



Mycological Quality of Relief Maize Meal and Nutritional Supplement of Maize Product (Unimix) in Moyale, Northern Kenya

Samson Chebon^{1*} and Mohamed Asafa Aila²

¹*Department of Medical Microbiology, Jomo Kenyatta University of Agriculture and Technology (JKUAT), Nairobi, Kenya.*

²*Moyale Sub-county Referral Hospital, Marsabit County, Kenya.*

Authors' contributions

This work was carried out in collaboration between both authors. Author SC designed the study, wrote the protocol, wrote the first draft of the manuscript and managed both literature searches and aflatoxin analysis. Author MAA collected maize samples, also undertook both fungal and aflatoxin screening. Both authors read and approved the final manuscript.

Article Information

DOI: 10.9734/MRJI/2017/28684

Editor(s):

(1) Laleh Naraghi, Plant Disease Research Department, Iranian Research Institute of Plant Protection, Tehran, Iran.

Reviewers:

(1) Margarita Ester Laczeski, National University of Misiones, Argentina.

(2) Ogori A. F., Federal University, Nigeria.

Complete Peer review History: <http://www.sciencedomain.org/review-history/20492>

Original Research Article

Received 30th July 2016

Accepted 19th November 2016

Published 14th August 2017

ABSTRACT

Aims: Determination of mycological quality of relief maize meal and Nutritional supplement of maize product (Unimix) in Moyale, Northern Kenya as regards aflatoxin producing fungi and aflatoxin Contamination.

Study Design: Purposeful sampling technique was applied targeting homesteads depending on relief food supplies.

Place and Duration of Study: Homesteads in Manyatta and Burji regions, Moyale town in Northern Kenya were sampling sites. Sampling was undertaken in January-February, 2016 while laboratory analysis was in March-April 2016.

Methodology: A total of 32 samples (maize meal: $n=20$; unimix: $n=12$) were obtained from homesteads and put in clean paper bags ready for transport to Nairobi for laboratory analysis. Mycological analysis was on Potato Dextrose Agar where fungal load was expressed as Colony forming Units (CFU/g) while aflatoxin quantification applied competitive enzyme-linked

*Corresponding author: E-mail: sambonce06@gmail.com;

immunosorbent assay (ELISA) for total aflatoxin levels using Helica Biosystems' kit. Data analysis using student's t-test and Pearson's correlation test was undertaken on various variables namely; mean fungal load, mean aflatoxin levels, frequency of fungi and aflatoxins alongside correlation between fungal and aflatoxin levels ($P=0.05$).

Results: Prevalence of aflatoxin producing fungal spp. among maize meal and Unimix samples were 90% and 0%, respectively with mean fungal load of 288 CFU/g and 27 CFU/g, respectively. The mean fungal load for Unimix samples was 26.7 ± 9.9 CFU/g while for maize meal samples it was 287.7 ± 83.2 CFU/g. Incidence of aflatoxins was in 100% and 91.7% of maize meal and Unimix samples, respectively with the mean contamination levels 6.6 ppb and 3.1 ppb, respectively thereby demonstrating that the difference was not significantly different ($P=0.05$). Similarly, a negative correlation coefficient ($r^2 = -0.212$) was established between fungal and aflatoxin contamination in maize meal ($P=0.05$). The prevalence of other potential mycotoxigenic fungal spp., *Fusarium* and *Penicillium* exhibited similar contamination patterns having been 100% among Maize meal samples, respectively while Unimix lacked any contamination.

Keywords: Mycological; aflatoxins; relief; maize; Unimix; Moyale; Kenya; Ethiopia.

1. INTRODUCTION

Maize is Kenya's principle staple food crop, providing substantial caloric intake to most urban and rural households of diverse socio-economic spectrum. It is estimated that more than 40% of both rural and urban diets consist of maize and maize products with a per capita consumption of about 100 Kg [1,2]. The annual national production is approximately 33.3 million 90 Kg bags (3.0 million metric tons) with production largely concentrated in Rift Valley Province which accounts for more than 50 percent of the national maize production. Trans-Nzoia, Uasin-Gishu, Elgeyo-Marakwet and Nandi counties of Rift Valley Province have the highest per capita yield [3]. This production is mainly under rain-fed farming system. However, with 70% of the country being arid and semi-arid with annual rainfall averages of 400 mm, the national maize production hardly attains the national per capita consumption. This deficit compromises the national food safety whereby in a bid to avert hunger, microbiologically low quality maize meal forms regular affordable diet among most households [4,5].

Marsabit county of Northern Kenya where Moyale town is located is non-agricultural region due the ambient high temperatures, low rainfall and infertile soils. It is one of the driest counties of Kenya, with temperatures as high as 30.2°C during the hot months. Annual rainfall ranges between 200-1000 mm, with the average precipitation being 254 mm. Therefore, the local population relies heavily on maize produced in Kenyan highlands supplied in form of relief food supply and commonly stored indoors. Additional food supply is from Ethiopia where its proximity

with the larger Ethiopian Moyale town allows for uncontrolled cross border trading in substandard uninspected foodstuffs. This scenario predisposes the local population to dietary aflatoxin chronic exposure particularly under conditions of informal food trading with ambient high temperatures favorable for proliferation of aflatoxin producing fungi.

Previously, maize samples from other semi-arid regions of Kenya, particularly lower and upper Eastern Province have been found contaminated with dangerously high aflatoxins levels due to among others; ambient conditions ideal for fungal growth, limited foods diversity, poverty and unsound farming practices [6,-12]. In a comparative study, factors including high levels of aflatoxigenic fungi in soils and the semi-arid climatic conditions of Kitui, Machakos and Makueni counties were found to correlate with the high levels of aflatoxins in maize from these regions compared to areas with humid and abundant rainfall including Uasin-Gishu, Nandi, Trans-Nzoia, Nyeri, Bungoma and Siaya counties where contamination levels were lower [10].

The northern frontier regions of Kenya including North Eastern have climatic conditions much more harsh than Eastern province, a region historically associated with recurrent epidemiological episodes of aflatoxicoses [7,13]. Fundamentally, these conditions not only exacerbate food insecurity through inability to practice farming, it also reduces food availability and economic empowerment of the residents as little home-grown food crops are available for domestic consumption and commercial trading, respectively. The relationship between the two

variables: economic empowerment and food security on one hand, and increased likelihood of aflatoxin exposure was demonstrated in a recent study aimed at establishing the correlation between aflatoxin exposure levels (measured in serum samples) and the socio-economic status of pregnant mothers alongside those with children under 24 months of age. Aflatoxin levels among poorest women were 5-7 times higher compared to the least poor [11]. These alarming findings from Meru and Tharaka-Nithi regions of upper Eastern Province complement observations by Malusha [12], whereby higher household aflatoxin levels in maize samples were correlated with lower household socio-economic and food security index in Kibwezi and Kilome sub-counties of Makueni County, both in lower Eastern Province.

These observations, together with the fact that aflatoxin biosynthesis is enhanced by stressful periods, particularly high temperatures and prolonged low moisture content [14], strongly imply that despite no serious incidence of aflatoxicoses, the combinations of conditions predisposing fungal proliferation and subsequent human dietary aflatoxin exposure do perfectly exist in Moyale. This possibility has previously been proven when Unimix supplement diet, produced by Proctor and Allan Co. and distributed to the local residents by Kenya Red Cross Society (KRSC) were found with aflatoxin contamination thereby occasioning recall of 28 tons [15]. This recall clearly illustrates both the economic and health burdens arising from aflatoxin contamination of foods [16].

The prevailing climatic and socio-economic conditions in Moyale could only be some of the factors likely to contribute to dietary chronic aflatoxin exposure among the local population. The phenomena implies that food supply has to either come from the other agriculturally rich regions of either Kenya, Ethiopia or both considering the far-flung boarder-point location of the town. This poses great food safety challenge because toxin contamination and production can occur throughout the food value chain supply system: from the field, during harvesting, processing, storage, transportation and consumption. The maize meal could therefore harbor storage mycotoxigenic fungi and mycotoxins by the time it reaches consumption stage [17]. The distance from Nairobi to Moyale is about 800 Km. This together with the additional distance of approximately 600 Km from North Rift Valley Province, the Kenyas'

grain basket, predisposes maize to prolonged period of grain handling likely to exacerbate aflatoxin contamination.

The other major maize supply source of maize to Moyale is from Ethiopia whereby unlike Kenya, despite no documented history of previous history of acute aflatoxicosis, findings on mycological quality of foods including maize, sorghum and teff have shown significant fungal and aflatoxin contamination, notwithstanding [18,19,20]. Fungal infection and aflatoxin contamination findings of maize collected from Gedeo zone indicated >50% of samples had aflatoxin concentration of >50 ppb with mean concentration of aflatoxin of 53 ppb and 52.1 ppb as determined by immuno chromatographic assay and thin layer chromatography, respectively [18]. Fungal prevalence was: *A. flavus* (64%), *A. parasiticus* (11%), *Fusarium* spp. (11%) and *Penicillium* spp (8%). Studies on effect of storage time regarding incidence of *Aspergillus* spp. and aflatoxin levels in Sorghum in Ethiopia also indicated total aflatoxin levels reached a high of 344 ppb after a two year period [19]. Findings on aflatoxin contamination of maize from Ethiopia similarly detected 88% prevalence with highest concentrations of 27 ppb [20]. Species of *Aspergillus*, *Fusarium* and *Penicillium* occurred in 94%, 76.5% and 64% of the samples in this study, respectively. These findings indicating comparatively lower prevalence of aflatoxins and aflatoxin producing fungi in maize from Ethiopia relative to those from Kenya could partly be due to its being third staple local staple food after wheat and teff [18,20].

It is therefore on the account the prevailing climatic and soil conditions unfavorable for crop cultivation besides the poor socio-economic status in Moyale that food supply in form of relief maize meal and unimix cushions the populations against foods insecurity. This study therefore aimed to determine mycological quality of relief maize meal and nutritional supplement of maize product (Unimix) in Moyale, Northern Kenya as regards aflatoxin producing fungi and aflatoxin Contamination.

2. METHODOLOGY

2.1 Study Area

Moyale is a market town on the border of Ethiopia and Kenya, whereby it is split between the two countries: the larger portion is in Ethiopia

(in the Oromia region), and the smaller is in Marsabit county, Kenya.

2.2 Sampling Method

Purposeful sampling technique was applied in identifying homesteads from which the food samples were obtained. Selected households in Moyale and Manyatta Burji were identified in Moyale using Multiple Indicator cluster survey data of Kenya National Bureau of Statistics, KNBS that indicates about 85% of households are registered for food Relief services with KRCS [21]. The list of food-relief dependent households was obtained from local area administration Chief, and 20 randomly selected households were visited for collection of maize flour samples (500 g). Another 12 samples (packets) of Unimix flour were collected from health care facilities offering food supplements to pregnant malnourished mothers. These samples were put in clean paper bags with proper identification and transported to JKUAT Medical Microbiology Lab, Nairobi for analysis of microbial quality and subsequent total Aflatoxin levels at Government Chemist food quality laboratory.

2.3 Isolation and Identification of Fungal Species

The isolation of fungi from the maize flour and unimix samples was undertaken using modified Potato Dextrose Agar. Fungi isolation was by dilution plating technique recommended for isolation of fungi from powdered foods [22]. Triplicate sub-samples each of 10 g maize flour were diluted in 100 ml of sterile double distilled water and vortexed for 1 min. Dilutions of 1ml suspension were inoculated on a set of triplicate agar plates using sterile pasteur pipette and glass spreader. Plates were then incubated at 30°C for 3-7 days. Fungal load was thereafter established and expressed as Colony Forming Units (CFU/g) at 10^{-1} . The fungal isolates were identified using obverse colony sizes, color, conidial head and conidia surface texture features and reverse pigmentation [23].

2.4 Determination of Total Aflatoxin Levels in Maize Flour and Maize Unimix Samples

Total Aflatoxin levels in maize and maize products were determined by determine by ELISA using an Helica Biosystems' kit. Briefly the procedure involved aflatoxin extraction from 10 g food samples in methanol: double distilled water phase in 70:30 ratio v/v, where the mixture was

shaken for 30 mins before filtration using whatman filter paper. A volume of 50 μ l of both aflatoxin standard and sample extract were added to aflatoxin coated to a polystyrene microplate microwells. The extracted sample and HRP-conjugated Aflatoxin B1 were mixed and added to the antibody-coated microwell. Aflatoxin from the extracted sample and HRP-conjugated Aflatoxin B1 compete to bind with the antibody coated to the microwell. Microwell contents were decanted and non-specific reactants were removed by washing with distilled water severally. An enzyme substrate (TMB) was added upon which color (blue) developed. The intensity of the color was directly proportional to the amount of bound conjugate and inversely proportional to the concentration of Aflatoxin in the sample or standard. Therefore, as the concentration of Aflatoxin in the sample or standard increases, the intensity of the blue color will decrease. An acidic stop solution was added which changes the chromagen color from blue to yellow. The intensity of resulting color, both in the sample extracts and standards was determined by reading absorbance at 450 nm using a spectrophotometer ELISA plate reader (Uniskan II_Labsystems, Finland [24]. Aflatoxin levels was expressed in μ g/kg, equivalent to ppb.

3. RESULTS

3.1 Incidence of Fungi with Regard to Frequency and Fungal Load in Unimix and Maize Samples

The study undertook isolation of fungi from a total of 32 samples (maize meal; $n=20$; Unimix; $n=12$). The results on the prevalence of aflatoxin producing fungi was 0% and 90% for Unimix and maize, respectively. The mean fungal load for Unimix samples was 27 CFU/g while for maize meal samples it was 288 CFU/g (Table 1). Both the mean fungal load and frequency among these two types of foods was significantly different ($P=0.05$).

The fungal load was profiled into five categories namely; <10 CFU/g, $\geq 10 < 100$ CFU/g, $\geq 100 < 500$ CFU/g, $\geq 500 < 1000$ CFU/g, and ≥ 1000 CFU/g. The fungal load profile among Unimix samples was limited to only two ranges namely, ≤ 10 CFU/g and $\geq 10 < 100$ CFU/g, while for maize meal samples, it was spread across the entire spectrum of five categories. Similarly, the highest fungal for Unimix samples was only 80 CFU/g while for maize meal samples, it was 1,286 CFU/g (Table 1).

The prevalence of aflatoxin producing fungi, particularly *A. flavus* and *A. parasiticus* was 0% for both spp. among Unimix samples while for maize samples it was 75% and 25%, respectively (Table 2). In this study, besides the two spp. of aflatoxin producing fungi, other potential mycotoxin producing fungal spp., particularly *Penicillium* and *Fusarium* spp. were predominantly isolated from maize meal rather than Unimix samples with *Penicillium* occurring in all the 20 maize samples (100%) at ≥ 10 CFU/g fungal load. Similarly, *Fusarium* spp. occurred in 50% of maize samples (Table 3: Pate 1). Findings of this study regarding the mycological quality of the two brands of foods demonstrated that the 32 samples were within the national statutory fungal load limit of $\leq 1.0 \times 10^4$ CFU/g stipulated by Kenya Bureau of Standards, (KEBS) [25].

3.2 Incidence of Aflatoxins in Unimix and Maize Meal

A total of 32 samples (maize meal; $n=20$; Unimix; $n=12$) were screened for total aflatoxin levels whereby results demonstrated that while all

maize samples (100%) had aflatoxin contamination, only one unimix sample (8.3%) was free from contamination (Table 4). The mean aflatoxin levels among these two types of foods was however significantly different having been 3.1 ppb and 6.6 ppb for Unimix and Maize meal samples, respectively ($P=0.05$). When the contamination levels were profiled into ranges, five categories were established namely; ≤ 2.0 ppb, $>2.0 \leq 4.0$ ppb, $>4.0 \leq 10.0$ ppb, $>10.0 \leq 100.0$ ppb, >100 ppb. Unimix samples had aflatoxin within the lower limit ranges of: ≤ 2.0 ppb, $>2.0 \leq 4.0$ ppb, $>4.0 \leq 10.0$ ppb at prevalence of 8.3%, 75%, and 16.7%, respectively. In contrast, maize meal samples were on the upper end contamination profiles: including $>4.0 \leq 10.0$ ppb, $>10.0 \leq 100.0$ ppb with frequency of 85% and 15%, respectively (Table 4).

Regarding the proportion of samples at aflatoxin safety levels of ≤ 10.0 ppb recommended by the Kenyan government through KEBS agency, all Unimix levels were within the safety levels while 15% of maize meal samples had unsafe aflatoxin levels of > 10 ppb (Table 4).

Table 1. Incidence of fungi in Unimix and maize meal flour samples

Food type	Aflatoxin producing fungi		Mean fungal load (CFU/g)	Fungal load profile (CFU/g)	Total aflatoxin producing fungi prevalence	
	AF ^a	RF ^b			AF ^a	RF ^b
Unimix $n=12$	AF ^a	RF ^b	26.7±9.9	≥ 1000	0	0.0%
	0	0%		$\geq 500 < 1000$	0	0.0%
				$\geq 100 < 500$	0	0.0%
Maize $n=20$	AF ^a	RF ^b	287.7±83.2	$\geq 10 < 100$	6	50%
				< 10	6	50.0%
				≥ 1000	2	10%
	18	90%		$\geq 500 < 1000$	2	10%
				$\geq 100 < 500$	8	40%
				$\geq 10 < 100$	6	30%
		< 10	2	10%		

AF^a Absolute frequency; RF^b Relative frequency; © Highest Fungal load

Table 2. Frequency of aflatoxin producing fungi in Unimix and maize flour

Food type	Samples (%)					
	Total fungi		<i>A. flavus</i>		<i>A. parasiticus</i>	
	AF ^a	RF ^b	AF ^a	RF ^b	AF ^a	RF ^b
Unimix $n=12$	0	0.0%	0	0.0%	0	0.0%
Maize $n=20$	18	90%	15	75%	3	15%

AF^a Absolute frequency; RF^b Relative frequency

Table 3. Fungal load profile of *Penicillium* and *Fusarium* spp in Unimix and maize flour samples

Food type	Mean		Contamination	Potential mycotoxigenic spp.			
				<i>Penicillium</i> spp.		<i>Fusarium</i> spp.	
				AF ^a	RF ^b	AF ^a	RF ^b
Unimix n=12	0 CFU		≥1000	0	0.0%	0	0.0%
			≥500<1000	0	0.0%	0	0.0%
	©	0	≥100< 500	0	0.0%	0	0.0%
			≥10<100	0	0.0%	0	0.0%
			<10	0	0.0%	0	0.0%
Maize n=20	104.32CFU		≥1000	1	5.0%	0	0.0%
			≥500<1000	8	40.0%	0	0.0%
	©	1000	≥100< 500	8	40.0%	3	15.0%
			≥10<100	3	15.0%	7	35.0%
			<10	0	0.0%	10	50.0%

AF^a; Absolute frequency; RF^b; Relative frequency; © Highest Fungal load (CFU/g)

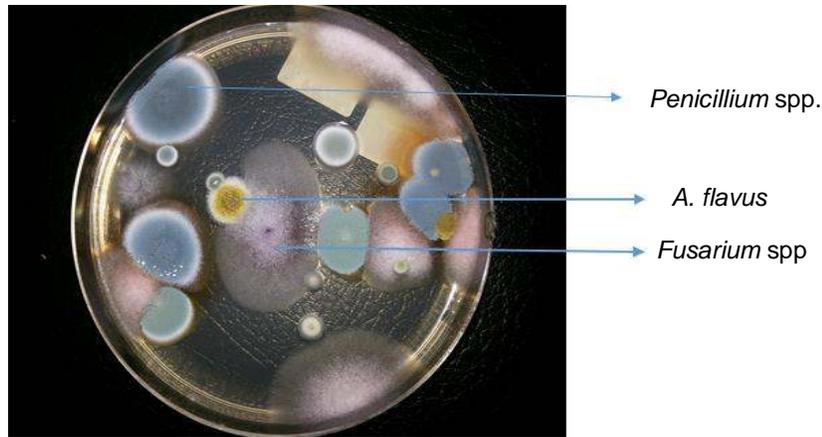


Plate 1. Mixed cultures of fungi isolated from maize flour from Moyale in PDA at 10⁻¹ after 7-day incubation.

Table 4. Aflatoxin load profiles (ppb) for maize meal and Unimix samples from Moyale

Food type	Mean ppb	Aflatoxin levels (ppb)										Max. level ppb
		≤2.0		>2.0≤4.0		>4.0≤10.0		>10.0≤100.0		>100.0		
		AF ^a	RF ^b	AF ^a	RF ^b	AF ^a	RF ^b	AF ^a	RF ^b	AF ^a	RF ^b	
Unimix n=12	3.1±0.7	1	8.3%	9	75%	2	16.7 %	0	0.0 %	0	0 %	4.23 ppb
Maize (n=20)	6.6±0.5	0	0.0%	0	0%	17	85.0%	3	15.0 %	0%	0 %	12.98 ppb

3.3 Correlation between Fungal Load and Aflatoxin Levels

This study attempted to establish the correlation between fungal loads for aflatoxin producing fungal spp. and aflatoxin levels among samples

from the two brands of foods. The results of bivariate Pearson’s correlation however, established a weakly negative correlation coefficient (R²= 0.212) between these two variables relation (P= 0.37).

4. DISCUSSION

4.1 Comparative Incidence of Aflatoxin Producing Fungi and Aflatoxins in Unimix and Maize

The findings of this study indicating that maize had significantly higher contamination levels of both aflatoxin producing fungi and aflatoxins compared to Unimix clearly illustrates the differences in the safety practices involved in the production of these foods. The magnitude of these differences is well demonstrated by the complete absence of *Fusarium* and *Penicillium* spp. besides the safety of all Unimix samples at the national statutory aflatoxin threshold safety limit of ≤ 10 ppb [25]. Unimix is industrial based product manufactured by Proctor and Allan Co. whereby being of an international company that targets the export market, quality production standards and food safety regulations are upheld through application Hazard Analysis and Critical Control Points (HACCP) [26].

Unimix is an enriched maize and bean flour designed and used to alleviate malnutrition. The ingredients for its production including maize flour, soy beans, oil, milk powder and sugar alongside vitamins and minerals usually undergo quality tests. These include fungal and aflatoxin tests which assures its safety for use among the target vulnerable populations cohorts, particularly children and pregnant mothers not only in Kenya but neighboring war-torn countries namely: Somali and Southern Sudan. Recent findings from Kisumu [27,28] and also from Meru and Tharaka-Nithi regions of upper Eastern Province [11] and Kibwezi and Kilome sub-counties of Makeni County, both in lower Eastern Province of Kenya [12] indicated the susceptibility of these cohorts of society to aflatoxin food poisoning. In contrast, the production practices of maize under conditions which target local market rather than export markets implies that stringent quality practices are not adhered to along the food value chain [16].

Supply of relief food in form of Unimix among refugees and internally displaced people is similarly undertaken in the western neighboring country of Uganda, albeit by World Food Program (WFP). The findings of a comparative study in Uganda on the quality of locally procured maize and imported maize grain indicated that despite the lower price and availability of locally cultivated maize, it is generally of very poor

quality. The moisture content was not only found to be high, the proportion of discolored, insect-infested and moldy grain was found comparatively higher than imported grain stock from Republic of South Africa. Informal trading in these poor quality maize flour has potentially serious health implications to consumers [29]. Further, despite the stringent WFP quality specifications, standards are occasionally compromised in a bid to avert disruptions in food supplies. The recommended grain moisture content by WFP in Uganda is 14.0% yet in Kenya it is 13.5% [25,30,31]. The study also established that the Ugandan formal maize trading sector has no experience of meeting the higher quality standards demanded by large maize buyers in Kenya where it is a staple diet. This complex scenario, together with the fact that maize is the fourth staple diet in Uganda after bananas, cassava and sweet-potatoes implies the bulk of lower quality and cheaper maize harvests from mainly the maize growing agro-ecological zones in Eastern Uganda may finally reach Moyale, Kenya almost 2,000 Km away through a prolonged informal food supply chain. This could explain the observed high fungal load and aflatoxin prevalence in maize meal from Moyale as established in this study.

During the 2004-2006 tragic aflatoxicoses outbreaks, homegrown maize was attributed in the affected regions of lower Eastern province [1]. Similarly, factors including high levels of aflatoxigenic fungi in soils and the semi-arid climatic conditions of Kitui, Machakos and Makeni counties were found to correlate with the high levels of aflatoxins in maize in these regions compared to areas with humid and abundant rainfall including Uasin-Gishu, Nandi, Trans-Nzoia, Nyeri, Bungoma and Siaya counties where contamination levels were lower [10]. Therefore, with the much more harsh climatic conditions and perennial food insecurity problem in the northern frontier regions of Kenya including North Eastern and specifically Moyale town, foods are more prone to fungal and aflatoxin contamination than in Eastern province, a region historically associated with recurrent epidemiological aflatoxicoses episodes.

Findings of this study indicating that all maize samples (100%) and 91.7% of Unimix samples had aflatoxin contamination clearly demonstrates this scenario. In these findings, despite the comparatively low levels of aflatoxins of maize meal (highest contamination=12.98 ppb) compared to the exceptionally elevated levels

(8,000 ppb) of the 2004-2006 aflatoxin epidemiological period [7,13], chronic dietary ingestion of low aflatoxin dosages has been linked to some cancers, particularly esopharyngeal cancer in North Rift Valley region of Kenya where acute aflatoxicoses outbreaks have never occurred [32,33].

In a critical review on aflatoxins and food safety in African perspective by Milani, [34], Wild & Gong, [35] and Ayale [36], the pervasive problem of chronic aflatoxin exposure was found exacerbated by the prevailing climatic conditions, traditional crop production practices, low awareness levels and exorbitant aflatoxin tests that deters most stakeholders along the maize value chain [37]. The town of Moyale town and the Ethiopia as a country, have never had incidences of acute aflatoxin outbreaks. However, the prevailing climatic conditions in Moyale are favorable for fungal proliferation. The fact that aflatoxin biosynthesis is enhanced by environmentally stressful conditions, particularly high temperatures and prolonged low moisture content [14], strongly implies that despite no serious incidence of aflatoxicoses, the combinations of conditions predisposing fungal proliferation and subsequent dietary exposure do perfectly exist in Moyale. This observation could explain the finding that Unimix, notwithstanding having been manufactured under the rigorous HACCP system, still had a significantly high prevalence of aflatoxin contamination (91.7%). The previous recalling of 28 tons of Unimix in 2011 due to suspected aflatoxin contamination corroborates this finding [15]. However, considering that Unimix labeling has instructions on storage conditions alongside manufacturing and expiry dates, its safety is comparatively assured relative to maize meal.

Manufactured infant foods including UNIMIX can provide consistent nutrient density and digestibility using a wide range of recipes among not only children but also mothers during pregnancy. The fact that these cohorts of population are vulnerable to aflatoxin food poisoning due to their low immunity, implies that these food supplement's ought to be of high quality both nutritionally and microbiologically [11,12]. However, while Unimix was established to be microbiologically of higher quality compared to maize meal in this study, the fact that only these two cohorts among the wider population are enlisted to the UNIMIX ration supply leaves the larger segment of population to depend on the informally traded low quality

maize meal as staple diet. This thereby gravely predisposes the greater society to not only possible chronic aflatoxin dietary exposure but also intoxication from *Fusarium* and *Penicillium* toxins, these two species having predominantly been isolated from maize meal in the current study.

4.2 Incidence of *Fusarium* and *Penicillium* spp. in Relation to Agro-ecological Location of Moyale: Kenyan-Ethiopian Boarder Town

The prevalence of other potential mycotoxin producing fungi namely *Fusarium* and *Penicillium* in maize meal samples at comparatively higher levels than *Aspergillus* spp. in this study could be attributed to the geographical location of Moyale town at Kenya-Ethiopia boarder. The different agro-ecological zones of maize-producing areas both in Kenya and Ethiopia together with the geographical distance from Moyale implies that maize is transported for long distances (over 1,600 km) thereby predisposing it to prolonged periods under the hot and humid confinement of packaging material and vehicles metallic containers favorable for fungal growth [38]. Further, the findings indicating prevalent contamination of maize in the major agro-ecological zones both in Kenya [39,40] and Ethiopia [20], by these two fungi together with the aflatoxin contamination of maize [18,41,42,43, 44] implies that maize could have higher contamination upon reaching Moyale. Findings by Ayale [20], regarding mycotoxins alongside surface and internal fungi of maize from Ethiopia similarly established that *Aspergillus*, *Fusarium* and *Penicillium* occurred in 94%, 76.5% and 64% of the samples, respectively. Similarly, the level of total aflatoxins, AFB₁, AFB₂, AFG₁ and AFG₂ in sorghum in Ethiopia during a two year storage study period increased to maximum of 344.26 µg/kg, 153.72 µg/kg, 91.82 µg/kg, 139.64 µg/kg and 52.02 µg/kg, respectively [19]. In a similar study in Nigeria on total aflatoxin levels and fungal contamination in maize and maize products by Sule et al. [45], old maize had a comparatively higher mean total aflatoxin level of 177 ppb while new maize had a mean 102 ppb.

This study aimed to determine mycological quality of relief maize meal and nutritional supplement of maize product (Unimix) in Moyale, Northern Kenya as regards aflatoxin producing fungi and aflatoxin contamination. The interesting findings that *Penicillium* and *Fusarium* spp. occurred exceptionally at higher prevalence than

the targeted fungal spp., particularly, *A.parasiticus* implies that dietary co-ingestion of *Penicillium* and *Fusarium* mycotoxins including ochratoxins, fumonisins, trichothenes and zearalenone alongside aflatoxins could be an unfortunate possibility among the residents, notwithstanding this study not having undertaken tests for presence of some of these mycotoxins. This poses a serious public health challenge, particularly considering that acute mycotoxicoses often remain unrecognized and misdiagnosed by medical professionals, except when large numbers of people are clinically affected [46].

5. CONCLUSION

The maize samples obtained from Moyale were significantly contaminated with both aflatoxin producing fungi and aflatoxins than Unix. Due to the observed higher prevalence (100%) of aflatoxins alongside the comparatively higher prevalence of *Penicillium* and *Fusarium*, the residents are at higher risk of dietary co-ingestion of multiple mycotoxins. Urgent educational intervention measures including public awareness campaigns on importance of hygienic grain storage alongside tests for other mycotoxins would help abate the problem. Cross border regular food quality surveillance is also important to forestall any likelihood of acute aflatoxin poisoning given the hot conditions favorable for fungal growth and aflatoxin biosynthesis in Moyale.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Mwhia JT, Straetmans M, Ibrahim A, Njau, J, Muhenje O, Guracha A. Aflatoxin levels in locally grown maize from Makueni District, Kenya. East African Medical Journal. 2008;85(7):311–317.
2. Nkonge C. Highlight of maize aflatoxin research in Kenya. Round table of Aflatoxin expert meeting. Brussels, Belgium; 2016.
3. USAID-KAVES. Maize value chain analysis. USAID-KAVES, Kenya. Fintrac In. Washington, DC; 2014.
4. Azziz-Baumgartner E, Lindblade K, Gieseke K, Rogers HS, Kieszak S, Njapau, et al. Case-control study of an acute aflatoxicosis outbreak, Kenya, 2004. Environmental Health Perspectives. 2005; 1779–1783.
5. Moturi KN. Factors likely to enhance mycotoxin introduction into the human diet through maize in Kenya. African Journal of Food Agriculture Nutrition and Development. 2008;8(3):265–277.
6. Shephard GS. Aflatoxin and food safety: Recent African perspective; In Aflatoxin and food safety. Taylor and Francis Group, LLC. CRS Press, New York; 2005.
7. Probst C, Njapau H, Cotty PJ. Outbreak of an acute aflatoxicosis in Kenya in 2004: Identification of the causal agent. Applied and Environmental Microbiology. 2007; 73(8):2762–2764.
8. Musyoka KR. Diversity of food consumption by household and their exposure to aflatoxins from maize products in Kibwezi district, Makueni County. Masters thesis, department of crop protection, University of Nairobi. Nairobi, Kenya; 2012.
9. Muthomi JW, Njenga LN, Gathumbi JK, Chemining'wa GN. The occurrence of aflatoxins in maize and distribution of mycotoxin-producing fungi in Eastern Kenya. Journal of Plant Pathology. 2009;8: 113–119. Available:<http://scialert.net/abstract/?doi=pj.2009.113.119>
10. Muthomi JW, Mureithi BK, Chemining'wa, GN, Gathumbi JK, Mutitu EW. *Aspergillus* species and Aflatoxin B1 in soil, maize grain and flour samples from semi-arid & humid regions of Kenya. International Journal of Agri Science. 2012;2(1):22–24.
11. Leroy JL, Wang J, Jones KM. Serum aflatoxin B1-lysine adduct level in adult women from Eastern Province in Kenya depends on household socio-economic status, A cross-sectional study. Social Science and Medicine. 2015;146:104–110.
12. Malusha JM, Karama M, Makokha A. The influence of household socio-economic characteristics and awareness on aflatoxin contamination of maize in Makueni County, Kenya. East African Medical Journal. 2015;92:5.
13. Daniel JH, Lewis LS, Rybak ME, Paliakov EM, Kim AA. Human aflatoxin exposure in Kenya, 2007: A cross-sectional study. Food Additives & Contaminants: Part A. 2013;30(7):1322–1331.
14. Medina A, Rodriguez A, Magan N. Effect of climate change on *Aspergillus flavus* and

- aflatoxin B₁ production. *Frontiers in Microbiology*. 2014;5:348.
15. UNIX Recalled from Moyale. Sunday Nation newspaper; 2011.
 16. Wu F, Norrad C, Tiongco M, Liu Y. The health economics of aflatoxin: Global burden of Diseases. Working Paper No. 4: IFRI; 2011.
 17. Kang'ethe EK. Situation analysis: Improving food safety in the maize value chain in Kenya. FAO; 2011.
 18. Nitin M, Chauhan H, Alemayehu P, Washe, Tesfaye M. Fungal infection and aflatoxin contamination in maize collected from Gedeo zone, Ethiopia. *Springerplus*. 2016;5(1):753.
 19. Weledesemayat GT, Gezmu TB, Woldegiorgis AZ, Gemede HF. Study on *Aspergillus* species and aflatoxin levels in sorghum (*Sorghum bicolor* L.) Stored for different period and storage systems in Kewet Districts, Northern Shewa, Ethiopia. *Journal of Food Science & Nutrition*. 2016; 2:10.
 20. Ayale WA. Mycotoxins and surface and internal fungi of maize from Ethiopia. *African Journal of Food, Agriculture, Nutrition and Development*. 2010;10(9): 4109-4123.
 21. KNBS. Kenya - Eastern Province, moyale district multiple indicator cluster survey; 2008.
Available: <https://www.google.com/webhp?sourceid=chromeinstant&ion=1&espv=2&ie=UTF-8#q=knbs%202008%20moyale>
 22. Pitt JI, Hocking AD. Fungi and food spoilage. New York: Springer. 2009;519.
 23. Rodrigues P, Soares C, Kozakiewicz Z, Paterson RRM, Lima N, Venancio A. Identification and characterisation of *Aspergillus flavus* and aflatoxins. In *Communicating current research and educational topics and trends in applied microbiology*. FORMATEX Publications, Barcelona, Spain. 2007;527-534.
 24. Lee NA, Wang S, Allan RD, Ivan R, Kennedy IR. A rapid aflatoxin B1 ELISA: Development and validation with reduced matrix effects for peanuts, corn, pistachio, and soybeans. *Journal of Agriculture Food and Chemistry*. 2004;52:2746–2755.
 25. KEBS. Specifications of maize and maize products. KEBS, Nairobi, Kenya; 2009.
 26. Wu F, Guclu H. Aflatoxin regulations in a network of global maize trade. *PLoS ONE*. 2012;7(9):e45151.
DOI: 10.1371/journal.pone.0045151
 27. Obade MI, Andang'o P, Obonyo C, Lusweti F. Aflatoxin exposure in pregnant women in Kisumu County, Kenya. *Current Research Nutrition Food Science*. 2015;3(2).doi.
Available: <http://dx.doi.org/10.12944/CRNF.SJ.3.2.06>
 28. Obade MI, Andang'o P, Obonyo C, Lusweti F. 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.
 29. Wandscheinder T, Hodges R. Local food aid procurement in Uganda a case study report for EC PREP (UK Department for International Development). Natural Resources Institute, Chatham Maritime Kent, UK; 2005.
 30. Kaaya NA, Muyanja C. Factors associated with fumonisin contamination of maize in Uganda. *Journal of the Science of Food and Agriculture*. 2009;89(14):2393–2398.
 31. Mastersa WA, Kuwornub J, Sarpong D. Improving child nutrition through quality certification of infant foods: Scoping study for a randomized trial in Ghana. Working paper 10/0828. International Growth Centre. London, UK; 2011.
 32. Parker RK, Dawsey SM, Abnet CC, White RE. Frequent occurrence of esophageal cancer in young people in Western Kenya. *Diseases of the Esophagus*. 2010; 23(2):128–135.
 33. Patel K, Wakhisi J, Mining S, Mwangi A, Patel R. Esophageal cancer, the topmost cancer at MTRH in the Rift Valley, Kenya, and its potential risk factors. *ISRN Oncology*; 2013.
 34. Milani M. Aflatoxin and food safety: Recent African perspective; In *Aflatoxin and food safety*. Taylor and Francis Group, LLC. CRS Press, New York; 2013.
 35. Wild CP, Gong YY. Mycotoxins and human diseases: A largely ignored global health issue. *Carcinogenesis*. 2010;31(1):71–82.
 36. Ayale WA. Improving health, trade and food security through regional efforts to mitigate aflatoxin contamination: Regional workshop report on the aflatoxin challenge in Eastern and Southern Africa Theme: 11-13 March 2014, Golden Peacock Hotel, Lilongwe, Malawi; 2014.
 37. Guchi E, Ayalew A, Dejene M, Ketema M, Asalf B, Fininsa C. Occurrence of *Aspergillus* species in groundnut (*Arachis hypogaea* L.) along the value chain in

- different agro-ecological zones of eastern Ethiopia. *Journal of Applied & Environmental Microbiology*. 2015;2(6): 309–317.
38. Hell K, Cardwell KF, Poehling HM. Relationship between management practices, fungal infection and aflatoxin for stored maize in Benin. *Journal of Phytopathology*. 2003;151(11–12):690-698.
39. Njoroge KS. Occurrence of *Fusarium* species and *Fumonisin* mycotoxins associated with maize in Makueni and Makindu counties, Kenya food safety Summit. Africa-Conference; 2015.
40. Kibe EN. Occurrence of mycotoxigenic fungi in maize from food commodity markets in Kenya. (Masters Thesis, Department of Crop Protection, Ghent University). Ghent, Belgium; 2015.
41. Odhiambo BO, Murage H, Wagara IN. Isolation and characterization of aflatoxigenic *Aspergillus* species from maize and soil samples from selected counties of Kenya. *African Journal of Microbiology Research*. 2013;7(34):4379–4388.
42. Okoth S, Nyongesa B, Ayugi V, Kang'ethe E, Korhonen H, Joutsjoki V. Toxigenic potential of *Aspergillus* species occurring on maize kernels from two agro-ecological zones in Kenya. *TOXINS*. 2012;4(11): 991–1007.
43. Sirma AJ. Sources and levels of human exposure to aflatoxins in Nandi County, Kenya. (M.Sc. Thesis. department of public health, pharmacology and toxicology, university of Nairobi). Nairobi, Kenya; 2014.
44. Ouko EO. Sources and levels of human exposure to aflatoxins in Makueni county, Kenya. (M. Sc. Thesis, department of public health, pharmacology and toxicology, university of Nairobi). Nairobi, Kenya; 2014.
45. Sule EI, Orukotan A, Ado A, Adewumi AAJ. Total aflatoxin level and fungi contamination of maize and maize products. *African Journal of Food Science and Technology*. 2015;6(8):229-233. (ISSN: 2141-5455)
46. Khlangwiset P, Shephard GS, F. Wu. Aflatoxins and growth impairment: A review. *Critical Review Toxicology*. 2011;41(9):740-755.

© 2017 Chebon and Aila; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:
The peer review history for this paper can be accessed here:
<http://sciencedomain.org/review-history/20492>