



Prevalence and Environmental Determinants of Aflatoxin Contamination in Maize Sold in Open Markets across Nasarawa State, Nigeria

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Abstract

This study evaluated the prevalence of aflatoxins (AFB1, AFB2, AFG1, and AFG2) in maize sold across 13 Local Government Areas (LGAs) in Nasarawa State, Nigeria, and examined how environmental factors contribute to fungal growth and mycotoxin production. Maize samples (130) were subjected to total heterotrophic fungal count (THFC) analysis using standard microbiological methods, while aflatoxin levels were measured using thin-layer chromatography (TLC) combined with densitometry. Statistical methods, including analysis of variance (ANOVA) and regression modeling, were utilized to clarify spatial contamination trends and identify environmental variables that could predict contamination. The findings revealed significant aflatoxin contamination, with 85.4% of samples surpassing the 20-ppb safety limit set by the Standards Organisation of Nigeria (SON). *Aspergillus flavus*, the main producer of aflatoxin B1, was found in 94% of the samples, followed by *Fusarium verticillioides* (71%) and *Aspergillus niger* (59%). Regression analysis showed a strong correlation ($r = 0.710$, $P < 0.001$) between fungal load and AFB1 levels, with humidity explaining 41.1% of the variability in THFC. The highest AFB1 concentration (137.10 ± 15.10 ppb) was found in Doma, while Lafia showed consistently lower contamination levels, likely due to better post-harvest handling practices. This study underscores the urgent need for targeted interventions to reduce aflatoxin contamination, such as rapid drying, hermetic storage systems, and educating farmers on preventing fungal growth. It also recommends implementing aflatoxin surveillance programs and researching resistant maize varieties to improve food safety and public health in Nasarawa State.

Keywords: Aflatoxin, maize, *Aspergillus flavus*, contamination, Nasarawa State

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Introduction

Maize (*Zea mays* L.) is one of the most widespread agricultural species that can thrive in many different types of agroecological settings. It ranks third among all cereals globally, after rice and wheat [1, 2]. In sub-Saharan Africa, maize represents the major cereal crop, as a staple food for over 1.2 billion people across the continent and into Latin America. On the African continent, there are more than 300 million depending on maize as a staple food [3]. In Nigeria, maize remains the most widely grown cereal crop; its production was estimated at 11 million tons in 2019 [4]. It also provides an important source of cash income for about 98% of the rural households practicing small-scale agriculture [5]. Beyond its role as a staple food, maize supports various industries, including animal feed production, biofuels, syrups, starch, and alcohol [6]. Nigerian households directly consume 10–15% of the country's total maize production [3].

Maize, being rich in carbohydrates, proteins, fats, fiber, vitamins, and minerals is a common food ingredient in the traditional Nigerian diet, particularly for foods like aadun, ogi, kokoro, and tuwomasara, and drinks like kunu [1]. Despite the nutritional value of the crop and its economic importance, there are common contamination incidents with mycotoxins, particularly

aflatoxins, in Nigeria [7]. Aflatoxins, toxic secondary metabolites produced predominantly by *Aspergillus flavus* and occasionally by other fungal genera like *Penicillium* and *Fusarium*, pose significant health and economic challenges. Poor farming and storage practices exacerbate contamination, creating conducive conditions for aflatoxigenic fungi to thrive [8]. Aflatoxins, including AFB1, AFB2, AFG1, and AFG2, are highly resistant to a wide range of thermal and chemical treatments. Moreover, they pose a double threat not only to detriment national economies but also, what is of paramount importance, have carcinogenic effects and suppress immune systems, with even acute aflatoxicosis in humans and animals [9, 7]. Aflatoxins represent a widely recognized concern on a global scale, with Africa exhibiting the highest prevalence, trailed by Asia, Europe, and the Americas [9].

Measures of regulation have been enforced to reduce exposure levels. For instance, the FDA allows only 20 ppb of total aflatoxins in food, while in the EU, the limits are much lower: for total aflatoxins at 10 ppb and AFB1 at 5 ppb in maize and rice [10]. The Standards Organization of Nigeria and the National Agency for Food and Drug Administration and Control have thus set permissible thresholds for the incidence of



aflatoxins in foods within Nigeria for protection against such mycotoxins. SON limits the permissible limits of 10 ppb for maize and sorghum, 20 ppb for groundnuts, and 4 ppb for groundnut cake and sesame seeds. While the acceptable limits according to NAFDAC are 4 µg/kg for ready-to-eat food commodities and 10 ppb for raw food commodities [11, 12]. Several studies carried out in Nigeria reported widespread contamination of aflatoxins. Adetunji *et al.* [13] identified Ondo State as a region of significant concern, documenting aflatoxin concentrations reaching 1,548.96 µg/kg in maize samples. Likewise, in Kaduna State, 93.3% of maize samples collected from the Giwa community displayed differing levels of aflatoxin contamination [14]. Nonetheless, information regarding aflatoxin contamination in maize grains, especially in Nasarawa State, is still scarce. The level of aflatoxin in grains available within the major markets of Nasarawa State should therefore be of vital importance because it is a staple crop within the region. This study will, therefore, fill this gap by ascertaining whether maize grains are contaminated due to aflatoxin and thus provide necessary data to support food safety.

Materials and Methods

Sampling and sample collection

The collection was made across 13 LGAs of Nasarawa State, North-Central Nigeria; these are Obi, Awe, Keana, Lafia, Doma, Kokona, Karu, Keffi, Toto, Nasarawa, Akwanga, Nasarawa-Eggon, and Wamba. These LGAs were considered to be of equal and full representation as separate strata in terms of covering both the geographical and climatic variation of the state. Altogether, 130 maize samples were collected, amounting to 10 samples per LGA. Stratified sampling was used to account for regional differences in fungal contamination and levels of aflatoxin; thus, one kilogram of maize grains was purchased from five cluster points within major markets in each LGA. The samplings were carried out at three intervals, separated by one month each, between February 2 and July 1, 2024, in order not to miss any important temporal diversities and to avoid seasonal bias as far as possible. Also, environmental parameters were recorded properly during sampling. The ambient temperature was measured with the help of a thermometer, which was kept very close to the maize for determination of correct ambient exposure. The relative humidity was measured with a digital hygrometer and the moisture content with a moisture content meter. The samples of grains were carefully collected after procurement and kept in coolers containing crushed ice packs. Each sample was labeled properly and then enveloped in sterile plastic bags, after which transportation to the Federal University of Lafia, Microbiology Laboratory, for analysis, was done. Consequently, strict sterility was followed to avoid even the slightest risk of cross-contamination. Such comprehensive and systematic sampling, in conjunction with extended environmental monitoring, has produced quite exhaustive dataset

related to fungal infection and assessment of aflatoxin levels in maize over the whole studied area.

Fungal species assessment in groundnuts

Fungal diversity in maize was determined using the agar plate method. Apparently healthy maize seeds were randomly picked with a sterile spatula and surface-sterilised in a clean beaker containing 2% sodium hypochlorite for 3 min. The seeds were subsequently washed four times with sterile distilled water to remove the residual sterilising agent using the method of Akharenegbe *et al.* [8]. Then, the sterilised seeds of maize were immediately plated on pre-prepared PDA plates supplemented with 100 µg/mL chloramphenicol, which inhibits bacterial growth. The inoculated plates were incubated at 25°C, and the growth of fungal colonies was observed within 3-5 days. Identification was based on the gross and microscopic morphology of the isolated fungi. Morphological traits were juxtaposed with reference descriptions found in a mycology atlas to ensure precise identification, conforming to the methodology described by Chuku [15].

Determination of aflatoxin concentration

Aflatoxin content of the maize was determined using the method described by Sobolev and Dorner [16] with modification. Twenty grammes of the maize seeds were homogenized with 100 mL of 70% methanol to extract aflatoxins [17]. The resultant mixture was vigorously agitated for 60 seconds in a 250 mL conical flask. To this, 40 mL of the filtrate was added to 40 mL of a 10% sodium chloride solution and 25 mL of hexane in the flask. This solution was shaken on an orbital shaker Scientific Industries, Bohemia, NY for 30 min at 400 rpm. At the end of the shaking, 25 mL of dichloromethane was added to the solution, and allowed to separate for 60 seconds. Then, the lower phase was transferred in a beaker and brought to dryness under a fume hood on a bed of 20 g of anhydrous sodium sulfate [18]. The dry extracts obtained were dissolved in 1 mL of dichloromethane and transferred to Eppendorf tubes in order to be then prepared for scanning densitometry analysis. For analysis, 4 µL of the extracts, together with aflatoxin standards (Supelco, Bellefonte, PA), were spotted onto TLC aluminium silica gel 60 F254 plates (20 × 10 cm; Merck, Darmstadt, Germany). TLC plates were scanned by a CAMAG TLC Scanner 3 at 365 nm for the detection of aflatoxins. The intensities of each spot measured using Densitometry software (winCATS 1.4.2 Camag, AG, Muttenz, Switzerland). Concentrations of the aflatoxin compounds were calculated by comparing the peak area calibration curve of the aflatoxin standards to those of the sample chromatograms.

Data analysis

Prior to the analysis, quantitative data on aflatoxins in maize samples were systematically coded and entered into Microsoft Excel spreadsheets. Analysis of variance (ANOVA) was done to determine any statistically significant mean differences in the level of aflatoxin production between maize samples collected from the markets of 13 Local Government Areas (LGAs). One

way analysis of variance (ANOVA) was used to investigate whether the maize market samples from 13 LGAs are significantly different in aflatoxin production through post hoc assessment. Simple linear regression was used to evaluate the relationship between individual environmental factors (temperature, humidity, and moisture content) and total heterotrophic fungal count (THFC). A multiple regression model was thus fitted to investigate the interactions of these factors as well as the environmental conditions in order to determine the levels of aflatoxin contamination. A chi-square test was done to assess the strength of association between categorical variables which in this case were contamination categories (ND, <10 ppb, ≥20 ppb) of maize samples. These dual approaches helped in the comprehensive understanding of the regional differences in the aflatoxin contamination and the climatic conditions affecting the maize of the region.

Results and Discussion

Percentage frequency of fungal species in maize samples across the LGAs in Nasarawa State

The analysis of fungal species isolated from maize samples across LGAs in Nasarawa State reveals significant variability in their occurrence (Table 1). *Aspergillus flavus* emerged as the most prevalent species, detected in 94% of the samples across all LGAs, followed by *Fusarium verticillioides* (71%) and *Aspergillus niger* (59%). Moderately prevalent species included *Curvularia spp.* (53%), *Mucor spp.* (53%), and *Cladosporium spp.* (48%). Conversely, some fungal species, such as *Penicillium expansum* (7%) and *Fusarium subglutinans* (2%), were infrequently detected. The data show geographical variability in fungal occurrence. In Akwanga, *Aspergillus flavus* and *Fusarium verticillioides* were found in 100% of the samples, underscoring the high susceptibility of maize in this area to these fungi. On the other hand, Lafia exhibited the lowest frequency of *Aspergillus flavus* (40%).

Table 1: Percentage frequency of fungal species across Local Government Areas (LGAs) of Nasarawa State

Fungi species	Akwanga N(%)	Awe N(%)	Doma N(%)	Karu N(%)	Keffi N(%)	Kokona N(%)	Keana N(%)	Lafia N(%)	Nasarawa N(%)	N. Eggon N(%)	Obi N(%)	Toto N(%)	Wamba N(%)	Total
<i>Aspergillus flavus</i>	10 (100)	10(100)	10(100)	7(70)	7(70)	5(50)	5(50)	4(40)	8(80)	7(70)	5(50)	7(70)	9(90)	94(94)
<i>Fusarium verticillioides</i>	10(100)	10(100)	10(100)	3(30)	5(50)	3(30)	3(30)	6(60)	5(50)	3(30)	3(30)	5(50)	5(50)	71(71)
<i>Aspergillus niger</i>	7(70)	7(70)	10(100)	5(50)	4(40)	3(30)	1(10)	5(50)	4(40)	3(30)	2(20)	4(40)	4(40)	59(59)
<i>Curvularia spp</i>	0(0)	3(3)	1(10)	4(40)	5(50)	5(50)	6(60)	5(50)	5(50)	4(40)	4(40)	6(60)	5(50)	53(53)
<i>Mucor spp</i>	3(30)	3(30)	3(30)	6(60)	4(40)	5(50)	4(40)	5(50)	4(40)	5(50)	4(40)	4(40)	3(30)	53(53)
<i>Cladosporium spp</i>	1(10)	1(10)	2(20)	5(50)	4(40)	5(50)	4(40)	4(40)	4(40)	4(40)	4(40)	6(60)	4(40)	48(48)
<i>Fusarium proliferatum</i>	5(50)	7(70)	7(70)	0(0)	0(0)	0(0)	1(10)	3(30)	2(20)	3(30)	4(40)	2(20)	5(50)	39(39)
<i>Trichoderma spp</i>	3(30)	2(20)	1(10)	4(40)	2(20)	3(30)	3(30)	2(20)	2(20)	4(40)	4(40)	5(50)	4(40)	39(39)
<i>Penicillium verrucosum</i>	6(60)	6(60)	1(10)	0(0)	2(20)	2(20)	2(20)	1(10)	0(0)	1(10)	2(20)	2(20)	1(10)	26(26)
<i>Aspergillus parasiticus</i>	4(40)	3(30)	0(0)	2(20)	3(30)	2(20)	5(50)	2(20)	3(30)	2(20)	2(20)	4(40)	2(20)	34(34)
<i>Fusarium oxysporum</i>	4(40)	3(30)	2(20)	0(0)	3(30)	1(10)	2(20)	2(20)	2(20)	3(30)	3(30)	2(20)	3(30)	30(30)
<i>Penicillium citrinum</i>	4(40)	4(40)	0(0)	4(40)	2(20)	2(20)	2(20)	2(20)	2(20)	1(10)	0(0)	3(30)	3(30)	29(29)
<i>Aspergillus chevalieri</i>	0(0)	2(20)	1(10)	3(30)	2(20)	4(40)	2(20)	2(20)	0(0)	4(40)	2(20)	2(20)	3(30)	27(27)
<i>Rhizopus spp</i>	0(0)	2(20)	2(20)	3(30)	1(10)	2(20)	4(40)	3(30)	3(30)	1(10)	2(20)	1(10)	2(20)	26(26)
<i>Aspergillus viticola</i>	1(10)	2(20)	1(10)	3(30)	3(30)	3(30)	3(30)	1(10)	0(0)	3(30)	0(0)	0(0)	3(30)	23(23)
<i>Fusarium graminearum</i>	1(10)	1(10)	3(30)	2(20)	2(20)	1(10)	1(10)	3(30)	3(30)	1(10)	3(30)	1(10)	1(10)	23(23)
<i>Alternaria alternata</i>	5(50)	2(20)	2(20)	2(20)	3(30)	1(10)	1(10)	1(10)	1(10)	0(0)	1(10)	0(0)	2(20)	21(21)
<i>Aspergillus penicillioides</i>	1(10)	1(10)	0(0)	2(20)	2(20)	1(10)	2(20)	2(20)	1(10)	1(10)	3(30)	3(30)	1(10)	20(20)
<i>Penicillium expansum</i>	3(30)	1(10)	3(30)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	7(7)
<i>Fusarium subglutinans</i>	2(20)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	0(0)	2(2)

Table 2: Mean count and standard deviation of THFC and aflatoxin levels across different LGAs in Nasarawa State

Locations	THFC x 10 ³ CFU/g	Aflatoxin B1 (ppb)	Aflatoxin B2 (ppb)	Aflatoxin G1 (ppb)	Aflatoxin G2 (ppb)
	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD	Mean ± SD
Akwanga	4.92 ± 0.40 ^{ab}	85.00 ± 24.61 ^{ab}	16.50 ± 13.33	4.10 ± 6.06	1.60 ± 3.06
Awe	4.44 ± 0.84 ^a	82.40 ± 51.31 ^b	27.20 ± 24.02	6.10 ± 6.79	1.78 ± 2.64
Doma	6.50 ± 1.01 ^b	137.10 ± 15.10 ^b	22.20 ± 12.56	5.40 ± 2.88	1.30 ± 1.34
Karu	4.92 ± 1.42 ^{ab}	109.20 ± 46.58	22.90 ± 12.19	7.70 ± 5.76	1.50 ± 1.78
Keffi	5.51 ± 1.20 ^{ab}	103.60 ± 55.24 ^b	22.70 ± 17.14	4.80 ± 5.18	1.20 ± 1.55
Kokona	5.25 ± 1.36 ^{ab}	90.40 ± 65.36 ^{ab}	14.50 ± 16.10	4.20 ± 5.55	0.80 ± 1.14
Keana	4.77 ± 1.04 ^{ab}	82.70 ± 51.09 ^{ab}	16.90 ± 13.82	4.90 ± 5.36	1.20 ± 1.62
Lafia	4.37 ± 1.20 ^a	52.80 ± 57.65 ^a	15.40 ± 20.91	2.10 ± 3.57	0.20 ± 0.42
Nasarawa	4.96 ± 0.91 ^{ab}	110.80 ± 45.12 ^{ab}	28.40 ± 19.38	6.50 ± 6.10	0.90 ± 1.52
N. Eggon	4.70 ± 1.22 ^a	86.20 ± 51.24 ^{ab}	16.40 ± 14.58	2.40 ± 2.22	0.20 ± 0.42
Obi	5.39 ± 1.88 ^{ab}	86.70 ± 55.13 ^{ab}	19.00 ± 17.43	5.00 ± 6.00	1.00 ± 1.70
Toto	5.30 ± 1.14 ^{ab}	121.00 ± 14.17 ^b	23.10 ± 12.16	6.80 ± 4.21	1.80 ± 1.48
Wamba	5.99 ± 0.90 ^{ab}	124.90 ± 15.68 ^b	24.30 ± 12.04	5.80 ± 4.52	1.30 ± 2.00
F. Values	2.66	2.54	1.41	1.41	0.76
P. Values	0.003	<0.005	0.176	0.182	0.684

The values represent the means of three replicates and are expressed as Mean ± Standard Deviation. Means sharing the same alphabet are statistically different, while means with different alphabets are not significantly different at P= 0.05, Units: ppb (parts per billion); CFU/g (colony-forming units per gram)



Mean fungal counts and aflatoxin contamination across the LGAs in Nasarawa State

Table 2 presents the distribution of the mean total heterotrophic fungal count (THFC) and aflatoxin contamination across the 13 LGAs in Nasarawa State. Wamba recorded the highest mean THFC ($5.99 \pm 0.90 \times 10^3$ CFU/g), while Lafia exhibited the lowest ($4.37 \pm 1.20 \times 10^3$ CFU/g). Regarding aflatoxin levels, AFB1 concentrations were highest in Doma (137.10 ± 15.10 µg/kg) lowest in Lafia (52.80 ± 57.65 µg/kg). AFB2 levels peaked in Awe (27.20 ± 24.02 µg/kg), with the minimum found in Kokona (14.50 ± 16.10 µg/kg). For AFG1, Karu recorded the highest mean concentration (7.70 ± 5.76 µg/kg), while Lafia had the lowest (2.40 ± 2.22 µg/kg). AFG2 levels were highest in Toto (1.80 ± 1.48 µg) and lowest in Lafia (0.20 ± 0.42 µg/kg) and Nasarawa Eggon (0.20 ± 0.42 µg/kg).

Relationship between environmental factors and key parameters: Total heterotrophic fungal count (THFC) and aflatoxin B1 (AFB1) levels

The relationship between environmental factors (humidity and temperature) and two critical parameters is shown in Table 3: THFC and AFB1 levels. The

equation $THFC = 555.295 + (73.792 \times \text{Humidity})$ indicates a strong positive correlation between humidity and fungal growth ($r=0.641$, $r^2=0.411$, $F^*=89.49$, $P<0.001$), with humidity accounting for 41.1% of the variation in THFC. Conversely, temperature showed a weaker relationship, explaining only 19.1% of THFC variability as per the equation $THFC = -4864.683 + (308.091 \times \text{Temperature})$, which exhibited a moderate correlation ($r=0.437$, $r^2=0.191$, $F^*=30.23$, $P<0.001$). The strong relationship between THFC and AFB1 was evidenced by the equation $AFB1 = -43.861 + (0.0275 \times THFC)$, where fungal load explained 50.4% of the variation in AFB1 levels ($r=0.710$, $r^2=0.504$, $F^*=130.01$, $P<0.001$). Humidity and temperature both positively correlated with AFB1 levels, with humidity exerting a stronger influence. The equation $AFB1 = -84.480 + (2.926 \times \text{Humidity})$ yielded $r=0.657$ $r^2=0.431$, while $AFB1 = -336.413 + (13.354 \times \text{Temperature})$ showed a weaker correlation ($r=0.489$, $r^2=0.239$). The combined effect of humidity and fungal counts yielded the highest predictive accuracy in the equation $AFB1 = -95.034 + (1.523 \times \text{Humidity}) + (0.0190 \times THFC)$, which exhibited a robust correlation ($r=0.757$ $r^2=0.573$, $F^*=85.10$).

Table 3: Regression analysis of environmental factors on total heterotrophic fungal count (THFC) and aflatoxin B1 across the LGAs

Factors	Equation	r*	r ²	SEE	Adj r ²	F*
Humidity	$THFC = 555.295 + (73.792 \times \text{Humidity})$	0.641	0.411	965.936	0.407	89.49
Temperature	$THFC = -4864.683 + (308.091 \times \text{Temp})$	0.437	0.191	1132.451	0.185	30.23
Humidity, Temperature	$THFC = -177.639 + (70.820 \times \text{Humidity}) + (28.232 \times \text{Temp})$	0.642	0.412	968.959	0.403	44.57
THFC	$AflaB1 = -43.861 + (0.0275 \times THFC)$	0.710	0.504	34.355	0.500	130.01
Humidity	$AflaB1 = -84.480 + (2.926 \times \text{Humidity})$	0.657	0.431	36.792	0.427	96.96
Temperature	$AflaB1 = -336.413 + (13.354 \times \text{Temp})$	0.489	0.239	42.545	0.233	40.24
THFC, Humidity	$AflaB1 = -95.034 + (1.523 \times \text{Humidity}) + (0.0190 \times THFC)$	0.757	0.573	32.011	0.566	85.10

* $P < 0.001$

Distribution of aflatoxin contamination levels in maize samples across various LGAs in Nasarawa State based on different safety category

Table 4 shows that 5.4% of the maize samples had no detectable aflatoxin levels, primarily from Awe and Kokona. An additional 7.7% of the samples exhibited very low aflatoxin levels, falling within the European Union (EU) permissible limit of 4 ppb. Only 1.5% of the samples were within the <10 ppb category, meeting the Standards Organisation of Nigeria (SON) permissible limit for maize. Alarming, the majority of samples (85.4%) were highly contaminated with aflatoxins, exceeding the 20-ppb threshold and considered unfit for human consumption. All LGAs recorded samples in this unsafe category, with Akwanga, Wamba, Doma, and Toto showing the highest proportions. The contamination range spanned from 0 to 223 ppb, significantly surpassing both national and international regulatory limits.

Table 4: Prevalence and concentration of aflatoxins based on safety category in maize samples from markets across Local Government Areas in Nasarawa State

LGA	Category of Aflatoxin Concentration (ppb)				Range
	ND	<4	<10	≥ 20	
	N (%)	N (%)	N (%)	N (%)	
Akwanga	0 (0.0)	0 (0.0)	0 (0.0)	10 (7.7)	0 – 223
Awe	2 (1.5)	0 (0.0)	0 (0.0)	8 (6.2)	0 – 222
Karu	0 (0.0)	1 (0.8)	0 (0.0)	9 (6.9)	0 – 221
Keana	0 (0.0)	2 (1.5)	0 (0.0)	8 (6.2)	0 – 221
Lafia	0 (0.0)	4 (3.1)	0 (0.0)	6 (4.6)	0 – 221
Wamba	0 (0.0)	0 (0.0)	0 (0.0)	10 (7.7)	0 – 206
Nasarawa	0 (0.0)	1 (0.8)	0 (0.0)	9 (6.9)	0 – 205
Keffi	1 (0.8)	0 (0.0)	1 (0.8)	8 (6.2)	0 – 200
Doma	0 (0.0)	0 (0.0)	0 (0.0)	10 (7.7)	0 – 196
Kokona	2 (1.5)	1 (0.8)	0 (0.0)	7 (5.4)	0 – 196
Obi	1 (0.8)	1 (0.8)	0 (0.0)	8 (6.2)	0 – 188
Toto	0 (0.0)	0 (0.0)	0 (0.0)	10 (7.7)	0 – 185
N. Eggon	1 (0.8)	0 (0.0)	1 (0.8)	8 (6.2)	0 – 167
Total	7 (5.4)	10 (7.7)	2 (1.5)	111 (85.4)	

ND = Not detected; limit of detection = 0.1 ppb; <4= EU permissible limit; SON permissible limits of 10 ppb for maize, ≥ 20 ppb not fit for human consumption

The high prevalence of *Aspergillus flavus* agrees with its well-documented role as the primary producer of aflatoxins, particularly aflatoxin B1 (AFB1), a potent carcinogen [19]. This fungus thrives in warm, humid climates such as those in Nasarawa State, which explains its widespread prevalence across LGAs [19]. The high frequency of *Fusarium verticillioides* is similarly consistent with its role in fumonisin production, another harmful mycotoxin commonly associated with maize [20]. The diversity in fungal frequencies across LGAs may be attributed to environmental factors (e.g., temperature, relative humidity) and post-harvest handling practices. In areas like Akwanga and Doma, where *Aspergillus flavus* and *Fusarium verticillioides* were detected in 100% of samples, inadequate drying, and substandard storage practices likely contributed to fungal proliferation. By contrast, Lafia's relatively low prevalence of *Aspergillus flavus* may suggest better post-harvest handling, such as hermetic storage bags or well-ventilated storage facilities. The high prevalence of *Aspergillus flavus* is of public health concern due to its ability to produce aflatoxins, which pose serious health risks. Chronic exposure to aflatoxins has been linked to hepatocellular carcinoma, immune suppression, and stunted growth in children [21, 22]. The presence of *Fusarium verticillioides* raises concerns about fumonisin contamination, which has been associated with esophageal cancer and neural tube defects [23]. These findings agree with studies conducted in other places south east, Nigeria, where *Aspergillus flavus* and *Fusarium verticillioides* were identified as the predominant fungal contaminants in maize [24]. However, this study's relatively high prevalence rates for non-toxicogenic fungi, such as *Mucor spp.* and *Cladosporium spp.*, highlight potential regional differences. These differences may reflect differences in climate, agricultural practices, and maize cultivation methods. The widespread occurrence of *Aspergillus flavus* and *Fusarium verticillioides* necessitates urgent interventions to reduce fungal contamination and associated mycotoxin risks. Improved post-harvest practices, such as proper and quick drying after harvesting and better storage infrastructure, are critical to reducing fungal proliferation. Additionally, non-toxicogenic fungi suggest that some environmental niches may support fungal competition, which could be used for the biocontrol programs.

This study investigated the prevalence of aflatoxin B1 and other aflatoxin types (AFB2, AFG1, and AFG2) in maize sold in open markets across 13 Local Government Areas (LGAs) in Nasarawa State. The findings revealed significant geographical diversity in both fungal contamination and aflatoxin levels, underlining the pressing need for public health interventions to reduce aflatoxin exposure. Wamba exhibited the highest THFC ($5.99 \pm 0.90 \times 10^3$ CFU/g), reflecting potential environmental conditions such as humidity, which favours fungal proliferation. Contrarily, Lafia had the lowest THFC ($4.37 \pm 1.20 \times 10^3$), likely due to better post-harvest

handling practices. For aflatoxins, Doma recorded the highest mean AFB1 levels (137.10 ± 15.10 μ g), far exceeding the acceptable limits of 10 ppb recommended by the Standards Organization of Nigeria (SON) and the National Agency for Food and Drug Administration and Control (NAFDAC). Again, Lafia showed the lowest levels of AFB1 (52.80 ± 57.65 ppb). High AFB2 concentrations were observed in Awe (27.20 ± 24.02 ppb), and the lowest in Kokona (14.50 ± 16.10 ppb). Notably, Karu had the highest AFG1 levels (7.70 ± 5.76 ppb), while Toto recorded the highest AFG2 levels (1.80 ± 1.48 ppb). Lafia consistently exhibited the lowest contamination for AFG1 (2.40 ± 2.22 ppb) and AFG2 (0.20 ± 0.42 ppb). These findings agree with a previous study that identified climate and storage conditions as factors that contributed to aflatoxin contamination [25]. However, the significant AFB1 levels in Doma are of public health concern and may reflect local storage or processing practices requiring further scrutiny. The observed aflatoxin contamination levels are consistent with studies from similar agro-climatic regions, such as Kaduna State, Nigeria, where AFB1 levels also exceeded safety limits [26]. The high incidence of aflatoxins, especially AFB1, poses serious health risks, including hepatocellular carcinoma and immune suppression. Economically, contaminated maize may affect export potential and domestic food security. The findings emphasize the urgency for implementing LGAs-specific mitigation programs, including improved drying methods and aflatoxin-binding agents during storage.

The findings of this study demonstrate that humidity is an important determinant of fungal growth in maize samples across Nasarawa State, accounting for over 40% of THFC variability. This study aligns with established microbiological concepts where higher humidity provides a conducive environment for fungal sporulation and growth [27]. However, temperature showed a weaker influence, reflecting its secondary role compared to moisture content. The combined model incorporating humidity and temperature slightly improved predictive accuracy, suggesting interactive effects between these variables. This finding of this study is consistent with earlier studies indicating that varying temperature levels help to change the impact of humidity on fungal growth [28]. The strong correlation between THFC and AFB1 levels underscores the role of fungal load in high aflatoxin contamination. This is in agreement with previous research emphasizing that aflatoxin production is primarily dependent on the metabolic activity of fungal species, particularly *Aspergillus flavus* and *Aspergillus parasiticus* [29]. Humidity has also shown a robust direct relationship with AFB1, highlighting its dual role in promoting fungal growth and stimulating secondary metabolite production. Temperature, while significant, showed a weaker correlation, indicating that extreme temperature variations may inhibit aflatoxin biosynthesis despite fungal proliferation. These results underline the need for strict humidity control during maize storage to



minimize fungal growth and aflatoxin contamination. The significant predictive value of THFC for AFB1 suggests that monitoring fungal counts could serve as an early warning system for potential aflatoxin contamination, providing actionable insights for stakeholders in maize storage and distribution.

The findings highlighted a significant issue of aflatoxin contamination in maize samples throughout Nasarawa State, with a staggering 85.4% of the samples surpassing the 20-ppb limit established by international regulatory bodies like the European Union (EU) and the Standards Organisation of Nigeria (SON). This alarming prevalence of unsafe aflatoxin levels raises serious food safety concerns that could impact public health, agriculture, and trade. The high number of samples in the ≥ 20 ppb category indicates an urgent need for government action. Aflatoxins, especially aflatoxin B1, are known hepatocarcinogens associated with liver cancer, immune system suppression, and stunted growth in children [22, 30]. The widespread contamination of maize, a key crop in Nigeria, threatens food security and public health. Regular consumption of contaminated maize, even in small amounts, can lead to long-term health issues. The elevated contamination levels found across all local government areas (LGAs) are likely exacerbated by environmental conditions that favor the growth of *Aspergillus flavus* and *Aspergillus parasiticus*, the main aflatoxin producers [8]. Factors such as warm temperatures, high humidity, and poor storage practices, including delayed drying and inadequate storage facilities, significantly contribute to fungal growth and toxin production. LGAs like Akwanga, Wamba, Doma, and Toto, which exhibited the highest rates of unsafe contamination, may face more severe environmental challenges or inadequate post-harvest handling. These findings align with previous research in similar agro-climatic regions, where aflatoxin levels in maize frequently exceed safe limits due to environmental and handling issues [14]. Notably, the contamination levels in this study, reaching as high as 223 ppb, surpass those reported in many other regions of Nigeria, underscoring the critical situation in Nasarawa State. While only 1.5% of samples met the SON permissible limit (< 10 ppb), the presence of 7.7% of samples within the EU limit (< 4 ppb) suggests that some areas may already benefit from improved post-harvest practices, potentially serving as models for broader intervention.

Conclusion

The findings of this research indicates that there is a serious problem of aflatoxin contamination in maize across Nasarawa State, with 85.4 % of samples being above the 20 ppb maximum permissible limit, which endangers food security, human health and the economy. The high occurrence of a mould, *Aspergillus flavus* which is a major producer of aflatoxin and *Fusarium verticillioides* contaminant of fumonisin indicates the importance of climatic factors such as humidity, temperature and poor post-harvest handling in the outbreaks of fungi and toxins. Even though some

areas like Lafia were less contaminated due to more appropriate areas of storage, the troubling amounts of aflatoxins in Doma and Wamba require urgent measures.

Recommendation

Educating farmers and grain traders about the dangers of aflatoxins and other mycotoxins is essential for reducing contamination. Awareness campaigns led by stakeholders in public health and agriculture should highlight the health risks, including liver cancer and immune suppression, while also offering practical preventive measures like rapid drying and proper maize storage. Enhancing aflatoxin monitoring programs at market and storage levels will help ensure adherence to safety regulations and provide useful data for interventions. Promoting regular testing with affordable and quick detection kits is crucial. Improving post-harvest practices is vital. Farmers need training on effective drying methods and the use of hermetic storage bags to limit fungal growth. Furthermore, establishing infrastructure support, such as community drying facilities and better storage centers, is necessary. Investing in research to create maize varieties that are resistant to aflatoxins and exploring natural detoxifiers, like plant-based binders, can offer sustainable solutions to minimize contamination risks.

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