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Aflatoxin Assay of Home-Packed Meals of Selected Pupils in Benin City

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Abstract: **This study evaluated the presence and concentration of aflatoxins in home-packed meals consumed by primary school pupils in Benin City, Nigeria. Eighteen (18) food samples were collected from five (5) private schools and analyzed using standard microbiological and chromatographic methods. Fungal counts ranged from 2.0 × 10^3 CFU/ml to 4.5 × 10^4 CFU/ml, with predominant isolates including *Aspergillus fumigatus, A. niger, A. aculeatus,* and *Penicillium citrinum*. Aflatoxin types B_1, B_2, G_1, and G_2 were detected at concentrations between 0.71 and 1.36 µg/kg, and total aflatoxin levels ranged from 3.20 to 4.84 µg/kg. Although these levels were within Nigerian safety limits, the results indicate potential chronic exposure risks for schoolchildren. Regular aflatoxin surveillance and improved hygiene in food preparation are recommended to ensure the safety of home-packed meals.**

Keywords: **Aflatoxin, Aspergillus, Benin City, food safety, mycotoxin contamination, pupils.**

1. INTRODUCTION

Food safety among schoolchildren remains a significant public health concern, particularly in developing countries where food handling and storage practices are often inadequate (WHO, 2016; Maduka *et al*., 2022). In Nigeria, most pupils depend on home-packed meals as their primary mid-day food source, as this option is considered safer and more affordable than ready-to-eat foods sold by street vendors (Olusanya, 2010; Ugochukwu *et al*., 2014). However, improper storage, reheating, and handling of home-packed meals may promote microbial contamination and mycotoxin development (Akindele & Ibrahim, 2016; Edmonds-Wilson *et al*., 2015).

Aflatoxins; potent secondary metabolites produced mainly by *Aspergillus flavus* and *A. parasiticus* pose serious health risks when present in food, including hepatotoxicity, carcinogenicity, and growth retardation in children (Fapohunda *et al*., 2008; Ezekiel *et al*., 2019). Previous studies have reported the presence of aflatoxin-producing fungi such as *Aspergillus niger* and A. fumigatus in food items consumed by school-aged children (Maduka *et al*., 2022). In Nigeria, contamination of staple foods such as maize, groundnut, and melon seeds by aflatoxins has been widely documented (Makun *et al*., 2013; Obani *et al*., 2019). Despite these findings, limited data exist on aflatoxin contamination in home-packed meals consumed by school pupils, especially in southern Nigeria.

This study, therefore, aimed to determine the levels of aflatoxins (B_1, B_2, G_1, and G_2) in home-packed meals of selected pupils in Benin City and to identify the associated mycotoxigenic fungi. The findings are expected to provide baseline information on the safety of home-prepared meals and contribute to strategies for mitigating aflatoxin exposure among schoolchildren.

2. FOODBORNE FUNGI AND AFLATOXIN CONTAMINATION

Fungi are ubiquitous microorganisms that play both beneficial and detrimental roles in food systems. While some species are valuable in food production - such as *Saccharomyces cerevisiae* used in bread and beverage fermentation. These cause significant food spoilage and contamination (Odeyemi *et al*., 2020). It is estimated that fungal spoilage accounts for 5 - 10% of global food losses annually (Kwon *et al*., 2008). In poorly preserved foods, filamentous fungi like *Aspergillus*, *Fusarium*,

Penicillium, and *Rhizopus* species can proliferate and synthesize harmful secondary metabolites known as mycotoxins (Tomsikova, 2002).

Among these mycotoxins, aflatoxins are the most potent and widespread. They are produced predominantly by *Aspergillus flavus* and *A. parasiticus* under conditions of high humidity and temperature, such as those typical in southern Nigeria (Fapohunda *et al*., 2008; Ezekiel *et al*., 2019). Aflatoxins (B_1, B_2, G_1, and G_2) are known for their hepatotoxic, mutagenic, and carcinogenic properties and are classified as Group 1 carcinogens by the International Agency for Research on Cancer (IARC) (WHO, 2016). Children are particularly susceptible due to their developing immune systems and higher food intake relative to body weight (Coppock *et al*., 2018).

Foodborne fungal contamination and aflatoxin accumulation can occur at various points - from preparation to storage of home-packed meals. In households where cooked foods are left at ambient temperature for long periods before being packed for school, fungal spores from the environment can easily colonize and produce toxins. Species such as *A. niger*, *A. fumigatus*, and *Penicillium citrinum* have been commonly isolated from ready-to-eat and home-prepared foods in Nigerian settings (Makun *et al*., 2013).

Consumption of aflatoxin-contaminated foods can lead to aflatoxicosis, characterized by liver dysfunction, immune suppression, stunted growth, and in severe cases, hepatocellular carcinoma (Benkerroum, 2020). Although the Nigerian regulatory limit for total aflatoxins in food is 10 µg/kg, studies have reported contamination levels approaching this threshold in common school meals such as rice, beans, and maize-based dishes (Ezekiel *et al*., 2019). The detection of aflatoxins in home-packed meals highlights the need for strict hygiene during preparation, proper food storage, and public education on mycotoxin risks.

3. RISK OF FOODBORNE ILLNESSES TO PRIMARY SCHOOL CHILDREN

Nutrition during childhood is fundamental to the physical, cognitive, and psychosocial development of pupils aged 6–12 years. In developing countries such as Nigeria, schoolchildren typically depend on home-packed meals, foods purchased from roadside vendors, or, less commonly, school feeding programs for their daily nutrition (Maduka *et al*., 2022). These meals are usually consumed at ambient temperature during lunch hours without reheating, creating a favorable environment for microbial proliferation.

According to the Centers for Disease Control and Prevention (CDC, 2022), children under the age of five are three times more likely to be hospitalized due to Salmonella infection, while one in seven cases of E. coli O157:H7 infection in children can progress to hemolytic uremic syndrome (HUS), leading to kidney failure. Foodborne illnesses in children often manifest as diarrhea, vomiting, abdominal cramps, fever, and dehydration, which can become life-threatening if not promptly managed (WHO, 2016).

Reports indicate that children below 15 years constitute nearly half of global food poisoning cases, reflecting their greater vulnerability due to underdeveloped immune systems and poor hygiene practices (WHO, 2016). Inappropriate food storage, cross-contamination, inadequate cooking temperatures, and poor hygiene among food handlers are major contributors to these outbreaks.

A study by Maduka *et al*. (2022) in Benin City revealed that home-packed meals of schoolchildren contained diverse microbial contaminants, including *Escherichia coli* (30%), S*taphylococcus aureus* (25%), *Bacillus cereus* (15%), and *Aspergillus flavus* (8%), among these. The presence of these pathogens, particularly *A. flavus*, which can produce aflatoxins, underscores the potential risk of foodborne infections and mycotoxin exposure even in meals prepared at home for school consumption.

4. MATERIALS AND METHODS

Materials

Sterile universal containers, swab sticks, autoclave, pipettes, Petri dishes, test tubes, conical flasks, centrifuge, incubator, weighing balance, cotton wool, microscope, UV Visible spectrophotometer, filter paper, amber bottles, and rotary shaker were used for sample collection, microbiological analysis, and aflatoxin assay.

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The Study Area and Samples Collection

A total of fifteen (15) home-packed food samples were collected from five (5) selected private primary schools located along the Ogumnwenyin–Ugbor Road axis in Benin City, Edo State, Nigeria (Fig, 1). Each school provided three (3) different food samples representing the common meal types consumed by pupils - fried rice, jollof rice, white rice with stew, noodles, pasta, and beans. Three (3) additional samples obtained from street food vendors along the same axis served as control. All samples were collected aseptically between 2nd and 12th May 2023, placed in sterile containers, and transported on ice to the Microbiology Laboratory, Wellspring University, and the Mycotoxin Laboratory, Nigerian Institute for Oil Palm Research (NIFOR) for analysis within four hours of collection.

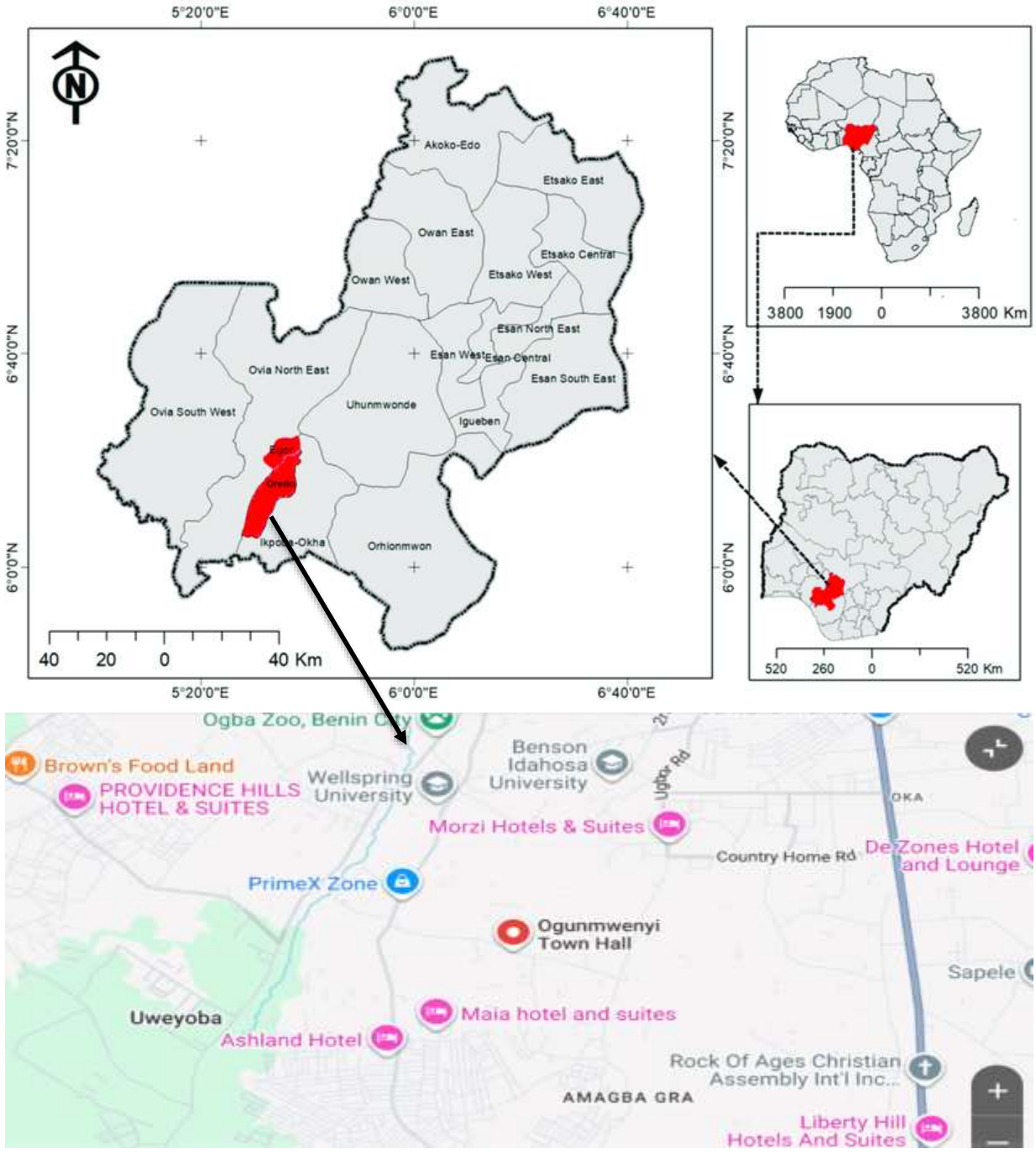

Figure 1: Map of the study area

Isolation of fungi

Food samples were homogenized aseptically, and serial dilutions were prepared using sterile 0.9% normal saline. One millilitre (1 mL) aliquots of the 10^{-3} dilution were plated in duplicate Potato Dextrose Agar and plates were incubated at room temperature (25 ± 2°C) for 7days.

The total number of fungal colonies were counted after the incubation period of 7 days using the hand tally counter. The result obtained for the TFC was calculated using the formula below.

$$\text{CFU/g} = \text{no. of colonies} \times \frac{1}{dilution\ factor} \times \frac{1}{volume\ plated}$$

Result was expressed as colony forming units per gram or CFU/g.

Morphological characteristics such as colony colour, texture, and growth pattern were recorded.

Microscopic identification was performed using lactophenol cotton blue staining as described by Gaddeyya *et al*. (2012). The prepared slides were examined under ×10 and ×40 objective lenses for structural identification of fungal spores and hyphae.

Molecular identification

Fungal genomic DNA extraction

Extraction was done using a ZR fungal DNA mini prep extraction kit supplied by Inqaba South Africa. A heavy growth of the pure culture of the fungal isolates was suspended in 200 microliters of isotonic buffer into a ZR Bashing Bead Lysis tubes, 750 microliters of lysis solution was added to the tube. The tubes were secured in a bead beater fitted with a 2 ml tube holder assembly and processed at maximum speed for 5 minutes. The ZR bashing bead lysis tube were centrifuged at 10,000x*g* for 1 minute.

Four hundred (400) microliters of supernatant were transferred to a Zymo-Spin IV spin Filter (orange top) in a collection tube and centrifuged at 7000x*g* for 1minute. One thousand two hundred (1200) microliters of fungal DNA binding buffer were added to the filtrate in the collection tubes bringing the final volume to 1600 microliters, 800 microliters were then transferred to a Zymo-Spin IIC column in a collection tube and centrifuged at 10,000x*g* for 1 minute, the flow through was discarded from the collection tube. The remaining volume was transferred to the same Zymo-spin and spun. Two hundred (200) microliters of the DNA Pre-Was buffer was added to the Zymo-spin IIC in a new collection tube and spun at 10,000x*g* for 1 minute followed by the addition of 500 microliters of fungal DNA Wash Buffer and centrifuged at 10,000x*g* for 1 minute.

The Zymo-spin IIC column was transferred to a clean 1.5 microliters centrifuge tube, 100 microliters of DNA elution buffer were added to the column matrix and centrifuged at 10,000x*g* for 30 seconds to elute the DNA. The ultra-pure DNA was then stored at -20 degree for oThere downstream reaction.

DNA quantification

The extracted genomic DNA was quantified using the Nanodrop 1000 spectrophotometer. The software of the equipment was lunched by double clicking on the Nanodrop icon. The equipment was initialized with 2 µl of sterile distilled water and blanked using normal saline. Two microlitre of the extracted DNA was loaded onto the lower pedestal, the upper pedestal was brought down to contact the extracted DNA on the lower pedestal. The DNA concentration was measured by clicking on the "measure" button.

Agarose gel electrophoresis

Agarose gel electrophoresis is used to determine wheThere or not the fragment of DNA that is amplified by PCR is the expected size. The presence of a fragment of the correct size in the reaction tube strongly suggests a successful PCR. In the case of a successful amplification, bacterial 16SrRNA bands will be at 1500bp, while fungal ITS bands will be at 500bp. 1% agarose gel was prepared by dissolving 1 gm of agarose powder in 1 mil of distilled water. 1X trace borate buffer (TBE) was added. The mixture was heated to boiling and 5 µl of Safeview tracking dye was added for visualization. The boiled mixture was poured in a casting tray/mould with a comb placed in the tray and allowed to cool down. The comb was removed and the wells formed were loaded with the amplified DNA samples.

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Fungal Internal Transcribed Spacer (ITS) amplification

The ITS region of the rRNA genes of the isolates were amplified using the ITS1F: 5'-CTTGGTCATTTAGAGGAAGTAA-3' and ITS4: 5'- TCCTCCGCTTATTGATATGC-3,

primers on an ABI 9700 Applied Biosystems Theremal cycler at a final volume of 50 microlitres for 35 cycles. The PCR mix included: the X2 Dream Taq Master mix supplied by Inqaba, South Africa (Taq polymerase, DNTPs, MgCl), the primers at a concentration of 0.4M and the extracted DNA as template. The PCR conditions were as follows: Initial denaturation, 95ºC for 5 minutes; denaturation, 95ºC for 30 seconds; annealing, 53ºC for 30 seconds; extension, 72ºC for 30 seconds for 35 cycles and final extension, 72ºC for 5 minutes. The product was resolved on a 1% agarose gel at 120V for 15 minutes and visualized on a UV transilluminator.

Sequencing of 16S rRNA and ITS

Sequencing was done using the BigDye Terminator kit on a 3510 ABI sequencer by Inqaba Biotechnological, Pretoria South Africa. The sequencing was done at a final volume of 10 µl, the components included 0.25 µl BigDye® terminator v1.1/v3.1, 2.25 µl of 5 x BigDye sequencing buffer, 10 µM PCR primer, and 2-10 ng PCR template per 100bp. The sequencing conditions were as follows 32 cycles of 96°C for 10s, 55°C for 5s and 60°C for 4min.

Phylogenetic analysis

Obtained sequences were edited using the bioinformatics algorithm Trace edit, similar sequences were downloaded from the National Center for Biotechnology Information (NCBI) data base using BLASTN. These sequences were aligned using ClustalX. The evolutionary history was inferred using the Neighbor-Joining method in MEGA 6.0 (Saitou and Nei, 1987). The bootstrap consensus tree inferred from 500 replicates (Felsenstein, 1985) is taken to represent the evolutionary history of the taxa analysed. The evolutionary distances were computed using the Jukes-Cantor method (Jukes and Cantor, 1969).

Aflatoxin determination in the food and fungal samples

The aflatoxins in the foods and fungal samples were determined according to the methods described by JAOAC (1988).

Extraction of the food samples

Exactly 15.0 g of the blended food sample was weighed into a sterile beaker, 3 g of sodium chloride and 75 mL of a mixture of methanol and water (7:3, v/v) was added to the food in the beaker. The mixture was homogenized for about 2 mins and centrifuged for about 10 mins. Fifteen (15) milliliters of the supernatant solution of the centrifuged mixture were transferred into an amber bottle and filtered through a filter paper. The filtrate was collected as the test solution.

Preparation of the fermenting medium

The isolated fungi were screened for their ability to produce aflatoxin in a growth medium. The screening was carried out in 250 ml Erlenmeyer flasks containing 100 ml of the fermenting medium (glucose, 10.0g; K_2HPO_4, 2.0g; $NaNO_3$, 0.5g; $MgSO_4.7H_2O$, 0.5g; pH 7.0). The experimental flasks were incubated on a rotary shaker.

Preparation of fungal test samples

The test samples were prepared by transferring the mycelia culture in the fermenting medium mentioned above into a centrifuged bottle. The centrifuged bottles were then centrifuged at 4000rpm for 10 mins. Thereafter, with the aid of a micropipette 5mls of the supernatant was transferred into a conical flask. This served as the test sample. This procedure was also repeated for the fermenting medium that was not inoculated with any of the fungal samples. This served as the control experiment.

Calibration of the Ultraviolet-Visible (UV-Vis) spectrophotometer

This was done using the absorbance of difference concentrations of $K_2Cr_20_7$ in H_2S0_4 (0.25, 0.125 and 0.0625mM) at absorbance near 350 nm against 0.009M H_2S0_4 as solvent blank. The molar absorptivity at each concentration was determined using the equation below:

$$\varepsilon = \frac{(A \times 1000)}{Concentration\ (mM)}$$

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Determination of the molar absorptivity (ε) of the different aflatoxin standards

One (1) milligram of the respective aflatoxin standards (B_1, B_2, G_1 and G_2) was accurately weighed into 100 ml volumetric flasks, then dissolved and diluted with methanol. The maximum absorbance (A) was measured using a UV-Vis spectrophotometer at a wavelength close to 350 nm.

$$\varepsilon = \frac{A_{350} \times M_W \times 1000}{\mu g \text{ aflatoxin/kg}}$$

Determination of Individual aflatoxin concentration in the food and fungal test samples

This was done by recording the UV spectrum of the test samples from 200 - 500 nm against the methanol solvent for the food samples; and the centrifuged fermenting medium for the fungal samples. The concentrations of individual aflatoxin in the food and fungal samples were determined by measuring the maximum absorbance (A) close to 350 nm using the equation below:

$$\text{Concentration of aflatoxin } (\mu g/kg) = \frac{A_{350} \text{ x } M_W \text{ x } 1000}{\varepsilon}$$

Where A_{350} = the absorbance of the aflatoxin at a wavelength of maximum absorption close to 350 nm,

M_w = the molecular weight of the aflatoxin

ε = the molar absorptivity of the aflatoxin in solution

5. RESULTS AND DISCUSSION

This study evaluated the fungal contamination of food samples from the home-packed meals of 15 pupils in five private primary schools and street-vended foods in Benin City. The total fungal count (TFC) ranged from 2.0×10^3 to 4.5×10^4 CFU/g in home-packed meals and from 2.8×10^3 to 3.5×10^4 CFU/g in street foods as seen in Table 1. Fungal growth was observed in nearly all samples after a 7-day incubation period at room temperature (25 ± 2°C).

Table 2 shows that the fungi isolated from these samples included *Aspergillus flavus*, *A. fumigatus*, *A. niger*, *Penicillium chrysogenum*, and *Saccharomyces cerevisiae*, as classified based on morphological features on Potato Dextrose Agar and microscopy. The prevalence of *Aspergillus spp*. aligns with earlier findings by Ekpakpale *et al*. (2021) and Perrone *et al*. (2007), indicating contamination during handling and storage, especially considering the moist and carbohydrate-rich nature of the foods.

All samples contained detectable levels of aflatoxins B_1, B_2, G_1, and G_2 with concentrations between 0.71 and 1.36 µg/kg - totaling 3.20 to 4.84 µg/kg as summarized in Figure 1. Although these levels are below the Nigerian regulatory limit of 10 µg/kg, they exceed the European Union's stricter threshold of 4 µg/kg (Nigerian FDA, 2018; Bande *et al*., 2022). The highest aflatoxin production was by A. aculeatus, while P. citrinum produced none, aligning with previous research suggesting multiple Aspergillus species are capable of producing mycotoxins under conducive conditions (Ekpakpale *et al*., 2021; Makun *et al*., 2013).

The relatively low aflatoxin levels may be due to short storage periods and heat application during cooking, which may be partly responsible for reducing toxin concentrations. Nonetheless, the presence of mycotoxins, even at low levels, poses potential health risks for children, emphasizing the need for improved hygiene, storage practices, and continuous monitoring to prevent long-term health implications (Abdelaziz, 2009; Makun *et al*., 2023)..

Table 1. Total fungal counts of home-packed schoolchildren's meals and street foods

Sample	CFU/g		
PS1	7.5×10^3	1.5×10^4	4.5×10^4
PS2	2.0×10^3	3.0×10^3	5.0×10^3
PS3	4.5×10^3	2.6×10^3	4.8×10^3
PS4	5.0×10^3	8.0×10^3	2.0×10^3
PS5	2.1×10^3	8.2×10^3	7.2×10^3
Street foods	3.5×10^4	2.8×10^3	3.2×10^4

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Table 2: Colonial characteristics and morphology of fungi isolated from home-packed meals of school children and street foods

Samples (school)		Colonial characteristics	Morphology and cellular structure	Suspected organism
PS1	a	Colonies are fluffy black with white to cream edges and mostly white when viewed from the reverse of the Petri dish	Spherical sporangia with brush-like sporangiophores	*Aspergillus flavus*
	b	Fluffy/woolly velvety white to cream/light yellow colonies	Conidiophores are present with brush-like heads containing abundant dark-coloured and spherical conidia	*Aspergillus flavus*
	c	Colonies are white and moist and smooth/slimy	There is the presence of blastoconidia. They are globose, and ellipsoid to elongate in shape	*Saccharomyces cerevisiae*
PS2	a	Cottony/fluffy colonies which are initially white to yellow and then turning black with time. The reverse is white to yellow.	Smooth and coloured conidiophores and conidia. The conidial heads appear radial and they split into columns (biseriate). There is the presence of phialides on the conidiophores,	*Aspergillus niger*
	b	Fast-growing Greenish-grey to Greyish-white fluffy/velvety colonies of moderate size with powdery surface	Conidiophores are broom/brush-like with small and spherical conidia	*Aspergillus fumigatus*
	c	Cottony/fluffy colonies which are initially white to yellow and then turning black with time. The reverse is white to yellow.	Smooth and coloured conidiophores and conidia. The conidial heads appear radial and they split into columns (biseriate). There is the presence of phialides on the conidiophores	*Aspergillus niger*
	c	Fast-growing Greenish-grey to Greyish-white fluffy/velvety colonies of moderate size with powdery surface	Conidiophores are broom/brush-like with small and spherical conidia	*Aspergillus fumigatus*
PS3	a	Fast-growing Greenish-grey to Greyish-white fluffy/velvety colonies of moderate size with powdery surface	Conidiophores are broom/brush-like with small and spherical conidia	*Aspergillus fumigatus*
	a	Colonies are fluffy black with white to cream edges and mostly white when viewed from the reverse of the Petri dish	Spherical sporangia with brush-like sporangiophores	*Aspergillus flavus*
	b	Fast-growing Greenish-grey to Greyish-white fluffy/velvety colonies of moderate size with powdery surface	Conidiophores are broom/brush-like with small and spherical conidia	*Aspergillus fumigatus*
	c	Colonies are fluffy/cottony green to yellow-green in appearance with a distinct radial/fan-like growth pattern	Septate hyphae. Relatively long, Smooth-walled conidiophores. The conidia are spherical/elliptical and produced in well-defined chains	*Penicillium chrysogenum*
	c	Fluffy/woolly velvety white to cream/light yellow colonies	Conidiophores are present with brush-like heads containing abundant dark-coloured and spherical conidia	*Aspergillus flavus*
PS4	a	Fast-growing Greenish-grey to Greyish-white fluffy/velvety colonies of moderate size with powdery surface	Conidiophores are broom/brush-like with small and spherical conidia	*Aspergillus fumigatus*
	b	Colonies are fluffy/cottony green to yellow-green in appearance with a distinct radial/fan-like growth pattern	Septate hyphae. Relatively long, Smooth-walled conidiophores. The conidia are spherical/elliptical and produced in well-defined chains	*Penicillium chrysogenum*

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	c	Colonies are fluffy black with white to cream edges and mostly white when viewed from the reverse of the Petri dish	Spherical sporangia with brush-like sporangiophores	*Aspergillus flavus*
PS5	a	Cottony/fluffy colonies which are initially white to yellow and then turning black with time. The reverse is white to yellow.	Smooth and coloured conidiophores and conidia. The conidial heads appear radial and they split into columns (biseriate). There is the presence of phialides on the conidiophores	*Aspergillus niger*
	b	Fast-growing Greenish-grey to Greyish-white fluffy/velvety colonies of moderate size with powdery surface	Conidiophores are broom/brush-like with small and spherical conidia	*Aspergillus fumigatus*
	c	Colonies are fluffy black with white to cream edges and mostly white when viewed from the reverse of the Petri dish	Spherical sporangia with brush-like sporangiophores	Aspergillus flavus
	c	Cottony/fluffy colonies which are initially white to yellow and then turning black with time. The reverse is white to yellow.	Smooth and coloured conidiophores and conidia. The conidial heads appear radial and they split into columns (biseriate). There is the presence of phialides on the conidiophores	*Aspergillus niger*
Street food 1	i	Colonies are fluffy/cottony green to yellow-green in appearance with a distinct radial/fan-like growth pattern	Septate hyphae. Relatively long, Smooth-walled conidiophores. The conidia are spherical/elliptical and produced in well-defined chains	*Penicillium chrysogenum*
	ii	Cottony/fluffy colonies which are initially white to yellow and then turning black with time. The reverse is white to yellow.	Smooth and coloured conidiophores and conidia. The conidial heads appear radial and they split into columns (biseriate). There is the presence of phialides on the conidiophores	*Aspergillus niger*
Street food 2	i	Fast-growing Greenish-grey to Greyish-white fluffy/velvety colonies of moderate size with powdery surface	Conidiophores are broom/brush-like with small and spherical conidia	*Aspergillus fumigatus*
	ii	Fast-growing Greenish-grey to Greyish-white fluffy/velvety colonies of moderate size with powdery surface	Conidiophores are broom/brush-like with small and spherical conidia	*Aspergillus fumigatus*
	iii	Colonies are fluffy black with white to cream edges and mostly white when viewed from the reverse of the Petri dish	Spherical sporangia with brush-like sporangiophores	*Aspergillus flavus*
Street food 3	i	Cottony/fluffy colonies which are initially white to yellow and then turning black with time. The reverse is white to yellow.	Smooth and coloured conidiophores and conidia. The conidial heads appear radial and they split into columns (biseriate). There is the presence of phialides on the conidiophores	*Aspergillus niger*

Table 3: Concentration of aflatoxins in schoolchildren's home-packed meals and street foods

Samples	Aflatoxins concentration				
	B_1(µg/kg)	**B_2(µg/kg)**	**G_1(µg/kg)**	**G_2(µg/kg)**	**Total aflatoxin (µg/kg)**
School A (Fried Rice)	1.00	0.96	1.23	1.16	4.35
School B (White Rice & stew)	0.98	0.93	1.19	1.12	4.22
School C (Beans)	0.74	0.71	0.90	0.85	3.20
School D (Noodles)	1.12	1.07	1.36	1.29	4.84
School E (Pasta)	0.96	0.86	1.12	1.02	3.96
Food vendor (White Rice & stew)	1.10	1.08	1.33	1.26	4.77
Acceptable maximum limits (EU)	2.00	2.00	2.00	2.00	4.00
Acceptable maximum limits (Nig)	2.00	5.00	5.00	5.00	10.00

Table 4: Concentration of aflatoxins produced by fungi isolated from the food samples

Fungal isolates	Aflatoxins concentration					
	B_1(µg/kg)	B_2(µg/kg)	G_1(µg/kg)	G_2(µg/kg)	Total aflatoxin (µg/kg)	Acceptable Maximum limit (µg/kg)
N8 (*Penicillium citrinum*)	0	0	0	0	0.00	20.00
N9 (*Aspergillus brunneoviolaceus*)	2.62	2.31	3.25	3.12	11.30	20.00
N10 (*Aspergillus brunneoviolaceus*)	2.60	2.28	3.24	3.10	11.22	20.00
N11 (*Aspergillus fumigatus*)	2.71	2.58	3.30	3.12	11.71	20.00
N12 (*Aspergillus niger*)	2.98	2.84	3.63	3.63	13.08	20.00
N14 (*Aspergillus aculaetus*)	4.00	3.91	3.91	4.71	16.53	20.00

6. CONCLUSION

The fungal profile of home-packed meals of pupils in Benin City revealed the presence of common spoilage and toxigenic species of *Aspergillus* and *Penicillium*. Detected aflatoxin concentrations were below the Nigerian permissible limits, indicating that the meals were generally safe for consumption. However, the occurrence of aflatoxigenic fungi highlights the potential for contamination under poor storage or handling conditions. Continuous surveillance, coupled with improved food hygiene and handling practices, is recommended to minimize mycotoxin exposure risks among schoolchildren.

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