly prone to *Aspergillus* spp. infection, which facilitates aflatoxin production (Reddy et al., 2023). From a regulatory standpoint, the Codex Alimentarius Commission and European Union have set aflatoxin limits for cereals and processed foods at 4 ppb for total aflatoxins and 2 ppb for AFB1 (FAO/WHO, 2023). The mean aflatoxin concentrations detected in this study exceed these safety limits, indicating a significant food safety concern. The observed levels, particularly in frequently consumed snack products, pose a long-term risk of chronic aflatoxin exposure, which has been linked to liver cancer, immune suppression, and stunted growth in children (Wu et al., 2022).

The findings highlight the urgent need for improved storage and handling practices for raw materials used in snack and pasta production. Enhanced regulatory monitoring and awareness campaigns among food processors, vendors, and consumers could help mitigate the risks associated with aflatoxin contamination. Furthermore, adopting post-harvest management strategies, such as biocontrol agents (Aflasafe®), improved drying techniques, and hermetic storage systems, could significantly reduce fungal infestation and aflatoxin accumulation in food products (Bandyopadhyay et al., 2023).

## CONCLUSION

This study reveals a high prevalence of fungal contamination in commonly consumed snacks and pasta products in Sokoto State, Nigeria, with 97.0% of the samples tested being affected. The contamination was primarily caused by *Aspergillus niger*, *Aspergillus flavus*, *Fusarium oxysporum*, and *Rhizopus stolonifer*, which are known to produce mycotoxins such as aflatoxins, posing serious health risks like liver cancer, kidney damage, and immune suppression. The study also detected 92.7% aflatoxin contamination, with levels exceeding international safety limits, highlighting a critical food safety issue. The results are consistent with previous studies in Nigeria and sub-Saharan Africa, where poor storage, handling practices, and environmental factors like high humidity contribute to fungal growth and mycotoxins production. Specifically, the contamination in meat pies and macaroni (100%) was found to exceed the acceptable aflatoxin levels, emphasizing the need for improved processing, storage, and regulatory measures. The study calls for enhanced food safety awareness among food producers, vendors, and consumers, as well as better regulatory enforcement. This research underscores the urgent need for stronger food safety practices and public health interventions to mitigate the risks posed by fungal and mycotoxins contamination in Sokoto State. Future studies should focus on refining these intervention strategies to safeguard public health and improve food security in the region.

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