Journal of Agriculture and Food Sciences
Volume 20, Number 2, October 2022 pp. 31-61

PREVALENCE OF AFLATOXIN B1 IN MARKETED HERBS AND SPICES IN NAIROBI COUNTY KENYA: CONSUMER AWARENESS, PRACTICES, AND ASSOCIATED HEALTH RISKS

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ABSTRACT

We conducted a study in Nairobi County to assess Aflatoxin B1 (AFB1) levels in selected herbs and spices and to ascertain product use and consumer awareness of fungi and mycotoxin contamination. Descriptive statistical analysis of the survey data was performed using R-studio software version 1.4. Results revealed significant differences in consumers' preference for herbs and spices (P= 0.03671). Products consumed in less than one month were significant in predicting preference by customers (P= 0.0207). 65% of the respondents were unaware of mycotoxins in herbs and spices, and 80% had never attended mycotoxin training. Enzyme-linked immunosorbent assay (ELISA) method was used to analyze the samples for AFB1 contamination. Aflatoxin B1 was found in all of the samples and the concentration in spices ranged from 2.17ng/kgto 32.40ng/kg while in herbs the range was from 6.60ng/kg to 27.20 ng/kg.Considering the consumption data collected from our survey, the AFB1 daily intake for spices ranged between 0.83 and 11.78 ng/kg bw/day and in herbs it was between 0.6 and 6.88 ng/kg bw/day. The results indicate a high possible risk of exposure to mycotoxins; thus, more stringent measures must be taken for the prevention of mycobiota contamination in herbs and spices.

Keywords: Herbs, Spices, Fungi, Aflatoxins, Consumer Awareness

https://dx.doi.org/10.4314/jafs.v20i2.3

INTRODUCTION

Food safety has gained relevance among consumers and authorities globally. The purpose is to keep consumers safe from compounds that could be harmful to their health and ensure producers do not suffer massive economic losses due to contamination outbreaks in the production chain. Based on food safety authorities' risk evaluation reports, companies and legal bodies have taken various recommendations and measures to ensure risk exposure levels to harmful compounds in food and feed are kept at minimal levels (Alshannaq & Yu, 2017). The existing regulations are based on scientific opinions from institutions such as the European Food Safety Authority

(EFSA) and the FAO/WHO Joint Expert Committee on Food Additives of the United Nations (WHO/FAO 2018). Most risk mitigation studies aim to determine possible human exposure to contaminants in food (Moretti et al., 2017). Such a form of evaluation often requires long-term monitoring of the frequency of occurrence and contamination levels of targeted harmful compounds in food. According to the FAO report, mycotoxins contaminate approximately 25% of the world's agricultural products (Bhat, 1999). However, following the release of extensive data on mycotoxin prevalence in food crops, with higher levels than 25% (Kovalsky et al., 2016; Streit, Naehrer, Rodrigues & Schatzmayr, 2013); there is a need for a more detailed investigation of mycotoxin contamination rates at the global level. Mycotoxin producing fungi are ubiquitous and can infect grains at all levels from production to processing and the supply chains (Reddy et al., 2013). A significant level of infection is encouraged by improper post-harvest handling, transporting, drying and storage (Tiffany, 2013).

Physical, chemical, or biological detoxification strategies have been employed to eliminate mycotoxins in food products, with the need for biotechnological advancement gaining interest among researchers (Moretti, Logrieco and Susca, 2017).Herbs and spices are prone to fungal contamination, including *Aspergillus, Penicillium, Fusarium,* and *Alternaria* species (Hammami et al., 2014; Kong et al., 2014). These fungi contaminate herbs and spices either in the field, e.g., *Fusarium* spp, or during storage, e.g., *Aspergillus* and *Penicillium* spp. They produce mycotoxins, whose effects on human health are a primary global concern. The most common mycotoxins produced by field and storage fungi include aflatoxins, ochratoxins, fumonisins, trichothecenes, zearalenone, patulin, and citrinin (Alassane-Kpembi et al., 2017). Mycotoxin contamination often results in mycotoxicoses in humans and animals expressed in the form of allergic reactions, metabolic, biochemical, and reproductive deficiencies, fetal alterations, immune diseases, and death in cases of chronic exposure (Singh and Cotty, 2017). The main food groups affected by fungal metabolites are cereals, dried fruits, nuts, coffee, and spices (Marin, Ramos, Cano-Sancho & Sanchis. 2013). Many *Aspergillus* species such as *A. flavus* and *A. nomius* have frequently been isolated from herbs and spices (Azzoune et al., 2016; Hell, Gnonlonfin, Kodjogbe, Lamboni & Abdourhamane, 2009) and are prone to producing aflatoxins such as AFB1, AFB2, AFGI, and AFG2. These mycotoxins, especially AFB1, are carcinogenic (group 1), immunosuppressive and mutagenic (Okoth and Kola, 2012). Ochratoxins are cytotoxic to the lymphocytes, nephrotoxic, carcinogenic, teratogenic and suppresses the functions of monocytes and granulocytes (Darwish, Ikenaka, Nakayama and Ishizuka, 2014). As a result, Ochratoxin has been classified as a group 2B carcinogen, with the kidney being its main target organ (IARC, 2012).

Human beings can be exposed to mycotoxin by consuming contaminated herbs and spices (Azzoune et al., 2016). Exposure to these toxins is predominant in developing nations with poor methods for controlling pest infestation in crops poor transportation, storage, and processing facilities (Mutegi, Wagacha & Kimani, 2013). However, it can also be rampant in societies with a high level of ignorance, where people assume herbs and spices are not prone to fungal contamination. Constant exposure to mycotoxins is common where rules and laws that protect the food intake of the populace are not adhered to (Moturi, 2008). In developed countries, especially in subgroups where poor handling and storage of culinary herbs and spices are common, there can be a risk of exposure to mycotoxins, making the populace vulnerable to mycotoxin-induced infections. However, because mycotoxins are natural contaminants of plant products (Singh and Cotty, 2017) and with the nutritious nature of herbs and spices, their susceptibility to mycotoxigenic fungi contamination is inevitable.

In Kenya, adherence to production standards regarding permissible levels of mycotoxins in food products is rare; therefore, there is the likelihood of human exposure to unsafe levels of various mycotoxins (e.g., aflatoxins) in herbs and spices (Mwangi, William, Nguta and Muriuki, 2014). Information regarding levels and occurrence of mycotoxins in spices and herbs sold on the Kenyan market is lacking; hence the resulting public health burden has also been overlooked. This study assessed consumer knowledge regarding fungal and mycotoxin contamination of herbs and spices. During the survey that took place between January 2021-March 2021, a structured questionnaire was administered with the primary aim of retrieving information from participants that could assist in (i) determining the frequency and use of herbs and spices (ii) assessing consumer-based knowledge regarding fungal and mycotoxin contamination of the spices used and (iii) evaluating storage and handling practices of the purchased spices and herbs. Prevalence of AFB1 in marketed herbs and spices in Nairobi was also assessed.

Materials and Methods

Study Area

The study was carried out in Nairobi County. Nairobi county is the central capital city of Kenya, situated at 1° 09'S 3639' and 1°27'S 3706'E at an altitude of 1600m above sea level (LatLong, 2022). It receives a mean annual rainfall of 925mm and mean annual minimum and maximum temperature of 12 and 28°C, respectively(Lisa van Aardenne, 2017). The city has an estimated 4.5 million people living within 696km^2(CAHF, 2020). Nairobi is home to thousands of Kenyan businesses and a cosmopolitan center contributing to the numerous elements of cultural diversity, including cuisines.

Household Survey

In December 2020, a reconnaissance study was carried out using purposive sampling to select participants who frequently bought and used spices and herbs in their households. With a target of 500 respondents for the study, a total of 385 eligible individuals completed the survey (a response rate of 77%), which is above the 50% set target for survey studies (Crowther & Lancaster, 2009). The high response rate was attributed to high motivation among consumers of herbs and spices in completing the survey. The reconnaissance study carried out in December 2020 aimed at household identification also contributed to the high response rate. The interviewees had prior information and were therefore aware of the study.

Herbs and spices sampling

A sample representation of 207 herbs and spices were purchased from open and local supermarkets for AFB1 analysis. Approximately 100 g of spice and herb samples were purchased from different sellers,i.e., open market, supermarket, and imported brands within Nairobi County. The collected samples were taken to the mycology lab at the University of Nairobi ChiromoCampus in conditions under which they were sold (sealed plastics) and dried in the oven at 50°C for three days. Before AFB1 analysis, the dried samples were ground in a blender, and final products were passed through a 500um sieve. A list of the herb and spice samples used in this study is provided in Table 7 of the supplementary data.

AFB1 determination

Aflatoxin B1 content in the spice and herb samples was determined using a quantitative Enzyme-Linked Immunosorbent Assay(ELISA) kit for aflatoxin B1 in corn, rice, soybeans, wheat, and spices from AgraQuant® Romer Labs (Product no: COKAQ8000/COKAQ8048). The extraction process was performed as per the manufacturer's protocol. For each sample, 20 g of ground sample was added into tightly sealed clean jars, and 100 ml of 70/30 (v/v) methanol (LC grade, Merck, Darmstadt, Germany)/water extraction solution was added. The mixture was then homogenized at 240 r/s for 3 min using an orbital shaker (Digisystem Laboratory Instruments Inc. Germany). The sample was allowed to settle, and the top layer was filtered through Whatman #1 filter to collect the filtrate. The filtrate was diluted (1:2) with assay buffer by adding 50ul of the filtrate to 100ul of buffer and mixed thoroughly. The supernatant obtained was used for the Competitive Direct Enzyme-Linked Immunosorbent Assay (CD-ELISA). The AFB1 content was inversely proportional to the color intensity established using an automated microplate reader at 450 nm. AFB1 content in each sample was calculated based on a calibration curve plotted in each experiment using the AFB1 standard at different concentrations (0, 2, 5, 20 and 50 ppb). The correlation coefficients were above 0.99. According to the supplier's information, the lower and upper limits of quantification of the kit were 2 and 50 ppb (µg/kg), respectively. Samples with toxin values below the limit of quantification (LOQ) were recorded as containing no detectable toxin.

Assessment of possible exposure to AFB1 in the screened spices and herb samples

To assess exposure to AFB1, a deterministic risk analysis as described by Coronel, Marin, Cano, Ramos and Sanchis (2011) was performed. An estimate of AFB1 daily intake (ng OTA/kg bw/ day) was done by multiplying spice/herb AFB1 concentration (ng/g) by individual consumption data (g/kg bw/day) obtained from the household survey. Based on studies by Assimitti, (2020) and Carlsen, Blomhoff, and Andersen, (2011), people in the age group 15-65 represent the highest frequency in using herbs and spices on a daily basis. Calculations were thus done assuming an average population's body weight of 60 kg with reference to a study by Nguegwouo et al., 2018.

Data analysis

Descriptive statistics (frequencies and percentages) of the data generated from the household survey was performed using R-studio software version 1.4. Pearson's chi-square test was performed to determine significant relationships between product type, form, and market source for both herbs and spices. Binary logistic regression analysis was used to examine the co-relationship between product preference and duration of use. Statistical significance was set at 0.05. The p-value of less than 0.05 was considered significant. The AFB1 content in the different spices and herbs was expressed as mean ± standard deviation, and Statistical software SAS version 9was used to perform a one-way ANOVA to establish significant differences between the concentration levels obtained ($p \leq 0.05$). This software was also helpful in generating min, max, and mean AFB1 daily intake per sample.

RESULTS AND DISCUSSION

Household survey on consumer use and practices

The majority of the respondents were females (59%) between 25-50 years of age (Table 1). A similar observation was made in studies by Isbill, Kandiah and Khubchandani, (2018) and Nguegwouo et al. (2018). People aged 25-60 years are commonly associated with frequent use of herbs and spices (Assimitti, 2020). Contrary to the findings in this study, reports by Chapman-Novakofski (2016) and Heinrichs, Nikolaus, Ellison, and Nickols-Richardson (2014) shows a higher liking for spicy flavors among men compared to women and such disparities concerning culinary herb and spice preferences are influenced by race, personal, interpersonal, and environmental factors(Mwangi et al., 2014).

In most instances, food choice and preference are related to age (Dean, Raats, Grunert, Lumbers & The Food in Later Life Team 2009). The frequency of using herbs and spices for culinary purposes has been reported to decline with age (Wang & Worsley, 2014). The findings in the current study (Table 1) also indicate a decline in the preference rate for herbs and spices with age. The majority were young adults and middle-aged (94%), while only 6% of the entire target population was aged 55 years and above. Most of the respondents were single (61%), 36% were married, and only 3% were separated. The composition and family set up in a household influences taste preference, especially when using spices as food additives (Laroche, Hofer & Davis, 2007).

The education levels ranged from primary (1%), high school (14%), and tertiary (82%), with only (3%) not having an academic background. The employed (41%) and self-employed (35%) respondents ranked highest in terms of response compared to the unemployed (12%) (Table 1).It is, therefore, evident that people from lower socioeconomic position (SEP) backgrounds (as indicated by education and income) tend to have less preference and interest in culinary herbs and spices and this is a reflection of awareness and affordability. A similar trend was observed in the Nguegwouo et al. (2018) sample population. In the study by Isbill et al. (2018), the majority of the respondents had attained only high school education (58%) compared to a 42% incidence rate of those with tertiary schooling. Bower, Marquez and de Mejia (2016) report indicate that those with higher income and higher education levels have a greater propensity to use spices and herbs. In the current study, the level of education had a positive impact on the knowledge regarding fungi and mycotoxin contamination in herbs and spices. This observation is in line with studies by Dosman, Adamowicz and Hrudey, (2001), whose findings indicate that individuals with a higher level of education are more aware of fungal contamination in food than those with less education.

Use and storage of herbs and spices

Most herbs and spices were consumed on an average basis, i.e., at least twice a week (51%), while 29% of consumers used the products highly i.e., thrice a week (Table 2). These statistics reflect consumers' increased demand for the products, attributed to increased consumer awareness regarding the role of herbs and spices in food flavoring, nutrition, and medicinal purposes (Smiechowska, Newerli-Guz & Skotnicka, 2021). The farming and trading of herbs and spices in Kenya are also rising (Farmbiz Africa, 2019), culminating in increased availability and use locally and internationally. Most consumers use culinary herbs and spices for food preparation (65%), while only a small fraction (12%) of the respondents reported their use for medicinal purposes (Table 2). Similarly, in the studies of Nguegwouo et al. (2018), respondents attributed white pepper, black pepper, and cloves mainly for food preparation compared to medicinal purposes.

During purchase, the most preferred form of packaging was plastic containers (32%), which consumers attributed to their availability on the market. However, it is interesting to note that most of the respondents (45%) did not base their product preference on the packaging of the culinary herbs and spices; instead, their product choice was determined by the prices. Therefore price comparison dominates consumer purchase preference, unlike packaging and branding (Tariq, Nawaz, Nawaz3 and Butt, 2013). As seen in Table 2, 68% of the respondents admitted that they do not check the expiry dates of the herbs and spices before purchase. This trend contributes to the use of compromised and hence contaminated products. In our study, we also recorded a 41% incidence (Table 2) where consumers stored their herbs and spices in open containers. Exposure of nutritious products such as herbs and spices to air creates room for mycobiota contamination, most of which are mycotoxigenic, contributing to mycotoxin contamination (Tančinová, Mokrý, Barboráková & Mašková, 2014).

Product preference, market source preference, and average quantities used per meal

In the current study, the majority of the households used spices (81%) compared to herbs (29%) (Table 3). As noted in Table 2, most culinary products are preferred for food flavoring, and since spices are aromatic, their preference automatically outweighs herbs. 29 out of 53 respondents preferred to use herbs in fresh form, while 135 out of 219 consumers preferred spices in their dry form (Table 3). The same preference for spicy flavors was reported by Törnwall et al. (2014). Table 1 and 2 of the supplementary data gives a detailed breakdown of the preferred form for the specific herbs and spices documented in the current study. In Kenyan markets, herbs and spices are sold in dried or fresh forms. Drying has been the most preferred form of product preservation from ancient days as it extends the shelf life of food products by reducing susceptibility to microbial contamination through lowering moisture contents (FDA, 2017).

Supermarkets were the primary source (52%) for culinary herbs and spices. In comparison, 46% of the respondents sourced their products from local/open markets, with only 6% of the consumers using imported products (Table 3). The reliance on supermarket products was attributed to the variety of herbs and spices at customers' disposal for preferred product choices. When probed further, consumers from the open market found the products affordable and claimed authenticity in aroma compared to the imported and supermarket ones. Contrary to the findings in the current study, Fritts et al. (2019) and Muhammad, Jibirb, Hamzab and Garba, (2020) reported a higher incidence in the use of spices from open markets than the supermarket ones by consumers. Results from the duration of product use, (Table 3) revealed that most spices were consumed in less than a month (48%) while herbs were consumed within the first six months (40%) upon purchase. This indicates that spices were consumed frequently compared to herbs. However, 9% and 13% of the responses acknowledged the quantity purchased and frequency of use as the main determinants for the duration of use of spices and herbs (Tables 3 and 4 of the supplementary data).

The current study indicates that both herbs (37%) and spices (40%) are used in quantities less than 5g for meal preparations (Table 3). Cayenne, chili, nutmeg, and white pepper were used in low quantities for meal preparation. In probing further, most consumers attributed the spicy nature of pepper and chilies to low use. At the same time, the acrid aroma in nutmeg makes it unpalatable, and thus preference use is in low quantities. A similar trend was observed by Koleosho, 2014, Muhammad et al., 2020 and Nguegwouo et al., 2018.While using herbs and spices in meal preparation, 31% and 27% (Table 3) of the respondents admitted that they did not measure the quantities used. It is essential to acknowledge that frequency of use combined with the amount of seasoning used in meal preparations is the main factor for risk assessment when determining possible exposure to mycotoxins in food products (WHO/FAO, 2018). With such information, an evaluation of health risks associated with mycotoxin contamination is feasible. A breakdown of estimated quantities of each spice and herb used in meal preparation has been provided in tables 5 and 6 of supplementary data.

Herb and spice preference

In the current study, garlic, ginger, black peppers were the most used while cayenne, red chili, and white pepper spice products were least used. For herb products, mint, rosemary, and oregano were the most used (Table 4). Ginger and garlic were used in large quantities because they are the most popular, readily available, and affordable in Kenyan markets (Mwangi et al., 2014). Mint and rosemary were the most preferred herbs because they have a variety of uses (food and health), and they easily complement spices for flavor and aroma (Okoye, 2018). Findings in this study establish a relationship between awareness and preference in the sense that consumers prefer spices and herbs that they are most aware of (Mulderij, 2017).

Knowledge of fungi and mycotoxin contamination

The majorities of respondents in the current study are aware of fungal contamination (83%) in food products and know the health risks (73%) associated with such contaminations. In relation to fungal contamination in herbs and spices, 72% of the study population admitted that they were unaware if herbs and spices are also susceptible to fungal contamination, and 70% of the respondents had never experienced fungal contamination in their purchased culinary products (Table 5). With reference to fungal contamination in food, 60% of the respondents admitted they could easily recognize fungi in culinary herbs and spices. 67% of the respondents were aware of mycotoxin contamination in different food products, while only 35% acknowledged that herbs and spices are also prone to toxin contamination (Table 5). Public knowledge of health risks associated with mycotoxicoses was averagely sufficient (66%). Similarly, in the findings of Magembe, Mwatawala, Mamiro and Chingonikaya (2016), majority of the respondents were not aware of the health risks associated with mycotoxins. In the current study, 80% of the respondents (Table 5) have never attended any training on mycotoxin contaminations in food thus reflecting a knowledge gap regarding mycotoxin awareness and its impact on food security and human health. The willingness expressed by 84% of the respondents to attend training on mycotoxin contamination in food is worth noting. Public awareness training often drives transformation in attitudes. When target groups express confidence in the lessons learned and apprehend the problems well enough, they can be persuaded to drop old habits and practices (Adekoya, Obadina, Phoku, Nwinyi & Njobeh, 2017).

Aflatoxin B1 contamination and exposure levels

AFB1 screening in herbs and spices

The AFB1 content of culinary herbs and spices is summarized in Tables 6 and 7. AFB1 was detected in all samples. Among the samples, 11 (79%) of the 14 spices and all(100%) the herb samples had AFB1 levels above the set regulatory limit of 5μg/kg by the European Commission(2006). In culinary spices, cloves and cinnamon had the highest mean concentration levels of AFB1, i.e., 32.40 ±18.01 SD max: 50μg /kg and 21.20 ± 22.46 max: 47.1 SD μg/kg respectively while among the herbs sage and mint had the highest mean concentration levels of AFB1, i.e., 27.20 ± 5.23 SD max: 30.9μg /kg and 23.50 ± 2.12 SD max: 25 μg/kg respectively. Paprika, coriander, and garlic had the lowest AFB1 levels among the spice samples. The low levels of AFB1 contamination in garlic samples may be attributed to inhibiting *Aspergillus niger* and *A. Flavus* growth by its indigenous antifungal activity (Ismaiel, Gama, Saied, Marwa & Abd EL-Aal, 2012). A significant difference in AFB1 content in the sampled spices was observed among cloves and the other spices apart from cinnamon (Table 6). There was also a significant difference in AFB1 mean concentrations between sage and the rest of the herb samples (Table 7). There is a continuous growth in the popularity of organic foods globally (FiBL, 2020). Thus the presence of high levels of AFB1 in the sampled organic herbs and spice samples should be a matter of interest and concern among researchers and the government. Several reasons instigate consumers' purchase of herbs and spices: perceived benefits to the environment and human health and the nutritional benefits (Törnwall et al., 2014). Several other studies have also reported prevalence of AFB1 in herbs and spices with different factors such as post-harvest handling and mycotoxin analytical techniques contribute to the varied levels of AFB1 (Khazaeli, Mehrabani, Heidari, Asadikaram & Najafi, 2017; Ozbey & Kabak, 2012; Torsun & Arslan 2013).

Dietary exposure to AFB1

The daily intake for AFB1 was highest in cinnamon with a maximum estimate of 11.775 ng AFB1/kg bw/day, followed by ginger and cloves, both having maximum estimates of 5.533 ng AFB1/kg bw/day. The most negligible intake was white pepper and garlic, whose maximum estimates were 1.067 and 0.833 ng AFB1/kg bw/day, respectively (Table 8). For herb samples, daily intake for AFB1 was highest in mint and rosemary, whose daily intakes were 6.875 and 4.45 ng AFB1/kg bw/day, respectively. The least daily intake was in thyme and basil (1.192 and 0.6 ng AFB1/kg bw/day, respectively) (Table 9). Ginger, mint, and rosemary were the most preferred culinary products by consumers in the current studies (Table 4) and are also among the samples with high AFB1 daily intake levels. This implies that consumers are at greater risk of exposure to aflatoxin-related illnesses. Among the storage options cited by customers, most of them store their spices in open plastic containers, which allow moisture into the spices creating a conducive environment for mycotoxigenic fungi to thrive (Ozbey & Kabak, 2012). To prevent mycotoxin exposure risks, there is a need to focus on post-purchase handling of the spices on the shelf or in storage (Passamani, et al., 2014). In the case of our studies and with reference to consumer practices, simple actions such as storing spices and herbs in air-tight containers and observing proper handling can help limit contamination.

CONCLUSION

Aflatoxin exposure is high in Kenya. The erratic tropical climate characterized by high humidity, periodic droughts, and high temperatures significantly contributes to spoilage fungi thriving in food products (Mutegi et al., 2013). Drying of grain on bare grounds, poor grain conditioning before storage, poor storage structures, use of propylene storage bags, poor transportation and handling of produce, insect infestation, and chronic poverty have been incriminated for the aflatoxigenic contamination of Kenyan foods (Koskei, Bii, Musotsi & Karanja, 2020).

Herbs and spices are mainly used for food preparation, with ginger, garlic, and black pepper being the most consumed spices, while rosemary and mint topped the preference list for herbs as per the current study. Cultural attributes, community setups, and dominating cuisines were the determinant factors for herb and spice preference among residents of Nairobi County. Owing to the vulnerability of herbs and spices to spoilage by storage fungi (Darwish et al., 2014) possibility of mycotoxin contamination is imminent. Risks of mycotoxin contamination with more devastating effects are associated with low-level chronic intake other than one-time level intake (Nguegwouo et al., 2018). On average, herbs and spices are consumed at least thrice a week, indicating the high frequency of use and thus increased demand among consumers. Therefore, the use of herbs and spices, even in low amounts, is a predisposing factor to mycotoxicoses.

Herein AFB1 analysis in selected herbs and spices and a comprehensive overview of consumer awareness concerning mycotoxin contamination of culinary herbs and spices is reported. All the samples in the current study were AFB1 contaminated, thus calling on the urgent need for the government and research institutions to channel more resources on studies aimed at regulating mycotoxins in culinary products. Considering the maximum limits of AFB1 content in spices and herbs by the European Commission and the concentrations recovered from the analyzed samples coupled with the amount consumed daily, there is an increased health risk directly associated with AFB1 contamination in spices and herbs sold on Kenyan markets. This study revealed the feasibility of human exposure following poor perception of mycotoxin contamination among consumers of herbs and spices. Therefore, efforts should be made to enhance consumers' awareness of mycotoxins and their associated impact on health and enable them to adopt mycotoxins preventive measures.

Storage of purchased herbs and spices in open containers is a common practice, thus exposing them to moisture and subsequent fungal contamination. Also, the awareness of fungal and mycotoxin contamination in herbs and spices was generally low. Consequently, interventions to limit mycotoxins in herbs and spices from harvest to storage should be prioritized. Based on the evidence of the data available in this study, frequent training on mycotoxin contamination of herbs and spices need to be a common practice with more efforts need to be directed towards awareness and educational intervention among producers and consumers of herbs and spices as much interest and attention has only been given to cereals and nuts. In the meantime, hygienic post-harvest handling practices are suggested in addition to routine surveillance for mycotoxins. These recommendations are crucial to improving human capacity in producing and processing safer foods for a healthier nation.

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APPENDICES

Table 1: Socio-demographics

Socio-demographic variables				
Parameters			**Incidence (%)**	
Gender			Male(19-60 years), N=158	Female(18-57 years), N= 227
			41%	59%
Age range(years)				
18-24			38(24.1%)	63(27.8%)
25-54			107(67.7%)	155(68.2%)
≥55			13(8.2%)	9(4%)
Level of education		N= 385[1]		
	None	12 (3.1%)	3 (1.9%)	9 (4.0%)
	Primary	4 (1.0%)	2 (1.3%)	2 (0.9%)
	Secondary	52 (14%)	14 (8.9%)	38 (17%)
	Tertiary	317 (82%)	139 (88%)	178 (78%)
Marital status	Single	234 (61%)	87 (55%)	147 (65%)
	Married	138 (36%)	65 (41%)	73 (32%)
	Separated	13 (3.4%)	6 (3.8%)	7 (3.1%)
Main activity	Employed	156 (41%)	96 (61%)	60 (26%)
	Self employed	136 (35%)	56 (35%)	80 (35%)
	Housekeeper	47 (12%)	26(16%)	21 (9%)
	Unemployed	46 (12%)	18 (11%)	28 (12%)

Table 2: Purchase, use, and storage of herbs and spices

Consumption frequency[a]	Incidence % N= 385
Highly	113 (29%)
Averagely	198 (51%)
Rarely	74 (19%)
Product usage	
Food preparation	250 (65%)
Medicinal purpose	47 (12%)
Both food and medicinal purpose	88 (23%)
The preferred form of packaging during purchase	
Glass containers	63 (16%)
Jute bags	21 (5.5%)
Plastic containers	123 (32%)
Others	5 (1.3%)
No preference	173 (45%)
Checking for expiry date	
Yes	124 (32%)
No	261 (68%)
The preferred form of storage	
Airtight containers	163 (42%)
Open containers	156 (41%)
Plastic papers	25 (6.5%)
No preference	41(11%)

[a]Rarely (At most once a week); Averagely (twice a week); Highly (At least thrice a week)

Table 3: Product and market source preference, storage period, and average quantities used per meal

Parameter	Incidence %		
		Product form	
	Overall, N = 272[1]	Dry, N = 159[1]	Fresh, N = 113[1]
Form Preference			
Herbs	53 (19%)	24 (15%)	29 (26%)
Spices	219 (81%)	135 (85%)	84 (74%)
Source preference			
	Overall, N = 980[1]	Herbs, N = 238[1]	Spices, N = 742[1]
Imported/sold in original packaging	6 (0.6%)	0 (0%)	6 (0.8%)
Open/ Local Market	449 (46%)	110 (46%)	339 (46%)
Supermarket	511 (52%)	123 (52%)	388 (52%)
Others(gifting,borrowing)	14 (1.4%)	5 (2.1%)	9 (1.2%)
Product Storage			
	Herbs, N = 238[1]	Spices, N = 742[1]	
<1 month	91 (38%)	354 (48%)	
1-6 months	95 (40%)	269 (36%)	
6-12 months	16 (6.7%)	42 (5.7%)	
>1 year	4 (1.7%)	10 (1.3%)	
It depends on quantity purchased and frequency of use	32 (13%)	67 (9.0%)	
Product use			
	Herbs, N = 238[1]	Spices, N = 742[1]	
<5g	88 (37%)	297 (40%)	
5g	62 (26%)	205 (28%)	
>5g	14 (5.9%)	35 (4.7%)	
Not measured	74 (31%)	204 (27%)	

Table 4: Herb and spice preference

Variable				Variable			
Herb	**Preference =53[1]**	**Count**	**N**	**Spice**	**Preference =219[1]**	**Count**	**N**
Basil	6 (11%)			Black pepper	19 (8.7%)		
Bay	5 (9.4%)			Cardamom	8 (3.7%)		
Mint	16 (30%)			Cayenne	2 (0.9%)		
Oregano	8 (15%)			Red chilli	4 (1.8%)		
Parsley	2 (3.8%)			Cinnamon	13 (5.9%)		
Rosemary	13 (25%)			Cloves	7 (3.2%)		
Thyme	3 (5.7%)			Coriander	13 (5.9%)		
				Cumin	19 (8.7%)		
				Garlic	57 (26%)		
				Ginger	48 (22%)		
				Paprika	13 (5.9%)		
				Turmeric	12 (5.5%)		
				White pepper	4 (1.8%)		

[1]Preference (Frequency and Percentage (%))

Table 5: Knowledge of fungi and mycotoxin contamination

Parameter	Incidence %
Aware of fungal contamination in food	
Yes	321 (83%)
No	64 (17%)
Known of health risks associated with fungal contamination in food	
Yes	282 (73%)
No	103 (27%)
Aware of fungal contamination in herbs and spices	
Yes	109 (28%)
No	276 (72%)
Experienced fungal contamination in herbs and spices	
Yes	115 (30%)
No	270 (70%)
Ability to identify contaminated herbs and spices	
Yes	230 (60%)
No	155 (40%)
Aware of mycotoxin contamination in food	
Yes	257 (67%)
No	128 (33%)
Aware of mycotoxin contamination in herbs and spices	
Yes	134 (35%)
No	251 (65%)
Aware of health risks associated with mycotoxin contamination	
Yes	253 (66%)
No	132 (34%)
Common mycotoxins contaminating food	
Aflatoxins	286 (81%)
Fumonisins	30 (8.5%)
Ochratoxins	24 (6.8%)
Other	12 (3.4%)
Common mycotoxins contaminating herbs and spices	
Aflatoxins	116 (63%)
Fumonisins	34 (19%)
Ochratoxins	28 (15%)
Other	5 (2.7%)
Training on mycotoxin contamination of food products	
Yes	78 (20%)
No	307(80%)
Willingness to attend training on mycotoxin contamination	
Yes	325(84%)
No	60(16%)

Table 6: Aflatoxin B1 levels in spice samples

Spice	AFB1 levels (µg/kg)		
	Mean + SD*	Min	Max
Cloves	32.40 ±18.01[a]	14	50
Cinnamon	21.20 ± 22.46[ab]	7	47.1
Red chilli	15.10 ± 7.84[bc]	9.8	24.1
Cumin	12.30± 3.58[bc]	9.4	16.3
Cayenne	11.67± 1.69[bc]	10.5	13.6
Ginger	10.87± 5.32[bc]	6.1	16.6
Nutmeg	9.47± 4.31[bc]	5	13.6
Black pepper	8.57± 1.26[bc]	7.4	9.9
Turmeric	8.00± 1.71[bc]	6.6	9.9
Cardamon	6.07± 2.63[c]	4.4	9.1
White pepper	5.77± 0.65[c]	5.1	6.4
Paprika	4.77± 2.06[c]	2.6	6.7
Coriandar	4.37± 0.93[c]	3.3	5.0
Garlic	2.17± 0.29[c]	2	2.5
LSD ($p \leq 0.05$)	13.92		

*Mean values not followed by the same letters are significantly different ($p \leq 0.05$)

Table 7: Aflatoxin B1 levels in herb samples

Herb	AFB1 levels (µg/kg)		
	Mean + SD*	Min	Max
Sage	27.20± 5.23[a]	23.5	30.9
Mint	23.50± 2.12[ab]	22	25
Parsley	16.50± 1.84[bc]	15.2	17.8
Oregano	16.35± 3.61[bc]	13.8	18.9
Rosemary	15.00± 3.96[c]	12.2	17.8
Bay	14.15± 1.06[c]	13.4	14.9
Marjoram	13.10± 3.82[cd]	10.4	15.8
Thyme	12.05± 3.18[cd]	9.8	14.3
Basil	6.60± 0.85[d]	6	7.2
LSD ($p \leq 0.05$)	7.17		

*Mean values not followed by the same letters are significantly different ($p \leq 0.05$)

Table 8: Aflatoxin B1 daily intake in relation to spice consumption

Spice	Consumption(g/day)			AFB1 daily intake[a](ng/kg bw/day			AFB1 daily intake[b](ng/kg bw/day		
	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean
Cumin	0.017	0.167	0.075	0.205	2.05	0.917	0.272	2.717	1.215
Coriander	0.008	0.25	0.091	0.036	1.092	0.399	0.04	1.2	0.439
Paprika	0.008	0.25	0.084	0.04	1.192	0.399	0.056	1.675	0.561
Black pepper	0.008	0.25	0.097	0.071	2.142	0.827	0.083	2.475	0.956
Turmeric	0.017	0.25	0.112	0.133	2	0.893	0.165	2.475	1.106
Cayenne	0.008	0.167	0.078	0.097	1.944	0.914	0.113	2.267	1.065
Cinnamon	0.017	0.25	0.114	0.353	5.3	2.417	0.785	11.775	5.369
White pepper	0.008	0.167	0.06	0.048	0.961	0.343	0.053	1.067	0.381
Red chilli	0.008	0.167	0.046	0.126	2.517	0.690	0.201	4.017	1.102
Nutmeg	0.05	0.083	0.072	0.473	0.789	0.684	0.68	1.133	0.982
Cardamom	0.033	0.2	0.102	0.202	1.213	0.617	0.303	1.82	0.925
Ginger	0.017	0.333	0.13	0.181	3.622	1.413	0.277	5.533	2.158
Cloves	0.017	0.167	0.085	0.54	5.4	2.754	0.553	5.533	2.822
Garlic	0.017	0.333	0.115	0.036	0.722	0.249	0.042	0.833	0.288

Table 9: Aflatoxin B1 daily intake in relation to the quantity of spice consumed

Herb	Consumption(g/day)			AFB1 daily intake[a](ng/kg bw/day			AFB1 daily intake[b](ng/kg bw/day		
	Min	Max	Mean	Min	Max	Mean	Min	Max	Mean
Basil	0.05	0.083	0.078	0.33	0.55	0.517	0.36	0.6	0.564
Thyme	0.033	0.083	0.073	0.402	1.004	0.884	0.477	1.192	1.049
Mint	0.017	0.275	0.100	0.392	6.463	2.358	0.417	6.875	2.509
Bay	0.017	0.167	0.085	0.236	2.358	1.199	0.248	2.483	1.262
Oregano	0.017	0.167	0.065	0.273	2.725	1.066	0.315	3.15	1.232
Rosemary	0.017	0.25	0.089	0.25	3.75	1.324	0.297	4.45	1.571
Marjoram	0.033	0.083	0.067	0.437	1.092	0.873	0.527	1.317	1.053
Sage	0.033	0.083	0.054	0.907	2.267	1.473	1.03	2.575	1.674
Parsley	0.033	0.117	0.073	0.55	1.925	1.21	0.593	2.077	1.305

Supplementary data

Table 1: Form preference for herbs

Herb	Overall, N = 53[1]	Dry, N = 24[1]	Fresh, N = 29[1]
		Product form	
Basil	6 (11%)	3 (12%)	3 (10%)
Bay	5 (9.4%)	1 (4.2%)	4 (14%)
Marjoram	0 (0%)	0 (0%)	0 (0%)
Mint	16 (30%)	6 (25%)	10 (34%)
Oregano	8 (15%)	4 (17%)	4 (14%)
Parsley	2 (3.8%)	2 (8.3%)	0 (0%)
Rosemary	13 (25%)	7 (29%)	6 (21%)
Sage	0 (0%)	0 (0%)	0 (0%)
Thyme	3 (5.7%)	1 (4.2%)	2 (6.9%)

[1]Product form preference (Frequency and Percentage (%))

Table 2: Form preference for spices

Spices	Overall, N = 219[1]	Dry, N = 135[1]	Fresh, N = 84[1]
		Product form	
Black pepper	19 (8.7%)	10 (7.4%)	9 (11%)
Cardamom	8 (3.7%)	6 (4.4%)	2 (2.4%)
Cayenne	2 (0.9%)	2 (1.5%)	0 (0%)
Chilies/red chilli	4 (1.8%)	4 (3.0%)	0 (0%)
Cinnamon	13 (5.9%)	9 (6.7%)	4 (4.8%)
Cloves	7 (3.2%)	6 (4.4%)	1 (1.2%)
Coriander/dhania	13 (5.9%)	6 (4.4%)	7 (8.3%)
Cumin	19 (8.7%)	14 (10%)	5 (6.0%)
Garlic/saumu	57 (26%)	33 (24%)	24 (29%)
Ginger	48 (22%)	25 (19%)	23 (27%)
Nutmeg	0 (0%)	0 (0%)	0 (0%)
Paprika	13 (5.9%)	10 (7.4%)	3 (3.6%)
Turmeric	12 (5.5%)	9 (6.7%)	3 (3.6%)
White pepper	4 (1.8%)	1 (0.7%)	3 (3.6%)

[1]Product form preference (Frequency and Percentage (%))

Table 3: Duration for spice storage

	Storage Duration				
Product	Less than 1 month, N = 445[1]	1-6 Months, N = 364[1]	6-12 Months, N = 58[1]	More than 1 year, N = 14[1]	Depends on quantity purchased and frequency of use, N = 99[1]
Black pepper	39 (8.8%)	33 (9.1%)	9 (16%)	1 (7.1%)	10 (10%)
Cardamom	17 (3.8%)	18 (4.9%)	2 (3.4%)	1 (7.1%)	2 (2.0%)
Cayenne	6 (1.3%)	3 (0.8%)	1 (1.7%)	0 (0%)	0 (0%)
Red chilli	8 (1.8%)	7 (1.9%)	1 (1.7%)	0 (0%)	1 (1.0%)
Cinnamon	28 (6.3%)	28 (7.7%)	4 (6.9%)	0 (0%)	7 (7.1%)
Cloves	13 (2.9%)	16 (4.4%)	2 (3.4%)	1 (7.1%)	2 (2.0%)
Coriander	26 (5.8%)	19 (5.2%)	2 (3.4%)	1 (7.1%)	7 (7.1%)
Cumin	20 (4.5%)	15 (4.1%)	2 (3.4%)	2 (14%)	11 (11%)
Garlic	72 (16%)	46 (13%)	4 (6.9%)	0 (0%)	12 (12%)
Ginger	56 (13%)	29 (8.0%)	4 (6.9%)	2 (14%)	5 (5.1%)
Nutmeg	4 (0.9%)	1 (0.3%)	2 (3.4%)	0 (0%)	0 (0%)
Oregano	9 (2.0%)	8 (2.2%)	3 (5.2%)	2 (14%)	6 (6.1%)
Paprika	22 (4.9%)	12 (3.3%)	4 (6.9%)	0 (0%)	2 (2.0%)
Turmeric	33 (7.4%)	37 (10%)	5 (8.6%)	2 (14%)	8 (8.1%)
White pepper	10 (2.2%)	5 (1.4%)	0 (0%)	0 (0%)	0 (0%)

[1]Product storage duration (Frequency and Percentage (%))

Table 4: Duration for herb storage

Product	Storage Duration				
	Less than 1 month, N = 445[1]	1-6 Months, N = 364[1]	6-12 Months, N = 58[1]	More than 1 year, N = 14[1]	Depends on quantity purchased and frequency of use, N = 99[1]
Basil	6	11 (3.0%)	0 (0%)	1 (7.1%)	5 (5.1%)
Bay	4 (0.9%)	11 (3.0%)	4 (6.9%)	0 (0%)	0 (0%)
Marjoram	2 (0.4%)	4 (1.1%)	1 (1.7%)	0 (0%)	0 (0%)
Mint	18 (4.0%)	16 (4.4%)	1 (1.7%)	0 (0%)	9 (9.1%)
Parsley	8 (1.8%)	4 (1.1%)	1 (1.7%)	0 (0%)	1 (1.0%)
Rosemary	35 (7.9%)	30 (8.2%)	3 (5.2%)	1 (7.1%)	8 (8.1%)
Sage	0 (0%)	5 (1.4%)	0 (0%)	0 (0%)	0 (0%)
Thyme	9 (2.0%)	6 (1.6%)	3 (5.2%)	0 (0%)	3 (3.0%)

[1]Product storage duration (Frequency and Percentage (%))

Table 5: Herb consumption per meal preparation

Product	Quantity Used(grams)			
	<5g, N = 385[1]	5g, N = 267[1]	>5g, N = 49[1]	Not measured, N = 278[1]
Basil	4 (1.0%)	7 (2.6%)	1 (2.0%)	11 (4.0%)
Bay	8 (2.1%)	7 (2.6%)	1 (2.0%)	3 (1.1%)
Marjoram	5 (1.3%)	0 (0%)	0 (0%)	2 (0.7%)
Mint	16 (4.2%)	13 (4.9%)	3 (6.1%)	12 (4.3%)
Oregano	10 (2.6%)	6 (2.2%)	2 (4.1%)	10 (3.6%)
Parsley	6 (1.6%)	3 (1.1%)	1 (2.0%)	4 (1.4%)
Rosemary	32 (8.3%)	21 (7.9%)	6 (12%)	18 (6.5%)
Sage	3 (0.8%)	0 (0%)	0 (0%)	2 (0.7%)
Thyme	4 (1.0%)	5 (1.9%)	0 (0%)	12 (4.3%)

Table 6: Spice consumption per meal preparation

Product	Quantity Used(grams)			
	<5g, N = 385[1]	5g, N = 267[1]	>5g, N = 49[1]	Not measured, N = 278[1]
Black pepper	31 (8.1%)	30 (11%)	7 (14%)	24 (8.6%)
Cardamom	15 (3.9%)	12 (4.5%)	1 (2.0%)	12 (4.3%)
Cayenne	5 (1.3%)	1 (0.4%)	1 (2.0%)	3 (1.1%)
Red chilli	6 (1.6%)	5 (1.9%)	0 (0%)	6 (2.2%)
Cinnamon	29 (7.5%)	13 (4.9%)	3 (6.1%)	22 (7.9%)
Cloves	11 (2.9%)	6 (2.2%)	1 (2.0%)	16 (5.8%)
Coriander	23 (6.0%)	12 (4.5%)	1 (2.0%)	19 (6.8%)
Cumin	19 (4.9%)	11 (4.1%)	3 (6.1%)	17 (6.1%)
Garlic	63 (16%)	40 (15%)	4 (8.2%)	27 (9.7%)
Ginger	32 (8.3%)	34 (13%)	4 (8.2%)	26 (9.4%)
Nutmeg	3 (0.8%)	1 (0.4%)	0 (0%)	3 (1.1%)
Turmeric	37 (9.6%)	19 (7.1%)	6 (12%)	22 (7.9%)
White pepper	5 (1.3%)	3 (1.1%)	3 (6.1%)	4 (1.4%)
Paprika	18 (4.7%)	18 (6.7%)	1 (2.0%)	3 (1.1%)