

**MYCOTOXIN HAZARDS IN THE KENYAN FOOD AND FEED MARKET
- A RETROSPECTIVE STUDY****Kibugu J^{1,2*}, Mburu D², Munga L², Lusweti F^{1α}, Grace D³ and J Lindahl^{3, 4, 5}****Kibugu Karuku James**

*Corresponding author email: jkkibugu1@yahoo.com

¹Kenya Agricultural and Livestock Research Organization (KALRO),
P. O, Box 362-00902, Kikuyu, Kenya

^{1α}KALRO (Retired), c/o P. O. Box 57811-00100, Nairobi, Kenya

²Kenyatta University, P. O Box 43844-00100, Nairobi, Kenya

³International Livestock Research Institute, Box 30709-00100, Nairobi, Kenya

⁴Swedish University of Agricultural Sciences, Uppsala, Sweden

⁵Uppsala University, Uppsala, Sweden



ABSTRACT

Mycotoxins are toxic fungal metabolites naturally found in food and feed as contaminants. Animal feed and human food samples (n=1818) from three major Kenyan laboratories were categorized as compliant and non-compliant according to Kenya, America (USA) and Europe (EU) mycotoxin regulatory limits. Quantitative risk assessment of dietary aflatoxin intake in maize, wheat, peanut and dairy products in relation to human hepatocellular carcinoma was carried out employing deterministic approach. Non-compliant samples' proportions were calculated, and logistic regression and chi-square test used to compare different commodities. Animal feed were least compliant, with 64% and 39% having total aflatoxin (AFT) levels above Kenya and USA standards, respectively. Peanuts were the most non-compliant food, with 61% and 47% samples failing Kenya and USA AFT standards respectively, while wheat was least compliant (84%) according to EU threshold for AFT. Half of baby food sampled had AFT level above Kenya and EU standards. High non-compliance rate with Kenya, USA and EU regulatory thresholds with respect to seven different mycotoxins (summarized as "mycotoxins"), and also AFT and aflatoxin M1 alone in edible materials is reported. Significant non-compliance is reported for compound animal feed, peanuts, wheat, baby food, feed ingredients, herbal healthy drink, maize and fodder feed in that order. High levels of aflatoxin residues in animal feed and human food was also observed. Lifetime human consumption of wheat and maize leads to high additional risk for primary liver cancer, human hepatocellular carcinoma (HCC) associated with dietary aflatoxin, wheat and its products causing the highest disease burden. Subsequent implications and limitations of current food safety standards are discussed. Humans and animals in Kenya appear to be chronically exposed to mycotoxin hazards: this calls for surveillance and risk management. There is urgent need for enhanced and consistent surveillance of the dietary mycotoxin hazards observed in this study employing representative sampling plans. Regulation and future research need to focus on reliable analysis techniques, collection of data on toxicological effects of mycotoxins and food consumption pattern, and regulatory limits accordingly set and compliance enforced to protect vulnerable groups such as paediatric, geriatric and sick members of the society to reduce cancer burden in Kenya.

Key words: Mycotoxins, food, feed, risk analysis, human hepatocellular carcinoma, Kenya



INTRODUCTION

Mycotoxins are toxic secondary metabolites produced by fungi that infest food and feed. Commonly encountered mycotoxins are aflatoxins, ochratoxins, zearalenone, fumonisins, trichothecenes and patulin, which are produced by the fungal genera *Aspergillus*, *Penicillium* and *Fusarium* [1]. Humans in Africa are often exposed to mycotoxins in food. Chronic exposure is associated with aggravation of disease pathogenesis in experimental animals and humans [2,3], reduced animal productivity and impaired animal nutrition [4]. Mycotoxins can be teratogenic, carcinogenic, mutagenic, estrogenic, nephrotoxic, hepatotoxic and immunosuppressive [2]. Aflatoxin is an important contributor to primary hepatocellular carcinoma, childhood stunting and immunosuppression [5]. Aflatoxigenic fungi are widely distributed in Kenya, and acute aflatoxicosis in human and animals resulting in deaths has been reported [6]. Besides threatening human and animal health, mycotoxins also affect international trading and contribute to food insecurity [1]. Most mycotoxins are stable to normal cooking and processing. After consumption, some mycotoxin metabolites can be carried over *in utero* [7], breast milk [8] and animal products: all these contribute to mycotoxin exposure in humans.

To protect humans from exposure, many countries have regulatory limits for some mycotoxins. However, standards are rarely enforced in the developing world [9]. In Kenya, there are commercial and government laboratories that offer laboratory testing for food material destined for export and local consumption. The purpose of this study was to review available mycotoxin contamination data from testing laboratories in order to identify potential mycotoxin hazard-prone edible materials in Kenya for better understanding of the associated health risks.

MATERIALS AND METHODS

Data collection and management

A list of 23 facilities involved in mycotoxin analysis in Kenya based on a report by Kang'ethe was developed [6]. A three-step sampling procedure was employed to collect the data. Briefly, official request for participation were sent to all the 23 facilities, and 7 responded positively. Based on initial review of the size, completeness of data and records' quality, three major mycotoxin-testing laboratories were further selected from the sub-sample of seven laboratories. It is because of these reasons and lack of active involvement in mycotoxin analysis that most of the laboratories mentioned above declined to participate in this study. The participants were coded Lab1, Lab2 and Lab3 for purpose of confidentiality. All samples analysed in Lab1 (a government research facility) were randomly collected by researchers while those analysed in Lab2 and Lab3 (private facilities) were collected by scientific staff under instructions of the analyzing laboratory and delivered for analysis by clients. These samples were therefore presumed to be representative of the targeted consignments. All the three laboratories employed quantitative monoclonal antibody-based enzyme immunoassays whose limit of detection (LOD) and upper limit of detection range (ULDR) for aflatoxin analysis were as follows: Lab 1 (Helica Total Aflatoxin Kit: LOD= 1.5 ppb, ULDR= 30 ppb); Lab 2 (Romer Labs Total Aflatoxin Kit: LOD= 0.05



ppb; ULDR=100 ppb); Lab 3 (Ridascreen Total Aflatoxin Kit: LOD= 1.75 ppb; ULDR= 141.75 ppb and Ridascreen Aflatoxin M1 Kit: LOD= 5 ppt; ULDR= 80 ppt.).

The data were broadly grouped into animal feed and human food and then animal feed categorized into compound feed, feed ingredients, and fodder while human food were categorized as baby food, herbal health drink, maize, peanuts, dairy products, tea, wheat, on-the-plate meals comprising of maize slurry, vegetables and omena (silver fish). The aflatoxin level data from the laboratory records were examined and appropriately posted. Those entered as \leq LOD were re-entered as zero while those indicated as \geq ULDR were taken as the upper limit value. Details such as sample type and origin, test conducted, analytical method employed and test results obtained were recorded. All mycotoxin regulatory limits applied were within the range of analytical methods employed by the participant laboratories. When the mycotoxin level results were indicated as $<$ LOD, or more than $>$ LOD, the binary dependent response values were respectively compliance (denoted by 0) and non-compliance (denoted by 1) with regulatory thresholds.

Kenyan standards for animal feed and human food [10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21] were applied to categorize the materials as either above or below maximum admissible levels. Similarly, this was repeated with United States Food and Drugs Administration (FDA) and European Union (EU) standards [22,23,24,25,26,27,28]. Tables 1 and 2 show the maximum limits (MLs) of mycotoxin residues as stipulated in these standards for regulation of contaminants in food. A sample was considered non-compliant with regulations if its mycotoxin level surpasses the threshold prescribed by regulatory standard. A sample had analysis result of only one type of mycotoxin. Ethical approval of this study was given by the National Commission for Science, Technology & Innovation of the Republic of Kenya (License No: NACOSTI/P/19/1276).

Risk analysis

Quantitative risk assessment of dietary aflatoxin intake in maize, wheat, peanut and dairy products was done employing deterministic approach designed by FAO/ WHO [29] as described by Benkerroum [30]. The risk was calculated as carcinogenic potency multiplied by dietary exposure to AFB1 using equations 1 and 2 where the former is a function of seropositive and seronegative individuals for surface antigen of hepatitis B1 virus. The estimated per capita consumption for maize, wheat, milk (and their products) in the year 2018 was as per KNBS [31]. For peanuts, daily consumption 11.3 g/day, an estimation generalized for all African countries [29] while average body weight (bw) for an adult male in Kenya is 60 kg [29,32].

$$P_{\text{cancer}} = (\text{PHBsAg}^+ \times \text{FHBsAg}^+) + (\text{PHBsAg}^- \times \text{FHBsAg}^-) \quad \text{Equation 1}$$

Where,

P_{cancer} = Hepatitis B virus-adjusted carcinogenic potency = 0.083 cancers/ 100,000 people/ amount of aflatoxin (ng) / bw (kg)/ day = 0.083 for aflatoxin B1 [30] and 10-fold lower for aflatoxin M1 [32]



HBsAg⁺, and HBsAg⁻ = Seropositivity and seronegativity respectively for hepatitis B virus surface antigen
 PHBsAg⁺ and PHBsAg⁻ = Carcinogenic potency of aflatoxin B1 in HBsAg⁺ and HBsAg⁻ individuals=0.3 and 0.01 cancers/year/100,000 individuals per ng aflatoxin B1 per kg bw per day respectively
 FHBsAg⁺ and FHBsAg⁻ = Population fractions of HBsAg⁺ (25%) and HBsAg⁻ (75%) respectively

$$R = P_{\text{cancer}} \times \text{EDI} = 0.083 \times \text{EDI}$$

Equation 2

Where,

R= Probability (risk) of primary liver cancer for lifetime exposure or annual incidence expressed as number of cancers per year per 100,000 individuals
 EDI= Estimated daily intake expressed as ng aflatoxin B1 per 60 kg bw per day and calculated by multiplication of contamination level (ng/g) by the consumption rate (g/day) of contaminated food per kg bw.

Data analysis

Proportions of samples not compliant with mycotoxin regulatory thresholds were computed and expressed as a percentage of the total number of samples. Association between matrices (animal feed and human food) as predictors, and the binary response, that is non-compliance or compliance with regulatory limits for AFT content, was further determined employing Pearson chi square-test of independence. Logistic regression analysis was carried out to obtain odds ratios for the dependent response; above (non-compliance with standard=1) or below (compliance with standard=0) legal limits as stipulated in aflatoxin regulatory standards. The explanatory variables were 12 different feed/ food commodities while the dependent variable was the dichotomous outcome: failed (non-compliant) or met (compliant) the regulatory standards. The data had a binomial distribution, B(n, p), where n = number of feed or food samples and p= probability of non-compliance with food safety standards. The following binary logistic regression model was fitted to the data on statistical computer program (IBM SPSS Statistics 20):

$$\log\{p/(1-p)\} = \beta_0 + \beta_1 \text{Food/Feed matrix} + \epsilon_i$$

Equation 3

Where,

β_0 = Intercept,

β_1 food/feed matrix= regression coefficient for food/feed matrices

ϵ = random error

Using this model, effect of feed/ food matrix on the dependent binary outcome variable response was determined.



RESULTS AND DISCUSSION

Laboratories and samples

Three laboratories (Lab1, Lab2, and Lab3) participated in this study. Results for 1818 samples (323 and 1495 samples of animal feed and human food respectively) analysed for mycotoxin residues in Kenya between 2010 and 2015 were received. Since, this study was not designed to assess laboratories' proficiency, no attempt was made to compare their competence. Further, the purposes for analysis were varied ranging from research, routine monitoring to outbreak of gastrointestinal conditions. It was noted that maize was the most frequently tested food material. Sample sizes were in some cases inadequate and some important food items were left out. Nevertheless, this report provides the first, credible and comprehensive compilation of large-scale laboratory data on mycotoxins in Kenya.

Comparison of rates of non-compliance with regulatory mycotoxin content thresholds in animal feed and human food

The USA and EU standards were used because of their credibility, stringency, global importance and relevance to Kenyan exports and imports. Results on failure to comply with standards for total aflatoxins (AFT) in animal feed and human food are shown in Table 3. By Kenyan standards, animal feed had significantly ($p < 0.01$) higher AFT content non-compliance (32%) than human food (25%). By USA standard, no significant association was observed. Highest non-compliance rates were observed in compound feed, feed ingredients, peanuts and wheat in that order. This agrees with other authors' observations that animals are fed products considered unfit for human consumption [33,34]. All feed materials: compound feed, feed ingredients and fodder feed in that order had significant rates of non-compliance to AFT regulatory thresholds. Presence of aflatoxin hazards in animal feed and human food is of concern since chronic dietary mycotoxicosis is associated with adverse health effects [4] and reduced productivity [33] resulting in economic loss [1].

Non-compliance rates and effect of feed/ food matrices

Non-compliance rates for three feed ($n=323$) and nine food materials ($n=1495$) tested for aflatoxins and non-aflatoxin mycotoxins are shown in Tables 3 and 4, respectively. Of these, there were results for 1607 feed and food products samples tested for AFT and 192 dairy products samples tested for AFM1 (Table 3), and only 19 samples tested for non-aflatoxin mycotoxins (Table 4). Compound animal feed was the most non-compliant feed type with proportions of 64 % and 39 % having AFT levels above regulatory limit by Kenyan and USA standards respectively of the 92 samples tested (Table 3). Peanuts were the most non-compliant human food with proportions of 62 % and 47 % of 180 tested samples having levels surpassing legal limit by Kenya and USA standards respectively for AFT. By EU standards for AFT, wheat had the highest non-compliance rate with 84 % of 105 samples tested having levels surpassing regulatory limit. Half (50%) of the baby food failed Kenyan and EU standards for AFT. This corroborates previous studies. Baby food samples from Turkey [35] and Pakistan [36] had 87% and 40% aflatoxin contamination rates respectively. Effects of infantile aflatoxin exposure is evident in Kenya [37]. By Kenyan, USA and EU standards, respectively, maize (a common staple food in Kenya) had AFT failure rates of 20, 14



and 25 %. Again, maize was the most frequently tested food. Dairy products, largely comprising of raw milk, had high non-compliance rate of 59.9 % according to EU standards (Table 3).

Table 5 shows logistic regression results of the explanatory variable giving odds ratios (OR) of dietary AFT hazard occurrence as a function of EU, USA and Kenya standards for regulation of food contaminants. Hosner & Lemeshow test ($p > 0.01$) indicates good model fit. Compared to reference food matrix (on-the-plate food with AFT mean level below the three regulatory thresholds and large sample size was considered a good background reference material), high frequency of AFT hazard were observed in several food/ feed materials relative to threshold limits set by Kenya, USA and EU standards. By EU standards, odds of surpassing AFT limits were higher in wheat (OR=17.0), peanuts (OR=7.1) and herbal healthy drink (OR=2.8). Considering the USA standards, the odds of surpassing AFT regulatory limit were higher in compound feed (OR=14.2), and fodder feed (OR=3.9). In human food, odds of surpassing AFT threshold were higher in peanuts (OR=19.7), wheat (OR=18.6), maize (OR=3.5) and baby food (OR=11.0) compared to reference. By Kenya standards, odds of surpassing AFT regulatory limits were higher in feed ingredients (OR=3.7), in compound feed (OR=17.6), peanuts (OR=15.9), wheat (OR=11.7), baby food (OR=9.9) and maize (OR=2.4), herbal health drink (OR =3.1) and fodder feed (OR=1.8).

Earlier surveys conducted between 1960 and 2018 show non-compliance with Kenyan standards for aflatoxins of 15-83% in maize, 37-43% in peanuts, 11% in sorghum, 10-63% in milk products, 95% in animal feed, and nil in medicinal herbs [38]. Peanuts and wheat were identified as food with the highest rate of non-compliance with AFT thresholds stipulated in food safety standards. Large proportions of non-compliance with Kenyan AFT standards of 62% in peanuts, 54% in wheat, 50% in baby food, 24% in herbal health drinks, 20% in maize, and non-compliance with EU aflatoxin M1 standard of 59.9 % in dairy products was observed. Of these, peanuts, wheat, baby food, herbal healthy drink, and maize, in that order had significant rates of non-compliance to AFT thresholds. These are readily available food items commonly consumed by children and adults. In Kenya, food such as cereals and dairy products are consumed by infants and young children. Aflatoxin is anti-nutritional, and is associated with child growth faltering and oncogenesis [7,39,40]. Few and scanty records for five non-aflatoxin mycotoxins, indicate that surveillance is limited to only one group of toxins, albeit a group of high importance to public health. This corroborates previous studies where over emphasis on aflatoxins at expense of other mycotoxins in developing countries was observed [5].

Aflatoxin levels and hepatocellular carcinoma (HCC) risk analysis of selected human food

Table 6 shows levels of dietary aflatoxins in animal feed and human food from Kenya, and risk for HCC as a function of dietary exposure to aflatoxin in maize, wheat, peanuts, milk. Compound feed had the highest levels of AFT. For human food, high levels of AFT were observed in peanuts, maize, baby food and wheat while high levels of AFM1 were observed in dairy products. Of the four items analysed, highest additional risk for HCC was associated with dietary aflatoxin in wheat and maize.



However it should be noted that the peanut consumption data used here were based on estimation of 1998 [29] and prevailing risk for this food item is likely to be higher. Negligible risk was observed for lifetime exposure to AFM1 in milk, corroborating Sirma *et al.* [32] who observed minimal risk of HCC from AFM1 in milk from Kenya.

The role of aflatoxins is well-documented in development of hepatocellular carcinoma through synergy with hepatitis B virus [5,30], and interaction with human papillomavirus in induction of oesophageal malignancy [34,39]. High frequency of dietary aflatoxin in food destined for consumption by paediatric and pregnant individuals was observed suggesting exposure to these carcinogens commences early in life. In fact, *in utero* and paediatric mycotoxin exposure is common in Africa [7,8]. Since malignancy depends on exposure in terms of dose and time [41], the young being more susceptible to carcinogens especially mycotoxins [40], this study confirms aflatoxin as a risk factor contributing to occurrence of cancer in relatively young individuals in Kenya. Indeed, there was increased burden of HCC associated with dietary aflatoxin in wheat and maize. Oncogenesis can be induced via reduced immuno-competence or potentiation of carcinogenic infections. Aflatoxicosis could be a risk factor in development of many infection-associated malignancies through either synergy, exacerbation of carcinogenic infections or immunosuppression. Aflatoxicosis was recently associated with risk of oncogenic human papilloma virus infection detection in cervical samples of Kenyan women [39]. Further studies are required to elucidate possible potentiation of carcinogenic biological agents by chronic aflatoxicosis and incidence of various cancers.

Revision in some areas of the national mycotoxin regulation standards is suggested. The EU standard is strict with very low thresholds. For example, the maximum limit for AFM1 in processed dairy products by Kenya [19], USA [22] and FAO/WHO's *Codex Alimentarius* [19] is 10-fold higher (500 ppt) than EU's 50 ppt [23]. Further, EU set the limit to 25 ppt in infant formulae and milk to protect infants and young children. Many items consumed locally are not covered by the safety standards. It seems international standards are adopted in their original version without tailoring them for local scenario. Further, although there is likelihood of other mycotoxins such as ochratoxins, zearalenone and T-2 toxin, they are not mentioned in the national standards. These agree with Trench *et al.* [5] that although various mycotoxins occur in developing countries, more emphasis is put on aflatoxins. Further, although some national standards address food safety of infants and children [14,16], their regulatory statements are vague, with no threshold and difficult to interpret. Another issue is that enforcement of regulatory standards is difficult in developing countries including Kenya [9]. In this case, there is a trade-off between very high standards that are difficult to meet and enforce but which provide high protection, and standards that are easier to meet and enforce but entail more risk to human health. Protection of people from dietary contaminants, is a function of both establishment of sound regulatory limits and their effective enforcement, and very strict standards might have the paradoxical effect of increasing exposure to mycotoxins. In sub-Saharan Africa, where food security is inadequate, need to feed increasing populations should be considered alongside health benefits of mycotoxin regulation. These have been echoed by other authors [42]. Lastly, consumption data of local food items including baby food need to

be collected. Recent proposal to revise Kenya's food safety standards at the East African Community platform is a welcome move.

CONCLUSION

Large proportions of feed and food samples surpassed aflatoxin regulatory thresholds, significant rates of non-compliant being observed in compound feed, peanuts, wheat, baby food, feed ingredients, herbal healthy drink, maize and fodder feed in that order. Animals and humans, including infants, children and expectant mothers, in Kenya appear to be exposed to dietary aflatoxin hazards which could lead to serious economic and health implications. Kenyan food safety regulatory system does not cover many food and feed items consumed locally, is largely silent on non-aflatoxin mycotoxins, and at times gives vague regulatory statements. Lifetime human consumption of wheat and maize leads to high additional risk for primary liver cancer (HCC) associated with dietary aflatoxin; wheat causing the highest disease burden. Surveillance of the mycotoxin hazards observed in this study should be enhanced, safety regulatory standards revised to include all mycotoxins observed in this study and compliance enforced to protect vulnerable groups such as paediatric, geriatric and the sick.

ACKNOWLEDGEMENTS

The CGIAR Research Program, Agriculture for Nutrition and Health, and KALRO funded this study. Permission to publish this paper was granted by the Director General, KALRO. All the data used herein was kindly provided by KALRO-Food Crop Research Institute-Kitale (Mr. Phochunatus Sifuna), Bora Biotech Ltd. (Mr. George Kimani), and Polucon Services (K) Ltd (Mr. Charles Maina). Mr. David Kinoti and Ms Joanna Auma (KALRO) provided statistical and bibliography softwares respectively. A former science teacher, Mr. Robert S. I. Karuku is acknowledged posthumously for inspiring first author to the world of food poisoning, the main drive in this communication.



Table 1: Feed safety regulatory standards used in the study

Feed matrix	Toxin	Feed safety regulatory standard					
		KEBS standard		US-FDA standard		EU standard	
		RL (ppb)	Reference	RL (ppb)	Reference	RL (ppb)	Reference
Wheat Bran (Feed ingredient)	AFT	20	[11]	20	[24,25,26]	NS ^{AFT}	-
Omena (Feed ingredient)	AFT	20	[12]	20			
Cotton Seed/ cake (Feed ingredient)	AFT	10	[10]	20			
Sunflower Cake (Feed ingredient)	AFT	10	[10]	20			
Maize products (Feed ingredient)	AFT	10	[21]	20			
Wheat Bran (Feed ingredient)	FUM	2000	[19]	NS			
Compound feed	AFT	10	[19]	20			
Fodder feed	AFT	20	[20]	20			
Wheat Bran (Feed ingredient)	DON	1000	[19]	5000			
Wheat Bran (Feed ingredient)	OTA	5	[19]	NS	250	[27]	
Feed ingredient	ZEA	NS		NS	2000	[27]	

RL-Regulatory limit; US-FDA- Food and Drug Administration of the United States of America; KEBS-Kenya Bureau of Standards; ppb-parts per billion; AFT-Total aflatoxins; DON-Deoxynivalenol; FUM-Total fumonisins; OTA-Ochratoxin A; ZEA-Zearalenone; NS^{AFT} –EU standard for animal feed has limit for AFB₁ but none for AFT

Table 2: Food safety regulatory standards used in the study

Food matrix	Toxin	RL (ppb)	KEBS standard	US-FDA standard		EU standard	
			Reference	RL (ppb)	Reference	RL (ppb)	Reference
Baby food	AFT	0	[14,16]	20	[24]	NS	[23]
Herbal health drink	AFT	10	[19]				
Maize			[13]				
Peanuts			[15,19]				
Sorghum			[17]				
Tea			[19]				
Wheat			[13]				
Wheat			DON	1000	[19]	1000	[26]
Wheat	T-2 toxin	NS	-	NS	-	100	
On-the-plate food	AFT	10	[12,13]	20	[24]	4	[23]
Barley (other food)			[13]				
Beans (other food)			[18]				
Chilli (other food)			[19]				
Macadamia (other food)			[19]				
Omena fish (other food)			[12]				
Dairy products	AFM1	0.5	[19]	0.5	[22]	0.05	[23]
Baby food	ZEA	MF	[16]	NS	-	20	
Baby food	DON	MF	[16]	1000	[26]	200	[28]

RL-Regulatory limit; NS-Regulatory limit not set, MF-Mycotoxin free; US-FDA- Food and Drug Administration of the United States of America; KEBS-Kenya Bureau of Standards; ppm-parts per million; ppb-parts per billion; ppt-parts per trillion; AFT-Total aflatoxins; AFM1-aflatoxin M1; DON-Deoxynivalenol; ZEA-Zearalenone



Table 3: Non-compliance rates with aflatoxin content standards in animal feed and human food

<i>Food/feed matrix</i>	<i>Aflatoxin</i>	<i>Proportion (%) of non-compliant samples</i>		
		<i>KEBS standard</i>	<i>FDA standard</i>	<i>EU standard</i>
<i>Animal feed (n=310)</i>	AFT	32**	20	NS
<i>Human food (n=1297)</i>		25**	18	36
<i>Feed ingredients (n=64)</i>		27	7	NS
<i>Compound feed (n=92)</i>		64	39	NS
<i>Fodder feed (n=154)</i>		16	15	NS
<i>Baby food (n=6)</i>		50	33	NS
<i>Herbal health drink (n=21)</i>		24	10	48
<i>Maize (n=561)</i>		20	13	25
<i>Peanuts (n=180)</i>		62	47	70
<i>Tea (n=37)</i>		0	0	0
<i>Wheat (n=105)</i>		54	46	85
<i>On-the- plate food (n=369)</i>		9	4	25
<i>Other food (n=18)</i>		17	0	25
<i>Dairy products (n=192)</i>	AFM1	0.52	0.52	59.9

NS=Regulatory limit not set. ** The two values differ significantly ($p<0.01$)

Table 4: Non-compliance rates for non-aflatoxin mycotoxins and their levels in animal feed and human food in Kenya (2010-2015)

Matrix	Specific feed/ food matrix	Type of mycotoxin	Mycotoxin content expressed as arithmetic range (ppb)	Proportion (%) of non-compliant samples		
				KEBS standard	FDA standard	EU standard
<i>Animal Feed</i>	Feed ingredient	Zearalenone (n=3)	0-2	-	-	0
		Ochratoxin A (n=1)	5	100	-	-
		Fumonisin (n=5)	0-222	0	-	-
		Deoxynivalenol (n=3)	30-200	0	-	-
	Compound feed	Fumonisin (n=1)	1370	0	0	-
<i>Human Food</i>	Baby food	Zearalenone (n=1)	28	100	-	100
		Deoxynivalenol (n=1)	0	0	0	0
	Wheat	Deoxynivalenol (n=2)	7-144.2	0	0	0
		T-2 Toxin (n=2)	26	-	-	0

ppb=parts per billion



Table 5: Association between non-compliance with regulatory thresholds of total aflatoxin contents as stipulated in EU, FDA and KEBS standards for regulation of contaminants in food

Predictors (Food commodities)	Food safety standards					
	European Union		Food & Drug administration (FDA)		Kenya Bureau of Standards (KEBS)	
	Regression coefficient	Odds Ratio	Regression coefficient	Odds Ratio	Regression coefficient	Odds Ratio
<i>Feed ingredients (n=62)</i>	-		0.19 (n=58)	1.2	1.31***	3.7
<i>Compound feed (n=92)</i>	-	-	2.65***	14.2	2.87***	17.6
<i>Fodder feed (n=154)</i>	-	-	1.35***	3.9	0.60*	1.8
<i>Baby food (n=6)</i>	1.12	3.1	2.40**	11.0	2.29**	9.9
<i>Herbal healthy drink (n=21)</i>	1.02*	2.8	0.84	2.3	1.12*	3.1
<i>Maize (n=561)</i>	0.04	1.1	1.24***	3.5	0.88***	2.4
<i>Other food (n=18)</i>	0.02 (n=12)	1.0	-18.11	1.4 x 10 ⁻⁸	0.21	1.2
<i>Peanuts (n=180)</i>	1.96***	7.1	2.98***	19.7	2.76***	15.9
<i>Tea (n=37)</i>	-20.09	0.0	-18.11	1.4 x 10 ⁻⁸	-18.92	6.1 x 10 ⁻⁹
<i>Wheat (n=105)</i>	2.83***	17.0	2.92***	18.6	2.46***	11.7
<i>Model intercept</i>	-1.12***	-	-3.09***	-	-2.29***	-
<i>Reference: On-the-plate food (n=369)</i>						

*p<0.05; ** p<0.01; *** p<0.001



Table 6: Aflatoxin levels in animal and human food in Kenya (2010-2015) and associated estimated burden of hepatocellular carcinoma

Matrix	Specific food/ feed matrix	Type of aflatoxin	Aflatoxin level (ng/g)		Food consumption rate		EDI	P _{cancer}	R
					Per capita consumption (kg/year)	Daily consumption (g/day)			
<i>Animal feed</i>	Feed ingredient-Maize Germ (n=1)	Aflatoxin total	25		-	-	-	-	-
	Feed ingredient-Corn Meal (n=1)		0						
	Feed ingredient-Cotton Seed (n=6)		23.38						
	Feed ingredient-Omena* (n=28)		6.91						
	Feed ingredient-Sunflower (n=2)		6.50						
	Feed ingredient-Wheat Bran (n=24)		1.10						
	Compound feed (n=92)		22.45						
	Fodder feed (n=154)		13.44						
<i>Human food</i>	Baby food (n=5)	Aflatoxin total	28.8		69.5	190.4	30.4	0.083	2.5
	Herbal health drink (n=21)		5.91						
	Maize (n=561)		9.57						
	Wheat (n=105)		17.01						
	Peanuts (n=178)		50.51						
	Tea (n=37)		-						
	On-the-Plate (n=369)		3.93						
	Other food (n=18)		2.76						
Dairy products (n=192)	Aflatoxin M1	0.062		93.3	255.6	0.3	0.0083	0.02	

*=*Rastrineobola argentea* (Silver fish/ "mukene"); EDI=Estimated daily intake; P_{cancer}=carcinogenic potency; R=Risk of hepatocellular carcinoma



REFERENCES

1. **Alshannaq A and JH Yu** Occurrence, toxicity, and analysis of major mycotoxins in food. *International Journal of Environmental Research and Public Health*. 2017; **14**.
2. **World Health Organization and International Agency for Research on Cancer**. IARC monographs on the evaluation of carcinogenic risks to humans: some traditional herbal medicines, some mycotoxins, naphthalene and styrene (Volume 82). IARC Press, Lyon, France. 2002: 601 pages.
3. **Kibugu JK, Ngeranwa JJN, Makumi JN, Gathumbi JK, Kagira JM, Muchiri MW and RE Mdachi** Aggravation of pathogenesis mediated by ochratoxin A in mice infected with *Trypanosoma brucei rhodesiense*. *Parasitology*. 2009; **136**: 273-281.
4. **Indresh HC, Devegowda G, Ruban SW and MC Shivakumar** Effects of high grade bentonite on performance, organ weights and serum biochemistry during aflatoxicosis in broilers. *Veterinary World*. 2013; **6(6)**: 313-317.
5. **Trench PC, Narrod C, Roy D and M Tiongco** Responding to health risks along the value chain. 2020 Conference: Leveraging Agriculture for Improving Nutrition and Health. February 10-12, 2011; New Delhi, India: International Food Policy Research Institute (IFPRI). 2011; 54 pages.
6. **Kang'ethe E** Situation analysis: improving food safety in the maize value chain in Kenya. Report prepared for FAO by Prof. Erastus Kang'ethe College of Agriculture and Veterinary Science University of Nairobi September 2011. 2011; 89 pages.
7. **Turner PC, Collinson AC, Cheung YB, Gong YY, Hall AJ, Prentice AM and PC Wild** Aflatoxin exposure *in utero* causes growth faltering in Gambian infants. *International Journal of Epidemiology*. 2007; **36**: 1119–1125.
8. **Obade M, Andang'o P, Obonyo C and F Lusweti** Exposure of children 4 to 6 months of age to aflatoxin in Kisumu County, Kenya. *African Journal of Food, Agriculture, Nutrition and Development*. 2015; **15(2)**: 9949-9963.
9. **Ferrão J, Bell V and TH Fernandes** Mycotoxins, food safety and security in Sub-saharan Africa. *SM Journal of Food and Nutritional Disorders*. 2017; **3(2)**: 1021.
10. **Kenya Bureau of Standards (KEBS)**. Oil-seed cakes for compounding livestock feeds- Specification. 1st Edition. KS EAS 287. 2002. ICS 67.120. Kenya Bureau of Standards 2002.



11. **Kenya Bureau of Standards (KEBS)**. Wheat bran for animal feeds- Specification. 1st Edition 2004. KS EAS 353:2004. ICS 67. 060. Kenya Bureau of Standards 2004.
12. **Kenya Bureau of Standards (KEBS)**. Dried rastrineobola argentea (omena/ dogaci/ mukene). 2nd Edition 2011. KS 1470. 2011a. ICS 67. 120: 30. Kenya Bureau of Standards 2011a.
13. **Kenya Bureau of Standards (KEBS)**. Milled cereals and pulses products – Specification. 1st Edition 2011. KS 2271:2011b. ICS 67.060. Kenya Bureau of Standards 2011b.
14. **Kenya Bureau of Standards (KEBS)**. Infant formula-Specification. 1st Edition. KS EAS 4:2013; ICS 67.230. Kenya Bureau of Standards 2014a.
15. **Kenya Bureau of Standards (KEBS)** (2014b). Peanut butter-Specification. 1st Edition 2014. KS EAS 60: 2013. ICS 67. 200.10. Kenya Bureau of Standards 2014b.
16. **Kenya Bureau of Standards (KEBS)**. Processed cereal-based foods for infants and young children- Specification. 1st Edition 2014. KS EAS 72: 2013. ICS 67.230. Kenya Bureau of Standards 2014c.
17. **Kenya Bureau of Standards (KEBS)**. Sorghum grains- Specification. 1st Edition 2014. KS EAS 757:2013. ICS 67.060. Kenya Bureau of Standards 2014d.
18. **Kenya Bureau of Standards (KEBS)**. Faba beans- Specification. 1st Edition 2014. KS EAS 763: 2013. ICS 67.060. Kenya Bureau of Standards 2014e.
19. **Kenya Bureau of Standards (KEBS)**. General standard for contaminants in feeds and foods. 1st Edition. KS Codex Stan 193:1995. ICS 67.050. Kenya Bureau of Standards 2017.
20. **Kenya Bureau of Standards (KEBS)**. Fodder hay-Specifications. Part 1: Grass hay. 1st Edition. KS 2832-1: 2019. ICS 65:120. Kenya Bureau of Standards 2019.
21. **Kenya Bureau of Standards (KEBS)**. Maize gluten meal - Specification. 1st Edition. KS EAS 2358: 2012. ICS 65.120. Kenya Bureau of Standards 2012.
22. **Food and Drug Administration of the United States of America**. Compliance policy guide (CPG Sec. 527.400). Whole milk, low fat milk, skim Milk - Aflatoxin M1. 2005 ed: U.S. Department of Health and Human Services Food and Drug Administration. Available from: <https://www.fda.gov/regulatory-information/search-fda-guidance-documents/cpg-sec-527400-whole-milk-lowfat-milk-skim-milk-aflatoxin-m1> Date accessed; 20th February, 2015.



23. **European Union.** Amending Regulation (EC) No 1881/2006 setting maximum levels for certain contaminants in foodstuffs as regards aflatoxins. Commission regulation (EU) No 165/2010 of 26 February 2010. Official Journal of the European Union. 2010; **27.2.2010**: L 50/8- L 50/12.
24. **Food and Drug Administration of the United States of America.** Guidance for industry: Action levels for poisonous or deleterious substances in human food and animal feed. August 2000: U.S. Department of Health and Human Services, US Food and Drugs Administration. Available from: <https://www.fda.gov/food/guidanceregulation/guidancedocumentsregulatoryinformation/ucm077969.htm> Date page last updated; 11th October, 2017 and *Date accessed*; 27th February 2019.
25. **Food and Drug Administration of the United States of America.** Sec. 683.100 Action levels for aflatoxins in animal food. U.S. Department of Health and Human Services Food and Drug Administration Office of Regulatory Affairs and Center for Veterinary Medicine. Available from: <https://www.fda.gov/ICECI/ComplianceManuals/CompliancePolicyGuidanceManual/default.htm> *Date accessed*; 29th June, 2019.
26. **Food and Drug Administration of the United States of America.** Guidance for Industry and FDA: advisory levels for deoxynivalenol (DON) in finished wheat products for human consumption and grains and grain by-products used for animal feed July 2010. Available from: <https://www.fda.gov/regulatory-information/search-fda-guidance-documents/guidance-industry-and-fda-advisory-levels-deoxynivalenol-don-finished-wheat-products-human> *Date accessed*; 17th September, 2018.
27. **European Union.** Commission recommendation of 17 August 2006 on the presence of deoxynivalenol, zearalenone, ochratoxin A, T-2 and HT-2 and fumonisins in products intended for animal feeding. 2006/576/EC). Official Journal of the European Union. 2006; **23.8.2006**: L 229/7- L 229/9.
28. **European Union.** Amending Regulation (EC) No 1881/2006 setting maximum levels for certain contaminants in foodstuffs as regards Fusarium toxins in maize and maize products. Commission regulation (EC) No 1126/2007 of 28 September 2007. Official Journal of the European Union. 2007; **29.9.2007**: L 255/14- L 255/17.
29. **Food and Agriculture Organization and World Health Organization.** Food and Agriculture Organization of the United Nations; Expert Committee on Food Additives Meeting World Health Organization. International Programme on Chemical, Safety. Safety Evaluation of Certain Food Additives and Contaminants, prepared by the Forty-Ninth Meeting of the Joint FAO/WHO Expert Committee on Food Additives (JEFCA) 1998; World Health Organization: Geneva, Switzerland. Available from: <https://apps.who.int/iris/handle/10665/42092> *Date accessed*; 18th December, 2019.



30. **Benkerroum N** Aflatoxins: Producing-molds, structure, health issues and incidence in Southeast Asian and Sub-Saharan African Countries. *International Journal of Environmental Research and Public Health*. 2020; **17**: 1215.
31. **Kenya National Bureau of Statistics (KNBS)**. Enhanced food balance sheets for Kenya (2014-2018 results). Kenya National Bureau of statistics, May 2019. 2019. Available from: <https://www.afdb.org/en/documents/food-balance-sheets-kenya-2014-2018-results> Date accessed; 5th December, 2020.
32. **Sirma AJ, Makita K, Randolph DG, Senerwa D and JF Lindahl** Aflatoxin exposure from milk in rural Kenya and the contribution to the risk of liver cancer. *Toxins*. 2019; **11**: 469.
33. **Grace D** Animals and aflatoxins. In: Unnevehr L and D Grace (Eds). *Aflatoxins Finding solutions for improved food safety*. International Food Policy Research Institute (IFPRI). 2033 K Street, NW, Washington, DC 20006-1002 USA, 2013: 16-17. Available from: www.ifpri.org/publication/aflatoxins-finding-solutions-improved-food-safety Date accessed; 14th November, 2013.
34. **Kigen G, Busakhala N, Kamuren Z, Rono H, Kimalat W and E Njiru** Factors associated with the high prevalence of oesophageal cancer in Western Kenya: a review. *Infectious Agents and Cancer*. 2017; **12**: 59.
35. **Baydar T, Erkekoglu P, Sipahi H and G Sahin** Aflatoxin B1, M1 and ochratoxin A levels in infant formulae and baby foods marketed in Ankara, Turkey. *Journal of Food and Drug Analysis* 2007; **15 (1)**; 89.92.
36. **Mushtaq M, Sultana B, Anwar F, Khan MZ and M Ashrafuzzaman** Occurrence of aflatoxins in selected processed foods from Pakistan. *International Journal of Molecular Sciences*, 2012, **13**; 8324-8337.
37. **Omara T, Kiprop AK, Wangila P, Wacoo AP, Kagoya, S, Nteziyaremye P, Odero MP, Nakiguli, CK and SB Obakiro** The Scourge of Aflatoxins in Kenya: A 60-Year Review (1960 to 2020). *Journal of Food Quality* **2021**; 31 pages.
38. **Mutegi CK, Cotty PJ and R Bandyopadhyay** Prevalence and mitigation of aflatoxins in Kenya (1960-to date). *World Mycotoxin Journal*. 2018; **11(3)**: 341-357.
39. **Zhang J, Orang'o O, Tonui P, Tong Y, Maina T, Kiptoo S, Muthoka K, Gropman J, Smith J, Madeen E, Ermel A, Loehrer P and DR Brown** Detection and concentration of plasma aflatoxin is associated with detection of oncogenic human papillomavirus in Kenyan women. *Open Forum Infectious Diseases*. 2019; 7 pages.



40. **Lombar MJ** Mycotoxin exposure and infant and young child growth in Africa: What do we know? *Annals of Nutrition and Metabolism*. 2014; **64(suppl 2)**: 42–52.
41. **Valavanidis A** Environmental carcinogenic substances, exposure and risk assessment for carcinogenic potential. Classifications and regulations by international and national institutions. 2017; Available from: www.chem-tox-ecotox.org *Date Accessed: 6th June, 2019*.
42. **Sirma AJ, Lindahl JF, Makita K, Senerwa D, Mtimet N, Kang'ethe EK and D Grace** The impacts of aflatoxin standards on health and nutrition in sub-Saharan Africa: The case of Kenya. *Global Food Security*. 2018; **18**: 57-61.

