



# Prevalence of aflatoxin, ochratoxin and deoxynivalenol in cereal grains in northern Uganda: Implication for food safety and health

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## ABSTRACT

Mycotoxin contamination of cereals is a significant health risk for humans and animals, particularly in developing countries. To gain insight into food safety related to agricultural practices, we assessed levels of mycotoxin contamination in 105 samples of food grains raised and stored for consumption by rural households in the post-conflict districts of Kitgum and Lamwo in Northern Uganda. Aflatoxin, ochratoxin and deoxynivalenol (DON) contamination was assessed by quantitative enzyme-linked immunosorbent assay. Total aflatoxin in the foods analyzed varied from nd (not detected) to 68.2 µg/Kg. Ochratoxin ranged from 0.1 to 16.4 µg/Kg. DON ranged from nd to 2606 µg/Kg. The mean concentration of total aflatoxins was significantly higher ( $P = 0.002$ ) in sorghum than in millet, maize and sesame seeds. Frequency of co-occurrence of two mycotoxins ranged from 8.3 to 100%, with the highest being aflatoxin and ochratoxin in sorghum. Co-occurrence of all three mycotoxins ranged from 8.3 to 35.3%, with the highest again being in sorghum. Mean levels of aflatoxins concentration in sorghum samples were 11.8 µg/Kg, exceeding the Ugandan national regulatory limits of 10 µg/Kg. Furthermore, 46.5% of the sorghum consumed in both districts exceeded this limit, and 86.1% of sorghum samples exceeded the European Union (E.U.) maximum tolerable limit of 4 µg/Kg. The Estimated Daily Intake (EDI) and Hazard Indices (HI) values were in the range of  $1.2 \times 10^{-5}$ –91.521 and  $1.3 \times 10^{-7}$  to 0.0059, respectively. In conclusion, our results provide evidence of high levels of mycotoxin contamination and co-occurrence in food grains in Northern Uganda with aflatoxins and ochratoxins at high levels in all the cereal types analyzed. Consumption of cereals cultivated in this region poses no health risk of mycotoxins exposure since HI values obtained were less than 1.

## 1. Introduction

Mycotoxins are secondary metabolites produced by fungi of the genera *Penicillium*, *Aspergillus* and *Fusarium* growing on grains and other agricultural products before harvest, during transportation and in storage [1]. Mycotoxin contamination reduces the quality and nutritional value of food, resulting in economic losses to smallholder farmers, traders and consumers [2]. When ingested, inhaled or absorbed through the skin, mycotoxins may cause liver disease, immune deficiency, toxicity, carcinogenicity, growth retardation and death in animals and humans [3–7].

In sub-Saharan Africa, mycotoxin contamination in foodstuff is a

serious public health issue with over 250,000 hepatocellular carcinoma-related deaths occurring annually as a result of aflatoxin alone [8]. In north-eastern Kenya, 125 deaths from consumption of contaminated maize were recorded in 2004 [9]. Mycotoxin contamination of foods and other agricultural products depends on storage, microclimatic condition and harvesting techniques [10].

Several mycotoxins exist and the ones of public health importance include aflatoxins, deoxynivalenol (DON or vomitoxin), zearalenone, ochratoxins, T-2 toxin, fumonisins, and T-2-like toxins [8]. Currently, there are more than 20 aflatoxins known but the six predominant ones are aflatoxin B1 (AFB1), aflatoxin B2 (AFB2), aflatoxin G1 (AFG1), aflatoxin G2 (AFG2), aflatoxin M1 (AFM1) and aflatoxin M2 (AFM2)

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[11]. In the developed countries, the tolerance limits for total aflatoxins range from 0 to 50 µg/Kg, while those of aflatoxin B1 in foodstuff is at 0 to 30 µg/Kg [10].

Information on the prevalence and human health risk of mycotoxins in most African countries is still lacking. This is due to combination of limited monitoring systems and lack of public awareness with regard to mycotoxins. Thus, addressing mycotoxin contamination should be given a priority in sub-Saharan Africa. Determination of mycotoxin contamination is an essential first step in understanding and addressing the mycotoxin problem in Africa. This will provide information needed for assessing risks of key mycotoxins and in identifying intervention targets.

In northern Uganda, the civil war that started in the mid 1980s to 2006 forced communities into internally displaced people's (IDP) camps and caused a collapse in the agricultural production system. Today, rural communities are still confronted with the problem of food insecurity and poor agricultural practices. The communities process their foods locally in homes and for quality do not employ checks and good food safety practices. The staple foods produced in northern Uganda, especially millet, maize, sorghum, groundnut and sesame, are highly susceptible to infection with toxigenic fungi such as *Penicillium* spp., *Aspergillus* spp. and *Fusarium* spp. We undertook a cross-sectional study to assess the levels of contamination of aflatoxins, ochratoxins and DON in grain-based foods in the post-conflict districts of Lamwo and Kitgum, northern Uganda, using an enzyme-linked immunosorbent assay. The objective was to gain insights into mycotoxin safety of foods and related health risks. We discuss our results with a view to raising public awareness to mycotoxins risk in the region.

## 2. Materials and methods

### 2.1. Study sites

We conducted this study in Kitgum (3°17'20.0"N, 32°52'40.0" E) and Lamwo (3° 32' 0" N, 32° 48'0" E) districts, northern Uganda neighbouring South Sudan. These districts were intensely affected by the Joseph Kony's Lord Resistance Army (LRA) rebellion between the mid 1980's and 2006 [12,13]. The incursion shifted many villagers into internally displaced people's (IDP) camps and distorted social services. The total population in the two districts is 215,904 [14]. The area has rainy and dry seasons with annual rainfall of approximately 1300 mm. With improved security and a reduction in LRA attacks, people in both districts began returning to their villages. Subsistence agriculture is the mainstay of the economy employing up to 98% of the population, with cash crops including tobacco and cotton, and food crops being millet, maize, groundnuts, rice, sorghum, sesame, green vegetables, sunflower, citrus and mangoes, beans, sweet potatoes, cassava and pigeon peas. Livestock such as goats, cattle, pigs, sheep and chickens are also reared and some farmers are involved in bee keeping and fish farming. Majority of households use family labour and rudimentary hand tools such as hoes for cultivation and the foods grown are primarily for home consumption.

### 2.2. Collection of food grain samples

The grain samples were collected from seven villages in Kitgum and Lamwo districts (Table 1) between November 2014 and July 2015. Food samples collected included millet, maize, sorghum and sesame. We sampled cereal grains produced by farmers and used for household consumption from food storage bags in 75 randomly selected households. From each household, 500 g of each cereal grain was sampled and stored separately in a labelled polyethylene bag. Overall, 105 cereal grain samples were collected from the two districts (Table 1). These samples were transported to the Gulu University Bioscience Research Laboratory and stored at 4 °C until mycotoxin extraction and analyses.

**Table 1**  
Sampling sites for cereal grains.

| District | Village       | No. of households visited | No. of food grain samples collected |
|----------|---------------|---------------------------|-------------------------------------|
| Kitgum   | Okidi Central | 11                        | 11                                  |
|          | Lamittumangu  | 19                        | 26                                  |
| Lamwo    | Beyagoya      | 13                        | 29                                  |
|          | Apeyta South  | 5                         | 11                                  |
|          | Apeyta West   | 3                         | 5                                   |
|          | Abam          | 21                        | 21                                  |
|          | Laraba        | 3                         | 3                                   |
| Total    |               | 75                        | 105                                 |

### 2.3. Sample preparation and analyses of mycotoxin

Enzyme-linked immunosorbent assay (ELISA) tests were performed using test kit procedures of Romer Labs Singapore Pte Ltd, to determine the mycotoxin levels within cereal grains. 20 g of each grain sample was ground to fine powder in a blender (IKA, Model M20, Germany). Ochratoxin and aflatoxin were extracted from the samples following manufacturers' procedures using methanol and distilled water (for DON). The supernatant was filtered through Whatman No.1 filter paper and the elute was subjected to competitive ELISA analysis.

Mycotoxin concentrations were quantified optically using a spectrophotometer ELISA microplate reader (MULTISKAN FC, model 357, China) with an absorbance and differential filters of 450 nm and 630 nm, respectively, and extrapolated from standard curves generated for each microplate. The kit detection range for ochratoxin was 2–40 ppb, 4–40 ppb for aflatoxin and 0.25–5.0 ppm for DON. To determine the level of co-occurrence of aflatoxin, ochratoxin and DON for each crop type, we used Microsoft Excel 2013 to identify those samples with these mycotoxins.

### 2.4. Estimation of daily intake and hazard index

The Estimated Daily Intake (EDI) was calculated using the mean levels of aflatoxins, ochratoxin and DON obtained in sorghum, millet, maize and sesame, the daily cereal intakes of 0.82 g/person/day [16] and the mean body weight was 72.3 kg/person as indicated by Kirunda [31]. The EDI (expressed in ng/kg of the body-weight/day (ng/kg bw/day) [17]) was calculated for aflatoxin, ochratoxin and DON as indicated in the formula below:

EDI = Daily Intake (of sample food) × mean level of Aflatoxins or ochratoxin or DON divided by average body weight

The Hazard Index (HI) was determined by dividing the EDI by TD50 (the daily dose (ng/kg/body weight/day) at which 50% of test animals would have developed tumors), divided by a safety factor of 50,000 as described by Ishikawa et al. [18], Ismail et al. [19] and Tsakiris et al. [16].

### 2.5. Statistical analysis

To examine whether the concentrations of each of the three mycotoxins differed among the four crops, we fitted a one-way ANOVA with crop type as fixed factor. Differences among treatment means was assessed using Tukey post-hoc tests, corrected for multiple testing (Bonferroni correction) [20]. Prior to statistical analyses, the concentrations of aflatoxin, ochratoxin, and DON were natural log (x + 1) transformed to improve normality. A *t*-test was used to determine whether or not there are significant differences in the concentrations of aflatoxin, ochratoxin and DON for each crop type between the two districts. All the above analyses were undertaken in SPSS version 23.

**Table 2**  
Mean levels of mycotoxins ( $\pm$  SE) in food grain samples from Lamwo and Kitgum districts separately and data pooled for both districts. Grain samples with different letters in a column indicate significant differences in pair-wise tests (one-way ANOVA followed by Bonferroni corrected Tukey post-hoc test).

| Grain sample                 | Kitgum          |                   |                     |                 | Lamwo             |                     |                 |                   | Pooled data for both districts |                   |                    |  |
|------------------------------|-----------------|-------------------|---------------------|-----------------|-------------------|---------------------|-----------------|-------------------|--------------------------------|-------------------|--------------------|--|
|                              | Total Aflatoxin | Total Ochratoxins | DON                 | Total Aflatoxin | Total Ochratoxins | DON                 | Total Aflatoxin | Total Ochratoxins | Total Aflatoxin                | Total Ochratoxins | DON                |  |
| Sorghum ( $\mu\text{g/Kg}$ ) | 16.0 $\pm$ 3.6b | 4.4 $\pm$ 0.8b    | 253.4 $\pm$ 107.5a  | 9.0 $\pm$ 1.8c  | 3.5 $\pm$ 0.7c    | 361.6 $\pm$ 115.7ab | 11.8 $\pm$ 1.8b | 3.8 $\pm$ 0.5c    | 11.8 $\pm$ 1.8b                | 3.8 $\pm$ 0.5c    | 318.8 $\pm$ 81.4ab |  |
| Maize ( $\mu\text{g/Kg}$ )   | 1.9 $\pm$ 0.9a  | 0.6 $\pm$ 0.3a    | 1092.6 $\pm$ 400.8b | 3.3 $\pm$ 0.8ab | 0.3 $\pm$ 0.1a    | 191.4 $\pm$ 76.0ab  | 2.8 $\pm$ 0.6a  | 0.9 $\pm$ 0.2a    | 2.8 $\pm$ 0.6a                 | 0.9 $\pm$ 0.2a    | 513.3 $\pm$ 185.2b |  |
| Millet ( $\mu\text{g/Kg}$ )  | 2.9 $\pm$ 1.2a  | 1.1 $\pm$ 0.3a    | 325.7 $\pm$ 315.9a  | 4.3 $\pm$ 1.5ab | 1.0 $\pm$ 0.3ab   | 32.0 $\pm$ 31.7a    | 3.9 $\pm$ 1.1a  | 1.1 $\pm$ 0.2ab   | 3.9 $\pm$ 1.1a                 | 1.1 $\pm$ 0.2ab   | 129.7 $\pm$ 106.5a |  |
| Sesame ( $\mu\text{g/Kg}$ )  | 2.4 $\pm$ 1.1a  | 1.5 $\pm$ 0.3ab   | 104.8 $\pm$ 48.4a   | 3.5 $\pm$ 2.9a  | 1.4 $\pm$ 0.2bc   | 284.4 $\pm$ 73.6b   | 3.2 $\pm$ 2.1a  | 1.4 $\pm$ 0.2b    | 3.2 $\pm$ 2.1a                 | 1.4 $\pm$ 0.2b    | 230.5 $\pm$ 55.2b  |  |

### 3. Results

#### 3.1. Aflatoxin contamination

All 105 food samples analysed had traceable amounts of total aflatoxin. This was followed by total ochratoxins while DON was least detected (Table 2).

The overall concentration of total aflatoxins varied significantly among the four crops studied (Pooled data, one-way ANOVA  $F_{3,101} = 18.1$ ,  $P < 0.001$ ; Lamwo,  $F_{3,64} = 7.9$ ,  $P < 0.001$ ; Kitgum,  $F_{3,33} = 14.7$ ,  $P < 0.001$  (Table 2). According to the pairwise tests (for pooled data), the mean concentrations of total aflatoxins was significantly higher in sorghum ( $11.8 \pm 1.8 \mu\text{g/Kg}$ ) than in millet ( $3.9 \pm 1.1 \mu\text{g/Kg}$ ), sesame ( $3.2 \pm 2.1 \mu\text{g/Kg}$ ) and maize ( $2.8 \pm 0.6 \mu\text{g/Kg}$ ) (all  $P < 0.05$ , Table 2). The range of aflatoxins in sorghum was nd (not detected)–68.2  $\mu\text{g/Kg}$ , millet nd–14.8  $\mu\text{g/Kg}$ , maize nd–8.1  $\mu\text{g/Kg}$  and for sesame nd–61.8  $\mu\text{g/Kg}$  (Appendices 1 & 2).

The mean concentration of total aflatoxins in sorghum samples from Kitgum district (mean  $\pm$  SE,  $16.0 \pm 3.6 \mu\text{g/Kg}$ ) was slightly higher than the mean concentration of total aflatoxins in sorghum samples from Lamwo district ( $9.0 \pm 1.8 \mu\text{g/Kg}$ ). However, the mean concentration of total aflatoxins in the other grain samples (maize, millet, sesame) was generally higher in samples from Lamwo than in samples from Kitgum district, though the differences were not statistically significant (Table 2). The 46.5% percent of the sorghum consumed in the two districts had a mean total aflatoxin concentration of 16  $\mu\text{g/Kg}$  that exceeded the Uganda national maximum tolerable or regulatory limits of 10  $\mu\text{g/Kg}$  (Table 3). In comparison to European Union, 86% of the sorghum consumed in the two districts exceeded the tolerable maximum limits of 4  $\mu\text{g/Kg}$ .

#### 3.2. Ochratoxin contamination

The concentration of pooled total ochratoxins also differed significantly among the four crops (pooled data,  $F_{3,101} = 19.7$ ,  $P < 0.001$ ; Lamwo,  $F_{3,64} = 10.2$ ,  $P < 0.001$ ; Kitgum,  $F_{3,33} = 9.5$ ,  $P < 0.001$ ). Like for total aflatoxins, sorghum had the highest mean concentration of pooled total ochratoxins (3.8  $\mu\text{g/Kg}$ ), followed by sesame (1.4  $\mu\text{g/Kg}$ ), millet (1.1  $\mu\text{g/Kg}$ ) and maize (0.4  $\mu\text{g/Kg}$ ). The range of the measured total ochratoxins was from nd–16.4  $\mu\text{g/Kg}$  for sorghum, nd–1.8  $\mu\text{g/Kg}$  for maize, nd–3.2  $\mu\text{g/Kg}$  for millet and nd–3.1  $\mu\text{g/Kg}$  for sesame (Appendices 1 and 2).

The mean concentration of ochratoxins in sorghum samples from Kitgum district was slightly higher than for those from Lamwo district (Table 2). The other samples (maize, millet and sesame) showed differences in the mean concentration of ochratoxins, but these differences were not statistically significant (Table 2). In comparison to European Union limits for ochratoxins, 25.6% of the sorghum consumed in the two districts exceeded the maximum levels of 5  $\mu\text{g/Kg}$  (Table 3).

#### 3.3. Deoxynivalenol contamination

The concentrations of DON also differed significantly among the four crops studied (pooled data,  $F_{3,101} = 4.7$ ,  $P = 0.004$ ; Lamwo,  $F_{3,64} = 4.7$ ,  $P = 0.026$ ; Kitgum,  $F_{3,33} = 3.7$ ,  $P = 0.022$ ). According to

**Table 3**  
Comparison of maximum tolerable limits of mycotoxin with EU market.

| COUNTRY       | MYCOTOXIN                |                          |                          | Reference |
|---------------|--------------------------|--------------------------|--------------------------|-----------|
|               | Total Aflatoxins         | Total Ochratoxins        | Deoxynivalenol           |           |
| EU            | 4 $\mu\text{g/Kg}$       | 5.0 $\mu\text{g/Kg}$     | 750 $\mu\text{g/Kg}$     | [21,22]   |
| Uganda        | 10 $\mu\text{g/kg}$      | –                        | –                        | [23]      |
| Current study | nd–68.2 $\mu\text{g/Kg}$ | nd–16.5 $\mu\text{g/Kg}$ | nd–1904 $\mu\text{g/Kg}$ |           |

**Table 4**

Daily intakes of the various food grains in Uganda.

| Food samples | Daily Intake (Kg/person/day) | References |
|--------------|------------------------------|------------|
| Sorghum      | 0.115                        | [24,25]    |
| Millet       | $1.8 \times 10^{-4}$         | [24,25]    |
| Maize        | 0.106                        | [24,25]    |
| Sesame       | $6.0 \times 10^{-6}$         | [26]       |

pairwise tests (pooled data), the concentrations of DON was significantly higher in maize (513.3 µg/Kg) and sesame (230.5 µg/Kg) than in millet, 129.7 µg/Kg (Table 2). Generally, low concentrations of DON were detected in all the samples with the highest (2606.2 µg/Kg) being in maize samples from Kitgum (Appendix 1). The highest concentrations of DON in all samples from Lamwo district was less than 1738.2 µg/Kg (Appendix 1). The range of DON in sorghum was nd-1738.2 µg/Kg, maize nd-2606.2 µg/Kg, millet nd-1904.4 µg/Kg and for sesame nd-955.3 µg/Kg (Appendices 1 and 2). In comparison to European Union limits, 18.6% of the sorghum consumed in the two districts exceeded the DON maximum tolerable limits of 750 µg/Kg (Table 3).

### 3.4. Human risk assessment of exposure to total aflatoxin, Ochratoxin and DON via consumption of cereals

#### 3.4.1. Daily intake

Daily intakes of foods were 0.115 Kg/person/day for sorghum,  $1.8 \times 10^{-4}$  (millet), 0.106 (maize) and  $6.0 \times 10^{-6}$  (sesame), Table 4.

#### 3.4.2. Hazard Index (HI)

The EDI calculated for total aflatoxin, Ochratoxin and DON for northern Ugandan people via consumption of cereals are presented in Table 5. The EDI values ranged from  $1.2 \times 10^{-5}$ –0.125 for total

**Table 5**

Estimated Daily Intake (EDI) and Hazard Indices (HI) for northern Ugandans via consumption of sorghum, maize, sesame foods.

| Food grain                 | Mean (µg/Kg) | Age                   | Average body weight (kg) | Estimated Daily Intake (EDI (µg/Kg/.bw/day) | Hazard Index (HI)     |
|----------------------------|--------------|-----------------------|--------------------------|---------------------------------------------|-----------------------|
| Total aflatoxin            |              |                       |                          |                                             |                       |
| Sorghum                    | 11.8         | 18 > 65 yrs (adults)  | 72.3                     | $3.1 \times 10^{-3}$                        | $4.7 \times 10^{-8}$  |
|                            |              | 6–59 months (infants) | 11.3                     | 0.125                                       | $1.9 \times 10^{-6}$  |
| Millet                     | 3.9          | 18 > 65 yrs (adults)  | 72.3                     | $5.4 \times 10^{-7}$                        | $8.3 \times 10^{-12}$ |
|                            |              | 6–59 months (infants) | 11.3                     | $2.1 \times 10^{-5}$                        | $3.2 \times 10^{-10}$ |
| Maize                      | 2.8          | 18 > 65 yrs (adults)  | 72.3                     | $1.6 \times 10^{-4}$                        | $2.5 \times 10^{-9}$  |
|                            |              | 6–59 months (infants) | 11.3                     | 0.007                                       | $1.0 \times 10^{-7}$  |
| Sesame                     | 3.2          | 18 > 65 yrs (adults)  | 72.3                     | $1.2 \times 10^{-5}$                        | $1.8 \times 10^{-10}$ |
|                            |              | 6–59 months (infants) | 11.3                     | $4.8 \times 10^{-4}$                        | $7.4 \times 10^{-9}$  |
| Total ochratoxin           |              |                       |                          |                                             |                       |
| Sorghum                    | 3.8          | 18 > 65 yrs (adults)  | 72.3                     | $3.2 \times 10^{-4}$                        | $6.1 \times 10^{-8}$  |
|                            |              | 6–59 months (infants) | 11.3                     | 0.013                                       | $2.5 \times 10^{-6}$  |
| Millet                     | 1.1          | 18 > 65 yrs (adults)  | 72.3                     | $4.1 \times 10^{-8}$                        | $8.0 \times 10^{-12}$ |
|                            |              | 6–59 months (infants) | 11.3                     | $1.7 \times 10^{-6}$                        | $3.2 \times 10^{-10}$ |
| Maize                      | 0.9          | 18 > 65 yrs (adults)  | 72.3                     | $1.6 \times 10^{-5}$                        | $3.2 \times 10^{-9}$  |
|                            |              | 6–59 months (infants) | 11.3                     | $6.7 \times 10^{-4}$                        | $1.3 \times 10^{-7}$  |
| Sesame                     | 1.4          | 18 > 65 yrs (adults)  | 72.3                     | $3.4 \times 10^{-5}$                        | $6.6 \times 10^{-9}$  |
|                            |              | 6–59 months (infants) | 11.3                     | $9.2 \times 10^{-5}$                        | $1.8 \times 10^{-8}$  |
| Deoxynivalenol (Vomitoxin) |              |                       |                          |                                             |                       |
| Sorghum                    | 318.8        | 18 > 65 yrs (adults)  | 72.3                     | 2.236                                       | $5.8 \times 10^{-5}$  |
|                            |              | 6–59 months (infants) | 11.3                     | 91.521                                      | 0.0024                |
| Millet                     | 129.7        | 18 > 65 yrs (adults)  | 72.3                     | 0.006                                       | $1.5 \times 10^{-7}$  |
|                            |              | 6–59 months (infants) | 11.3                     | 0.024                                       | $6.3 \times 10^{-7}$  |
| Maize                      | 513.3        | 18 > 65 yrs (adults)  | 72.3                     | 5.343                                       | $1.4 \times 10^{-4}$  |
|                            |              | 6–59 months (infants) | 11.3                     | 218.72                                      | 0.0057                |
| Sesame                     | 230.5        | 18 > 65 yrs (adults)  | 72.3                     | 0.061                                       | $1.6 \times 10^{-6}$  |
|                            |              | 6–59 months (infants) | 11.3                     | 2.497                                       | $6.5 \times 10^{-5}$  |

TD50 of total Aflatoxin = 1.3 µg/kg [29].

TD50 of total Ochratoxin = 0.103 µg/kg [30].

TD50 of Deoxynivalenol (Vomitoxin) = 0.77 µg/kg [26].

Average body weight of an adult in Uganda = 72.3 kg [31].

Average body weight of infants in Uganda = 11.3 kg [32].

aflatoxin,  $1.6 \times 10^{-5}$ –0.013 (total ochratoxin), and 0.06–91.521 (DON). The recorded HI values were in the range of  $1.3 \times 10^{-7}$  to 0.0059 (Tables 4 and 5). In general, it is accepted that an HI ≤ 1 indicates no significant health risk. Nonetheless, the possibility of long-term adverse health effects increases with increasing HI values as an HI between 1.1 and 10 reflects a moderate risk [27], and HI < 10 indicates high risk [28]. HI value for exposure to aflatoxin, Ochratoxin and DON via consumption of sorghum, maize, millet and Sesame, cereal based foods consumed by the northern Ugandan population is less than one. The aforementioned values imply that intake of cereal-based foods will most likely not pose high risk to health of Ugandan population.

### 3.5. Co-occurrence of mycotoxin

Co-occurrence of the mycotoxins was also observed in the majority of the samples (Table 6). The co-occurrence of aflatoxin and ochratoxin was 86% of sorghum samples, 57.9% of millet samples, 40% of maize samples and 27.6% of sesame samples testing positive for both. Co-occurrence of aflatoxin and DON was found in 34.9% of sorghum, 10.5% of millet, 66.7% of maize and 20% of sesame samples. Co-occurrence of ochratoxin and DON ranged from 37.2% of maize samples, 15.8% in millet, 26.7% of maize to 56.7% of sesame samples. Co-occurrence of all three mycotoxins ranged from not detected in millet samples to as high as 32.6% in sorghum samples (Table 6).

## 4. Discussion

We assessed mycotoxin contamination levels in food grains in northern Uganda in order to gain insight into food safety and good agricultural practices in post-conflict areas of northern Uganda. Our results indicate high level of mycotoxin contamination. Aflatoxin and ochratoxin levels were high in the cereal grains analyzed in both Lamwo and Kitgum districts. The highest concentration of aflatoxin was

**Table 6**  
Frequency of co-occurrence of mycotoxin.

| Sample  | Mycotoxin      | Co-occurrence (%) |
|---------|----------------|-------------------|
| Sorghum | Aflatoxin      | 86.0              |
|         | Ochratoxin     |                   |
|         | Aflatoxin      | 34.9              |
|         | Deoxynivalenol |                   |
|         | Ochratoxin     | 37.2              |
|         | Deoxynivalenol |                   |
| Millet  | Aflatoxin      | 32.6              |
|         | Ochratoxin     |                   |
|         | Deoxynivalenol |                   |
|         | Aflatoxin      | 57.9              |
|         | Ochratoxin     |                   |
|         | Aflatoxin      | 10.5              |
| Maize   | Deoxynivalenol |                   |
|         | Ochratoxin     | 15.8              |
|         | Deoxynivalenol |                   |
|         | Aflatoxin      | 0                 |
|         | Ochratoxin,    |                   |
|         | Deoxynivalenol |                   |
| Sesame  | Aflatoxin      | 40                |
|         | Ochratoxin     |                   |
|         | Aflatoxin      | 66.7              |
|         | Deoxynivalenol |                   |
|         | Ochratoxin     | 26.7              |
|         | Deoxynivalenol |                   |
|         | Aflatoxin      | 20                |
|         | Ochratoxin,    |                   |
|         | Deoxynivalenol |                   |
|         | Aflatoxin      | 26.7              |
|         | Ochratoxin     |                   |
|         | Aflatoxin      | 20                |
|         | Deoxynivalenol |                   |
|         | Ochratoxin     | 56.7              |
|         | Deoxynivalenol |                   |
|         | Aflatoxin      | 13.3              |
|         | Ochratoxin     |                   |
|         | Deoxynivalenol |                   |

recorded in sorghum followed by millet, maize and sesame. Our estimates of total aflatoxin in grains are similar to those obtained in Ethiopia [33], Malawi [22,34], Ghana [36] and in the neighbouring Democratic Republic of Congo [37]. In northern Uganda, sorghum is used for food, local alcohol brewing and source of household income. Small-scale farming households in northern Uganda preferentially grow the crop due to its inherent ability to resist drought. Foods are locally processed at homes with no quality checks and communities lack awareness of food safety concerns. The high levels of mycotoxin we found in these foods indicate the use of poor agricultural practices and implies public ignorance about mycotoxins in the region.

The differences between food grain crops contaminations are probably attributable to poor pre- and post-harvest practices, handling and environmental factors. Lack of difference in mycotoxin contamination between Kitgum and Lamwo districts may be due to similar climatic conditions in the two districts (both of them lie in the same agroecological zone) and the shared agricultural practices among farmers in both districts. These similarities in environment and farming methods likely result in more or less similar levels of mycotoxin observed.

Although ochratoxin and DON are not currently regulated in Uganda, aflatoxin is. Overall, 46.5% of the sorghum samples in Lamwo and Kitgum districts exceeded the Ugandan maximum tolerable limit of 10 µg/Kg. If measured against the E.U. standard of 4 µg/Kg, 86.1% of the sorghum samples exceeded the E.U. limit [21]. However, calculation of the HI value indicated that consumption of cereals poses no health risk to northern Uganda consumers. Open sun-drying after harvesting is a traditional method that can reduce mycotoxin contamination. However, communities and commercial grain producers in northern Uganda need to be educated on the importance of good

agricultural practices and food hygiene to ensure better standards are adhered to minimize mycotoxin contamination at every point during grain production. We suggest implementation of an integrated mycotoxin management system at household level in order to lessen mycotoxin contamination in this region. Mycotoxin prevention focusing on pre-harvest management is likely to be beneficial in reducing mycotoxin contamination.

The occurrence of multiple mycotoxins, even in low concentrations, are likely to worsen toxicity and results in serious health outcomes among the people in northern Uganda. Although low levels of mycotoxins may not result in an immediate observable effects, repeated exposures to multiple mycotoxins over a long period of time may result in detrimental health consequences [38]. Additionally, since it is a common practice by the communities in northern Uganda to regularly drink a lot of local brew prepared from sorghum, this increases their chances of daily exposure to high concentrations of different mycotoxins. This might explain the high incidence of oesophageal cancer in northern Uganda as reported in Alema and Iva [39]. Mycotoxins have been shown to cause several cancer related deaths depending on the duration of exposure. For example, high mortality rate are known to occur at an aflatoxin contamination levels of 6.25–15.6 ppm and at a mean daily consumption intake of 2–6 mg per person [42–44]. Northern Uganda also has the highest prevalence of chronic hepatitis B virus (HBV) in the country standing at 17.6% [45]. Two of the major risk factors to hepatocellular carcinoma are HBV infection and dietary exposure to aflatoxin B1 (AFB1) [46]. Aflatoxin B1 causes cancer by binding to and altering the structure of DNA, resulting in genomic mutation [48]. Ochratoxin A has also been linked to serious human health effects, including human endemic nephropathies, pouty ochratoxicosis, porcine nephropathy and urinary tract tumours [5,49].

Overall, our results provide evidence of high levels of mycotoxin contamination and co-occurrence in food grains in northern Uganda where the population is at high risk of exposure, with aflatoxins and ochratoxins at high levels in all the cereal types analyzed. However, the Hazard Indices recorded values less than 1 and so Ugandan consuming these products were not at risk. There is therefore an urgent need for heightened public awareness and both farmers and consumer education on the importance of exposure to mycotoxins as well as implementation of appropriate methods to reduce mycotoxin contamination in food produced and consumed at the household level. This would be best combined with a monitoring system that can provide feedback on the progress in reducing mycotoxin burden.

### Declaration of Competing Interest

The authors declare that there is no conflict of interest. The research was conducted with no financial conflict or other factors which is considered to be declared as conflict.

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### Appendix A. Supplementary data

Supplementary material related to this article can be found, in the online version, at doi:<https://doi.org/10.1016/j.toxrep.2019.09.002>.

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