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Post- harvest handling practices, moisture content and aflatoxin status in maize samples from selected hammer milling centers in Uganda

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ABSTRACT

Aflatoxins are potent carcinogenic, teratogenic, immunosuppressive, and growth-inhibitory secondary metabolites of selected fungal species affecting both humans and animals. In Uganda, emerging evidence points to an increasing prevalence of aflatoxin contamination in cereal-based food products at toxic concentrations, necessitating routine surveillance to safeguard public health. This study evaluated aflatoxin levels in maize collected from milling centers across eight districts and investigated associated post-harvest handling practices. Moisture content was determined using a hot-air oven; aflatoxins were detected by Thin Layer Chromatography (TLC) and quantified via competitive ELISA. Data were analyzed using Microsoft Excel (2016), Graph Pad Prism (v6.0), and IBM SPSS (2019). Key informant interviews revealed that 55% of moldy maize was processed for human consumption, 30% was diverted to animal feed, and 15% was discarded. Regarding drying practices, 58% of respondents used tarpaulins, 16% dried maize directly on bare ground, and 26% milled maize without prior quality assessment. Dryness assessment methods were largely informal: 38% used the biting test, 18% a metallic rod, 9% each used peeling or noise tests, while only 10% employed moisture meters. Notably, 17% did not assess dryness at all. While all samples exhibited moisture content below the recommended 12.5% threshold, significant inter-district differences were observed ($p = 0.0014$). Of the 119 maize samples analyzed, 50.4% tested positive for aflatoxins. Aflatoxin G predominated in maize samples from most districts, whereas aflatoxin B was more prevalent in samples from Kampala district. Mean total aflatoxin concentrations ranged from 0.90 ± 0.46 ppb (Kampala) to 54.18 ± 0.0 ppb (Mityana), with significant regional variation ($p = 0.0034$). Alarmingly, all maize samples from Mityana district had total aflatoxin concentrations above the Uganda National Bureau of Standards (UNBS) regulatory limit of 10 ppb. Regression analysis ($p = 0.474$, $R^2 = 0.0089$) indicated a weak association between moisture content and aflatoxin levels, suggesting additional contamination drivers. These findings underscore the need for improved post-harvest handling and stricter regulatory enforcement to mitigate aflatoxin-associated public health risks in Uganda.

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Key words: Aflatoxins, Aspergillus, Chromatography, Enzyme Linked-Immunosorbent Assay, Milling center, Mycotoxins, *Zea mays*

RÉSUMÉ

Les aflatoxines sont des métabolites secondaires puissamment cancérigènes, tératogènes, immunosuppresseurs et inhibiteurs de croissance produits par certaines espèces fongiques, affectant à la fois les humains et les animaux. En Ouganda, des preuves émergentes indiquent une prévalence croissante de la contamination des produits alimentaires à base de céréales par des aflatoxines à des concentrations toxiques, nécessitant une surveillance de routine pour protéger la santé publique. Cette étude a évalué les niveaux d'aflatoxines dans le maïs collecté auprès de centres de meunerie dans huit districts et a examiné les pratiques post-récolte associées. La teneur en humidité a été déterminée à l'aide d'un four à air chaud ; les aflatoxines ont été détectées par chromatographie sur couche mince (TLC) et quantifiées via ELISA compétitif. Les données ont été analysées à l'aide de Microsoft Excel (2016), Graph Pad Prism (v6.0) et IBM SPSS (2019). Les entretiens avec des informateurs clés ont révélé que 55 % du maïs moisi était transformé pour la consommation humaine, 30 % réorienté vers l'alimentation animale, et 15 % jeté. Concernant les pratiques de séchage, 58 % des répondants utilisaient des bâches, 16 % séchaient le maïs à même le sol, et 26 % le moulaient sans évaluation préalable de la qualité. Les méthodes d'évaluation de la sécheresse étaient largement informelles : 38 % utilisaient le test de morsure, 18 % une tige métallique, 9 % utilisaient le test de pelage ou le bruit, tandis que seulement 10 % utilisaient un humidimètre. Notamment, 17 % n'évaluaient pas du tout la sécheresse. Bien que toutes les échantillons aient montré une teneur en humidité inférieure au seuil recommandé de 12,5 %, des différences significatives entre les districts ont été observées ($p = 0,0014$). Sur les 119 échantillons de maïs analysés, 50,4 % étaient positifs pour les aflatoxines. L'aflatoxine G prédominait dans les échantillons de la plupart des districts, tandis que l'aflatoxine B était plus répandue à Kampala. Les concentrations moyennes totales d'aflatoxines allaient de 0,90 ± 0,46 ppb (Kampala) à 54,18 ± 0,0 ppb (Mityana), avec des variations régionales significatives ($p = 0,0034$). Alarmant, tous les échantillons de maïs du district de Mityana dépassaient la limite réglementaire de 10 ppb fixée par l'UNBS. L'analyse de régression ($p = 0,474$, $R^2 = 0,0089$) a montré une faible association entre la teneur en humidité et les niveaux d'aflatoxines, suggérant d'autres facteurs de contamination. Ces résultats soulignent la nécessité d'améliorer les pratiques post-récolte et de renforcer l'application de la réglementation pour atténuer les risques de santé publique liés aux aflatoxines en Ouganda.

Mots clés : Aflatoxines, Aspergillus, Chromatographie, Test immuno-enzymatique ELISA, Centre de meunerie, Mycotoxines, Zea mays

INTRODUCTION

Studies have reported aflatoxin presence in maize in levels that are above regulatory limits across the globe (Omara *et al.*, 2021). Aflatoxins are secondary metabolites produced by certain species of *Aspergillus* fungi, primarily *Aspergillus flavus* and *Aspergillus parasiticus*. These fungi thrive in warm [> 25^0C], humid conditions [relative humidity of > 40%] and can contaminate a variety of agricultural commodities, including maize. Aflatoxins are highly toxic and can cause severe health problems in both humans and animals. Exposure to aflatoxins has been linked to liver cancer, immune suppression, stunted growth, and reduced reproductive performance (Nasir *et al.*, 2021). The presence of aflatoxins in foodstuff in many regions, poses a

significant public health threat. This therefore calls for regular monitoring of aflatoxin status of staple foods more so in developing countries due to improper implementation of aflatoxin prevention and control strategies. There are several factors that contribute to the contamination of foodstuffs including maize with aflatoxins. One of the most important risk factors are climatic conditions; warm and humid environments that are ideal for the growth and development of *Aspergillus* fungi (Jaibangyang *et al*., 2021; Abel Palacios *et al*., 2022; Marín *et al*., 2024). Additionally, poor post-harvest handling practices such as improper drying, storage, and processing can create conditions conducive for fungal growth and aflatoxin production (Akumu *et al*., 2020). Crop stress including drought, insect damage, or other adverse conditions can also weaken plants and make them more susceptible to fungal infection (Kumar *et al*., 2021).

Previous studies conducted in Uganda to investigate the prevalence and risk factors of aflatoxin contamination in maize have demonstrated an increasing trend (Benkerroum, 2020; Omara *et al*., 2020; Wacoo *et al*., 2020; Mwesige *et al*., 2023). To address the aflatoxin burden in Uganda, the government has implemented various prevention and control initiatives. In particular, the government has educated farmers and trade agents on proper post-harvest handling practices, promoted the use of aflatoxin-resistant maize varieties in farming communities, and established monitoring programs to detect and prevent aflatoxin contamination (Akumu *et al*., 2020; Kaale *et al*., 2021). Despite all these efforts, aflatoxin presence in various food matrices has remained a challenge in Uganda (Akullo *et al*., 2023; Atukwase *et al*., 2024). With increasing hepatoma frequency in Uganda that may be attributed to aflatoxins presence in staple foods (Kitya *et al*., 2010; Kang *et al*., 2015), there is a dire need to continuously monitor aflatoxin status of foods in Uganda. Therefore, this study assessed post-harvest handling practices and established aflatoxin concentrations in maize samples [the most consumed foodstuff] from selected milling centers in the eight regionally distributed districts of Uganda.

MATERIALS AND METHODS

This study assessed post-harvest handling practices, effectiveness of drying methods on reducing moisture content and determined levels of aflatoxin in maize samples from randomly selected milling centers in eight (8) purposively identified districts of Uganda. Using the Hot-air Oven, moisture content determination was used to assess effectiveness of maize drying methods. Detection and quantification of aflatoxins in maize samples were done using Thin Layer Chromatography (TLC) and competitive- ELISA techniques respectively. The RidaScreen Total Aflatoxin ELISA Kit- ART No. R4701 total aflatoxin ELISA kit was procured from PerkinElmer. The reagents used in this study were of analytical grade and were supplied by PubMed Diagnostics- Uganda. Additionally, all tests were done in duplicates alongside the standards. This study was conducted between April and August, 2023.

Study area. Uganda, situated in East Africa, is a landlocked country bordered by Kenya in the East, Tanzania and Rwanda in the South, the Democratic Republic of the Congo in the West and South Sudan in the North (https://en.wikipedia.org/wiki/Uganda). Its diverse landscape features the majestic Lake Victoria to the south and the Rwenzori Mountains to the west. The capital city, Kampala, lies near the northern shores of Lake Victoria. Uganda's population is around 47 million people, with a youthful demographic, as over half are under 15 years old (Uganda Bureau of Statistics [UBOS] 2024). The selection of the two districts from each region was guided by the rate of production and

consumption of maize. Although maize production and consumption in Uganda varies greatly, the eastern region dominates at 37%, followed by northern region at 34%. The Western and Central regions represent 15 % and 14 % of national maize production and consumption respectively. Thus, in this study, maize samples were collected from 8 purposively selected districts considering the production and consumption rates of maize. From each region, two districts were purposively selected to represent urban and rural settings. In southwestern Uganda, Kasese district represented the urban setting whereas Masindi was considered a rural district. Lira and Pader were selected to represent Urban and rural districts of Northern Uganda respectively. In Central Uganda, Kampala City and Mityana districts were selected to represent Urban and rural areas respectively. In Eastern Uganda, Jinja city was the urban study area while Kamuli district represented the rural setting. From each district, milling centers were identified with the help of community-based research assistants. A milling center was defined as the hammer mill that processes maize for public consumption. Centers processing animal feeds were not selected for this study.

Sample size determination. In this study, the theoretical population was negligibly small as they were fewer hammer milling centers in each district. Initially, the number of hammer milling centers for each district was established via desktop review considering the available production records of each district. However, there was insufficient information about the number of maize hammer milling centers via this method. Thus, the location and number of these centers were established with the help of community- based research assistants. Eventually, 15 composite maize samples were collected from each district translating into a total of 120 samples.

SAMPLE COLLECTION

For each composite maize sample, 250 g were purchased from the mill owners. Thereafter, samples were immediately divided into two equal subsamples (125 g each) and placed into bags labeled as either moisture sample or aflatoxin sample. Samples for moisture content determination were immediately placed into a sealable box whereas aflatoxin samples were immediately placed in cool boxes containing ice packs. All samples were immediately transported to Analytical Biosciences Laboratory, College of Veterinary Medicine, Animal Resources and Biosecurity, Makerere University for either immediate processing or storage. The moisture samples were immediately placed into desiccators while aflatoxin samples were kept at -20^{o}C until analysis.

Study design. The philosophical underpinning of this study was that of a blended approach exploiting a cross-sectional study design. Key informant interviews were conducted using a check list of questions to assess the handling of maize at hammer mill centers in eight (8) districts of Uganda. Moisture content determination was done using a hot-air-oven method (Nielsen, 2010). Conversely, Thin Layer chromatography [TLC] (Hussain *et al*., 2021) and competitive Enzyme Linked Immunosorbent Assay [c-ELISA] (Schuller *et al*., 1976) were used in the detection of aflatoxin groups and quantification of total aflatoxins in maize samples respectively.

DATA COLLECTION METHODS. The qualitative and quantitative methods were used to collect data as described below:

Key informant interviews. Initially, a verbal consent was obtained from the caretaker of the milling center after explaining the purpose of the study. Using a checklist of questions, relevant information on post-harvest handling practices of maize was obtained.

Moisture content determination. For each sample, 5 g of maize flour were weighed in duplicate into labeled pre-weighed crucibles. Crucibles containing samples were transferred into a hot-air oven set at 105^0C and left to stand for 24 hours. After 24 hours of drying, samples were cooled in desiccators and reweighed. The procedure was repeated until a constant weight was recorded. The percentage moisture content was calculated using the formula;

$$\% \text{ Moisture content (\%MC)} = \frac{\text{Weight (g) of original sample - Weight (g) of dried sample}}{\text{Weight (g) of original sample}} \times 100$$

Detection of aflatoxin groups in maize samples. This was done in two phases following procedures described by Hussain *et al.* (2021). Initially, aflatoxins were extracted from composite maize samples by solvent method followed by screening using Thin Layer Chromatography.

Extraction of total aflatoxins from maize samples. During this phase, 50 g of maize flour were weighed into a labeled conical flask followed by addition of 150 mL of 70% HPLC-grade methanol. The mixture was then shaken using a vortexer at 2500 rpm for 10 minutes. The resulting suspension was allowed to separate for 5 minutes. After 5 minutes of separation, the supernatant was collected into labeled conical flasks and 10 mL of deionized water and 10 mL of absolute HPLC-grade chloroform were sequentially added. The chloroform mixture was then shaken at 2500 rpm for 10 minutes. The resultant mixture was transferred into a marked separating funnel. The contents were then allowed to stand for 5 minutes and the bottom chloroform emulsion layer was collected into labeled bijou bottle. Extracts were then stored at -20ºC until analysis.

Thin Layer Chromatography (TLC) screening of total aflatoxins The TLC plate was activated at 90ºC for 5 minutes in an electric oven. For every sample, 5µL of the chloroform extract were spotted onto activated TLC plate and allowed to dry at room temperature (25ºC) for 5 minutes. The dried plate was developed in acetone: chloroform mixture [1:9 v/v] until the solvent mixture moved ¾ of the plate length. The dried plate was then observed using CAMAG- viewing cabinet at 366 nm for detection of aflatoxin groups.

Quantification of total aflatoxins using c-ELISA. Quantification of total aflatoxins was done following instructions in the Manufacturer's Manual- RidaScreen Total Aflatoxin ELISA Kit- ART No. R4701 with minor modifications customized to our laboratory settings. Briefly, 50 µL of the standard and sample were added into wells in replicates according to the loading plan. This was followed by addition of 100 µL of aflatoxin-HRP conjugate into each well. The plate was then gently rocked for one minute and incubated at 25^0C for 30 minutes. After incubation, the wells were washed three times by adding 250 µL of the wash solution. Thereafter, 100µL of TMB substrate were added into each well, plate gently rocked and incubated at 25^0C for 15 minutes. To each well, 100µL of the stop solution were added. Reading of absorbance was done at 450 nm using a plate reader within 15 minutes. Aflatoxin concentrations in maize samples were computed from the standard curve.

RESULTS

Handling practices of maize samples from milling centers across eight districts in Uganda. In this study, four post- harvest handling practices were evaluated namely; drying platform, methods of assessing dryness level, how maize cereals are stored and fate of mouldy maize (Figure 1). Majority of the mill owners reported use of tarpaulins (58 %) as drying platforms and pallets as storage platforms (55 %). In addition, majority (38%) of the mill owners used biting test as a method

for assessing the level of dryness with only 10 % having used a moisture meter. This approach does not only increase the risk of consuming fungal spores in case of maize contamination with aflatoxigenic fungi but also it is an ineffective method of establishing the dryness status of farm produce including maize. Astonishingly, 55 % of moldy maize were processed for human consumption with 30 % being processed as animal feeds. The two latter practices pose a serious aflatoxin public health risk as they all lead to human aflatoxin exposure

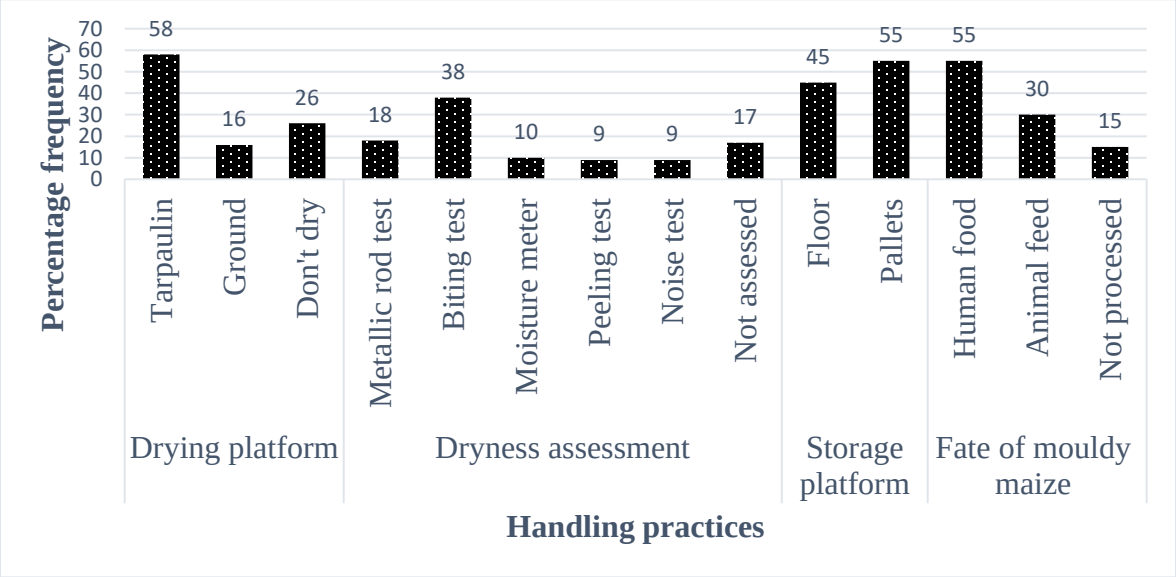

Figure 1. Handling practices of maize samples from four regions of Uganda

Percentage moisture content of maize samples from selected 8 districts of Uganda. This study revealed that the mean percentage moisture content of maize samples from eight districts was within the recommended range (< 12.5 %). These findings imply to a large extent that drying of maize before processing was adequately done. In particular, maize samples from Masindi district had the highest moisture content (9.97 $\pm$ 4.967 %) with samples from Kasese District having the least moisture content (0.29 $\pm$ 0.17 %) (Table 1). Notwithstanding, statistical analysis by ANOVA and Tukey's HD tests revealed significant differences in mean percentage moisture content among maize samples from eight districts of Uganda (p=0.0014; F [DFn, DFd] = (7,112) 3.637. Thus, the null hypothesis that there were no statistically significant differences in moisture content of maize samples from eight districts of Uganda was rejected at significance level of 0.05.

Aflatoxin contamination levels in maize samples per B and G groups. Out of 119 maize samples screened for aflatoxins, 60 samples (50.4%) were positive for aflatoxins. According to aflatoxin groups**,** results of this study revealed that overall, aflatoxin G was the predominant aflatoxin in maize samples. However, samples from Kampala district were largely contaminated with aflatoxin B (Figure 2). This finding is contrary to findings of the majority of the previous studies which reported that Aflatoxin B was the most prevalent aflatoxin. Strikingly, all maize samples from Kasese and Pader were only contaminated with aflatoxin G. These findings suggest that there could be a significant shift in the occurrence and distribution patterns of aflatoxigenic fungal species in food matrices in Uganda.

Table 1. Mean percentage moisture content of maize samples from 8 districts of Uganda

Region	**Central**		**Eastern**		**Western**		**Northern**	
District	Kampala	Mityana	Jinja	Kamuli	Kasese	Masindi	Lira	Pader
Sample size	15	15	15	15	14	15	15	15
Minimum	0.26	0.23	0.07	0.06	0.06	0.07	0.09	0.06
25% Percentile	0.35	0.28	0.12	0.09	0.08	0.10	0.28	0.07
Median	0.65	0.37	0.14	0.24	0.10	0.14	0.43	0.08
75% Percentile	1.32	0.73	0.26	1.68	0.18	3.99	0.51	0.10
Maximum	1.82	49.84	25.81	16.72	2.45	57.96	7.67	3.11
Mean % moisture + SEM	0.83 +0.14	8.99+4.67	2.99+1.94	2.072+1.14	0.29+0.17	9.77+4.967	0.88+0.49	0.30+0.20
Lower 95% CI	0.54	-1.03	-1.17	-0.37	-0.07	-0.89	-0.17	-0.14
Upper 95% CI	1.13	19.02	7.15	4.52	0.65	20.42	1.93	0.73
P value	P= 0.0014							
F (DFn, DFd) value	(7, 112) = 3.637							

Table 2. ANOVA on mean total aflatoxin levels in maize samples from mill centers in eight districts of Uganda

Region	**Central**		**Eastern**		**Western**		**Northern**	
District	**Kampala**	**Mityana**	**Jinja**	**Kamuli**	**Kasese**	**Masindi**	**Pader**	**Lira**
Number of values	6	2	5	15	11	5	10	6
Minimum	0.12	54.18	0.44	0.08	0.217	0.16	0.300	3.13
25% Percentile	0.15	54.18	6.71	7.170	0.37	0.25	0.32	3.88
Median	0.34	54.18	20.36	11.34	0.88	0.55	0.49	44.97
75% Percentile	1.83	54.18	62.23	49.04	3.96	46.11	4.87	62.79
Maximum	2.99	54.18	90.20	85.46	7.88	90.93	33.53	80.54
Mean aflatoxin (ppb)	0.90 + 0.46	54.18 + 0.0	31.65 + 15.63	27.13 + 7.321	2.35 + 0.85	18.65 + 18.07	5.13 + 3.26	39.10 + 12.43
Lower 95% CI	-0.29	54.18	-11.74	11.43	0.45	-31.52	-2.23	7.14
Upper 95% CI	2.09	54.18	75.04	42.83	4.25	68.83	12.50	71.06
P value	0.0034							
F (DFn, DFd) value	F (7, 52) =3.558							

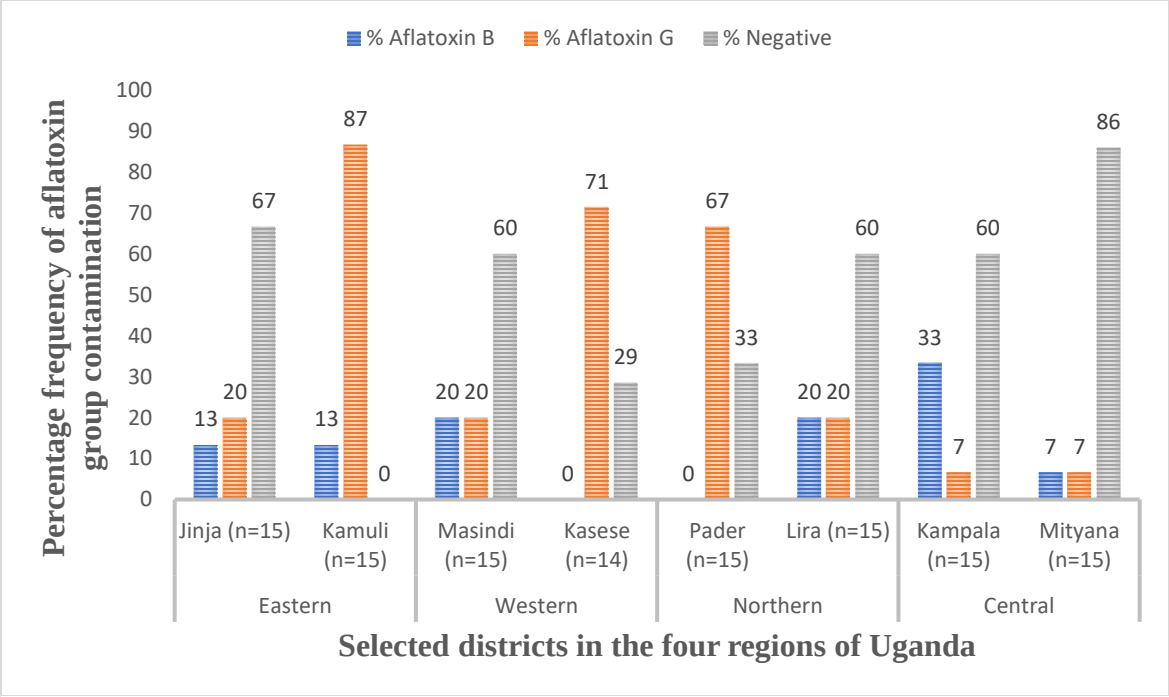

Figure 2. Aflatoxin groups' contamination of maize samples from selected hammer mill centers in Uganda

Total aflatoxin levels in maize samples from eight districts of Uganda. Results of Table 2 show that the mean total aflatoxin concentration of 60 positive maize samples from eight districts ranged from 0.90 $\pm$ 0.46 ppb to 54.18 $\pm$ 0.0 ppb. Maize samples from Mityana district had the highest concentration of total aflatoxins (54.18 $\pm$ 0.0 ppb) followed by Lira (39.10 + 12.43 ppb) and Kampala samples had the least total aflatoxin concentration (0.90 + 0.46 ppb). Analysis of mean total aflatoxin concentration of maize samples from eight districts using ANOVA and Tukey HD tests revealed statistically significant variations (p=0.0034; F [DFn, DFd] = (7, 52) 3.558. Thus, the null hypothesis that there were no statistically significant differences in mean total aflatoxin content of maize samples from eight districts of Uganda was rejected at significance level of 0.05.

Total aflatoxin levels against regulatory limit (10 ppb) by Uganda National Bureau of Standards. In this study, composite maize samples from four districts namely; Mityana, Jinja, Kamuli and Lira had concentrations of mean total aflatoxins above the regulatory limit [10ppb] set by Uganda National Bureau of Standards (UNBS). This finding presents a significant public health concern considering the toxic potential of aflatoxins. Strikingly, 100 % of contaminated samples from Mityana had concentrations of total aflatoxins above 10 ppb, followed by Jinja (80%) with 67 % of maize samples from Lira and Kamuli districts having aflatoxin levels above 10 ppb. On the contrary, positive maize samples from Kampala, Kasese, Masindi and Pader had majority of the samples with total aflatoxin content below the UNBS regulatory limit (Figure 3). These findings suggest that across regions in Uganda, consumption of posho might present a significant public health concern bearing in mind that posho is a staple food for majority of Ugandans.

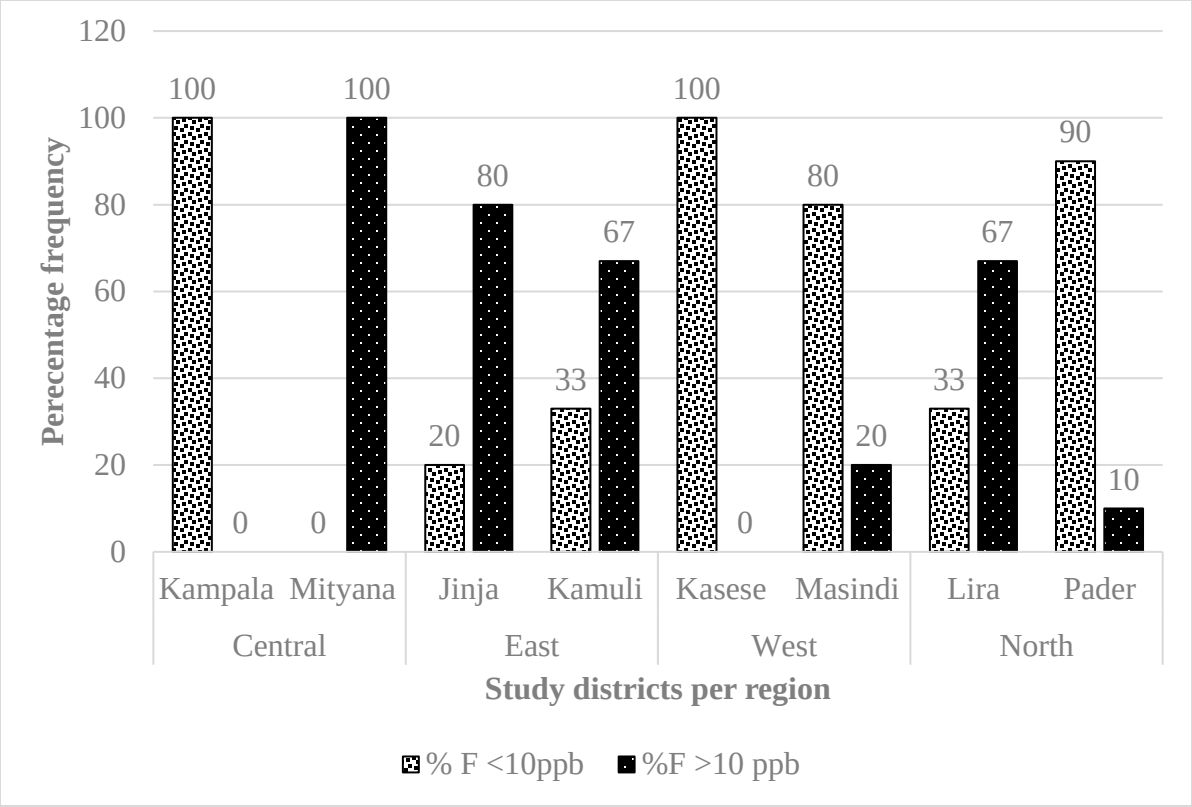

Figure 3. Total aflatoxin levels in maize samples from eight districts of Uganda with levels below and above UNBS regulatory limits (10ppb)

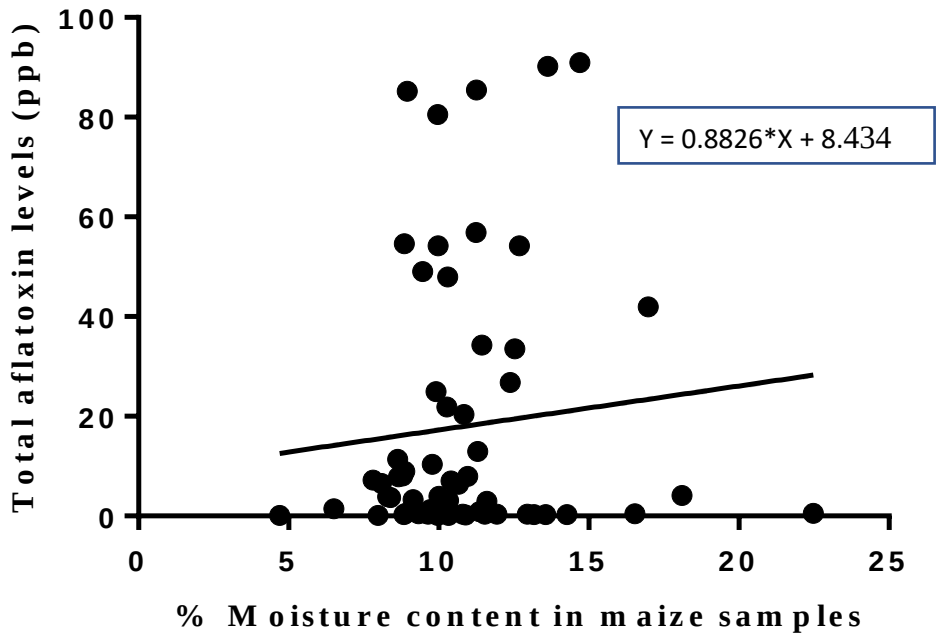

Figure 4: Moisture content against total aflatoxin levels in maize samples from eight districts of Uganda

Relationship between mean percentage moisture and mean total aflatoxin levels (ppb) in maize samples. In this study, it was hypothesized that the mean percentage moisture content levels do not influence mean total aflatoxin concentrations of maize samples analyzed. However, a regression analysis revealed a very weak correlation between the two variables (p=0.474; R^2=0.0089). The high p value obtained in this study at significance level 0.05 indicates that there was no relationship between moisture content and total aflatoxins of tested maize samples. In addition, the R-squared value indicates that only 0.89 % of the variation in mean total aflatoxin levels can be attributed to changes in mean percentage moisture content of maize samples analyzed in this study (Figure 4). Thus, the null hypothesis was accepted and concluded that variations in mean percentage moisture content did not influence changes in mean total aflatoxins of maize samples. This finding suggests that other factors, besides moisture content, could have influenced the variations in mean total aflatoxin concentrations of maize samples obtained in this study.

DISCUSSION

This study has revealed that ineffective methods for assessment of level of dryness of maize samples were used. In addition, drying of maize at milling centers depended on sunshine. Notably, moldy contaminated maize was either processes for human or animal consumption. Thus, the findings collaborate with findings of previous studies. It was reported that poor practices of maize in Sub-Saharan Africa remain a major contributor to losses incurred during post-harvest handling (Grace *et al*., 2018; Akumu *et al*., 2020). These findings combined allude to an increasing risk to aflatoxin contamination and human exposure. The results therefore call for an increased adoption of improved post- harvest handling practices to minimize the risk of aflatoxin contamination of maize in Uganda. This approach will not only safeguard the public from dangers of aflatoxins but also improve productivity and economic gains from this vital crop.

In addition, 50.4 % of composite maize samples screened were positive for aflatoxins with aflatoxin G being the dominant aflatoxin group. This finding contradicts with what has been previously reported that Aflatoxin B group is the most dominant group (Zhang and Banerjee, 2020). It has been documented previously that all known aflatoxigenic fungi produce aflatoxin B making it the dominant group (Coppock *et al*., 2018). The dominance of aflatoxin G in maize samples analyzed in this study could be attributed to climatic changes that are likely to cause genetic polymorphisms among aflatoxigenic fungi favoring production of aflatoxin G (Moore *et al*., 2017; Okoth *et al*., 2018).

The mean total aflatoxin content of maize samples analyzed in this study varied greatly across regions with samples from Masindi in Southwestern Uganda having the highest mean total aflatoxin concentration of 54.18 + 0.0 ppb. This finding implies that there is an increasing trend of aflatoxins levels in maize in Uganda since previous studies reported lower levels. For example, Akumu *et al.* (2020) reported a mean total aflatoxin concentration of 45.82 + 20.88 ppb in maize samples from eastern Uganda. In a study done in South Western Uganda by Murokore *et al*. (2023), a mean total aflatoxin content of 34.1 + 14.1 ppb in maize samples analyzed was reported. In contrast, a mean total aflatoxin concentration of 126.4 ppb in maize was obtained in a study conducted in Eastern and Northern Uganda (Akullo *et al*., 2023). The regional increases in mean total aflatoxin levels allude to an increasing trend in poor post-harvest handling practices of maize in Uganda. In this study, it was revealed that majority of the mill owners used ineffective traditional methods such as use

of biting test as a method of assessing degree of dryness, use of bare grounds as a drying platform and absence of use of solar dryers. These factors analyzed could partly explain the increasing aflatoxin contamination trend observed in this study.

Despite low moisture content levels of maize samples obtained in this study, significant concentrations of total aflatoxins were found in these samples. This finding contradicts with those of previous studies in which it was reported that water content (< 12.5 %) does not support growth of aflatoxigenic fungi and hence aflatoxin production (Jaibangyang *et al*., 2021; Abel Palacios *et al*., 2022; Wang *et al*., 2022; Marín *et al*., 2024). This finding suggests that maize contamination with aflatoxigenic fungi could have happened during the initial stages of the drying process since the moisture content levels of maize samples analyzed in this study were below 12.5 %. This finding indicates that there is a need for an effective method of drying of maize grains such as use of solar dryers. In agreement, this study revealed that none of the respondents reported use of solar dryers across the four regions of Uganda.

Limitations of the study. This study only analyzed 119 maize samples from selected hammer-mill centers in regionally distributed eight districts of Uganda. To fully understand the magnitude of aflatoxin occurrence in maize, a wider study should have been done to take into account variations in post-harvest handling practices under different cultural settings. However, in this study, it was assumed that composite samples from milling centers were representative of the samples from various culture settings. This is because in Uganda, processing of maize cereals into flour can only be done using a hammer mill and therefore, maize grains from different cultural settings and /or farmers converge at these milling centers.

CONCLUSION

The findings of this study highlight the need for regular monitoring of aflatoxin levels in maize grains and maize flour, use of proper post - harvest handling practices. In addition, these findings underscore the need for a collective action from all players to protect the public from dangers associated with consumption of aflatoxin contaminated maize and its products.

What is known about this topic?

Previous studies have demonstrated presence of aflatoxins in maize in Uganda and in levels above national regulatory limit. These studies further demonstrated an increasing trend in aflatoxin contamination of foodstuffs including maize and therefore called for regular monitoring of aflatoxin status of this staple food matrix.

What the study adds

This study has demonstrated an increasing trend in aflatoxin contamination of maize in Uganda. Unlike the findings of the majority of previous studies that reported the dominance of Aflatoxin B in food matrices, this study has revealed that Aflatoxin G was the major contaminant. The results of this study further demonstrate the dire need to urgently control aflatoxin contamination in maize in Uganda.

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DECLARATION OF CONFLICT OF INTEREST

The authors declare that they have no conflict of interest in the paper.

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