

Aflatoxin Contamination of Maize and Groundnuts in Barh-Koh Department, Southern Chad

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Abstract In Chad, maize and groundnut crops are faced with the proliferation of mycotoxins, particularly aflatoxins. The aim of the study is to determine aflatoxins in maize and groundnut and to design a simplified approach to practices to reduce aflatoxin contamination. A survey of cultivation practices likely to contribute to contamination was carried out using a form sent to growers. Followed by analysis of aflatoxins in maize and peanut samples, using the liquid chromatography-mass spectrometry (LC-MS) technique. The survey showed that 45% of growers harvest maize in October. Groundnuts are harvested in October by 39% of growers. The survey revealed that 33% of growers dry maize for two weeks, while 11% of growers dry maize intermittently for more than three weeks. The result showed that 56% of producers dry peanuts for two weeks. The results showed that 56% of maize dryers use tarpaulins, while 26% dry on the ground. The survey showed that 82% of groundnut drying was done on tarpaulins and 6% on the ground. Samples from maize batch M6 recorded a high total aflatoxin content of 380.83 $\mu g/kg$, followed by samples from batches M1 and M9. Low aflatoxin levels were observed in samples from lots M8 (1.05 $\mu g/kg$), M7 (2.10 $\mu g/kg$). Peanut lot A9 samples recorded high aflatoxin levels ($\geq 5.73\mu g/kg$), followed by lot A2 samples with an aflatoxin level of 2.48 $\mu g/kg$. Low levels were observed in samples from lots A10 (0.51 $\mu g/kg$), A5 and A6 (0.52 $\mu g/kg$). Seed treatment with fungicides and harvesting at the end of winter, 14-day drying on tarpaulins and storage in warehouses is proposed to reduce aflatoxin contamination of maize and groundnuts.

Keywords: maize, peanut, aflatoxins, contamination, cultivation practices, Chad

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1. Introduction

In Chad, agriculture is practiced by three-thirds (2/3) of the population and contributes around 23% to the formation of GDP (Gross Domestic Product) [1]. Cereals are the staple food of the Chadian population and are consumed in a variety of dietary forms. With production of 414.606 tonnes in the 2019/2020 crop year[2]. The maize is the main cereal consumed in Chad [3]. The same is true of groundnuts, whose production is estimated at 873,228 tonnes in the 2019/2020 crop year, grown in several of Chad's agro-ecological zones, notably the Sudanian, Sahelian and Saharan (oasis) zones [2]. A staple food for the Chadian population, its raw consumption and/or by-products are said to improve the quality of diets [4]. Given the popularity of maize and peanut crops in Chad among peasant farmers in the various performance zones, both crops are confronted with mycotoxin proliferation. Mycotoxins are toxic to humans and animals under specific environmental conditions [5]. They can develop on plants in the field or during storage [6] and easily contaminate organic matter. Mycotoxin

contamination of foodstuffs is a growing concern. This contamination may evolve simultaneously with variations in agricultural practices and is probably influenced by climate change. Surveys indicate that 70% of the world's production is contaminated by mycotoxins [7]. The Food and Agriculture Organization of the United Nations (FAO), meanwhile, estimates that around 25% of food is significantly contaminated by mycotoxins. In sub-Saharan Africa the World Health Organization (WHO) reports that over 500 million of the poorest people are exposed to dangerous levels of mycotoxins [8]. Mycotoxin contamination leads to increased mortality and morbidity. The maize and groundnut varieties grown come from traditional selection (local varieties) or agronomic research (improved varieties) and are consumed during the conservation period in granaries or storage warehouses. Cultivation practices, storage conditions and variety genetics are the factors that condition mold development. Molds are known to be the main producers of mycotoxins. Their great adaptability to hot, humid environments, their high enzymatic potential and their high level of biochemical synthesis make them veritable metabolite production factories. Mycotoxins are stable to a variety of chemical and physical treatments, and can persist along

production lines, ending up as contaminants in final foodstuffs destined for human and animal consumption. Mycotoxins are responsible for considerable economic loss for farmers, as they lead to lower quality raw materials, lower yields and reduced economic value. Endowed with a worrying toxic capacity, certain genera such as: Aspergillus, Fusaruim, and penicelluim, are often reported as agents responsible for several diseases on maize and peanuts in the field and in stock [9]. The WHO in 2018 considered that it is difficult to detect afflatoxicosis in humans and animals due to the variability of clinical signs and the potential presence of other factors such as immune system depression caused by infectious diseases. Numerous studies also claim that climate change could increase food safety hazards, which is why further research in the field is essential to gain a better understanding of emerging mycotoxins [10]. Beyond the literature on mycotoxin damage to peanuts and maize, Chad is one of the countries whose producers/researchers have not mastered this scourge in two key respects: Farmers-multipliers who produce seeds of improved and local varieties see their products downgraded at the certification analysis laboratory (by the Direction des Semences et Plants) due to poor germination and excessive mold development:

Peanuts sold on the market for consumption (in the form of pasta, meal and oil) come from areas where cultivation practices are thought to favor mold development (setting the variety in relation to its cycle, post-harvest management and conservation).

Several studies have been carried out on mycotoxin contamination of cereals and groundnuts [11-16]. The overall aim of this study is to propose simplified practices that can reduce aflatoxin contamination of maize and groundnuts.

Specifically, the aim is to assess the practices likely to promote mycotoxin contamination of groundnuts and maize, and to determine the aflatoxins produced by fungi in maize and groundnuts in the Bahr-koh department.

2. Materials and Methods

2.1. Location of the Study Area

A survey of cropping practices was carried out among growers in the Barh-koh department.

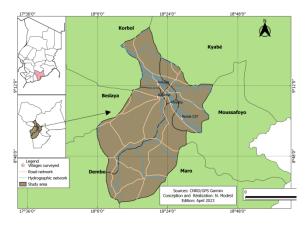


Figure 1. Location map of study area

2.2. Survey

The methodology used is a survey of producers in the study area. Data on practices likely to contribute to mycotoxin contamination of groundnuts and maize are collected using a survey form from growers in three villages in the Barh-Koh Department. Each grower was interviewed on the basis of a questionnaire on the techniques used to select the varieties to be grown, technical itineraries, harvesting, drying and storage periods, harvest losses and use of harvested produce.

Data (qualitative variables) are numerically coded and subjected to descriptive statistical analysis in terms of percentages and averages.

Determining Sample Size

Determining the sample size is an important step in any survey, as it determines the precision of the analysis. In order to implement this survey properly, we carried out cluster sampling on the entire population concerned by this study. This concerns the choice of the three villages, which is based on random sampling, and the population surveyed.

For the purposes of this study, the sample size is determined based on the formula [17], which is written as follows:

$$n = \frac{t^2 p \left(1 - p\right)}{e^2} where$$

n = sample size;

t = confidence level deduced from the confidence rate (traditionally 1.96 for a 95% confidence rate) - centered reduced normal distribution;

p = estimated proportion of the population with the characteristic studied in the study, expressed mathematically as p = n/N.

Given that the number of working people is 89031 and the rural population is 178063, the proportion of p is 50%.

At the significance level $\alpha = 7\%$; t = 1.96. The chosen margin of error is 0.07. The theoretical sample size is therefore 103.

2.3. Plant Material

The plant material consisted of samples of varieties (local and improved) of groundnut and maize collected from growers in the study area.

2.4. Methods

Analysis of aflatoxins in samples Maize and groundnut samples collected from producers in the study area were analyzed for aflatoxins at the CECOQDA laboratory in N'Djamena, using the liquid chromatography-mass spectrometry (LC-MS) technique.

2.5. Statistical Analysis of Data

Statistical analysis was carried out using SPSS (23.0) software for analysis of variance (ANOVA) and for tests of variance the confidence level is 95%.

3. Results and Discussion

3.1. Results of Surveys on Cultivation Practices and Storage of Maize and Groundnuts

The results of the survey on the gender of respondents practising agriculture in the study area are shown in Figure 2.

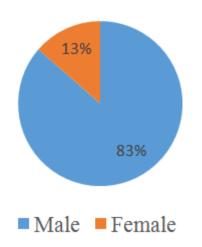


Figure 2. Gender of respondents farming in the study area

The results of our surveys show that 83% of men farm, compared with 13% of women (Figure 2).

The result of the survey on activities practiced by individuals is shown in Figure 3. The result of the survey showed that 33% of the people surveyed practiced agriculture as their main activity. 19% of those surveyed are civil servants and also practise agriculture. The results of the survey also revealed that 16% of individuals practice agriculture and trade as their main activities. 13% of those surveyed farm and fish at the same time. On the other hand, 11% of those surveyed practise farming and livestock breeding as their main activities. 7% of respondents study and practice agriculture as their main activities. The survey revealed that 1% of respondents are religious and practice agriculture.

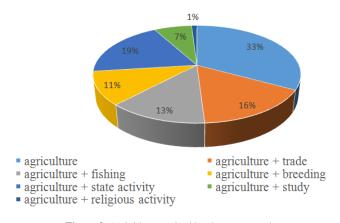


Figure 3. Activities practised by those surveyed

The results of the survey revealed that 74% of farmers produce for their own consumption, using "off-the-shelf" seeds paid for directly on the market, since the genetic identity of these seeds is not known 21% of farmers produce for their own consumption, but using improved seeds. 5% of farmers produce improved seeds for marketing.

The results of the survey revealed that 88% of farmers practise harnessed farming, while 9% use tractors for ploughing and only 3% use manual ploughing.

With regard to corn sowing dates, the survey revealed that 32% of growers sow from May 15 to May 25. 22% of growers from May 25 to June 06, 38% of growers from June 07 to June 17, 5% of growers from June 18 to June 28 and 3% of growers from June 29 to July 9. With regard to peanut sowing dates, 69% of growers surveyed sow from May 15 to May 25.21% of growers sow from May 25 to June 06.12% of growers sow from June 07 to June 17.2% of growers from June 18 to June 28. 2% of growers from June 29 to July 9 and 2% of growers from July 10 to July 20. The results of the survey revealed that 64% of corn seed is not mixed with thioral or other fungicides.

The survey showed that 80% of farmers grow pure maize, while 20% grow mixed crops. As for groundnuts, the result showed that 5% of growers practiced combined cropping and 49% pure cropping.

The results of the survey on the percentage of respondents and harvest periods for maize and groundnuts are shown in Figure 4.

The survey showed that 8% of growers harvest maize in August 47% in September and 45% in October. As for the peanut harvesting period, 3% of growers harvest in August 58% in September and 39% in October.

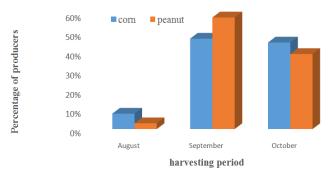
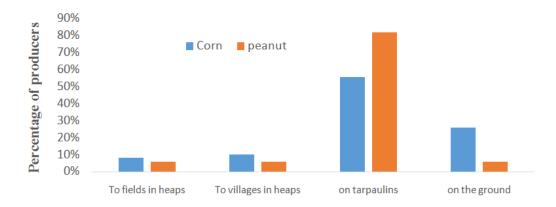


Figure 4. Percentages of respondents and harvest periods for maize and groundnuts

The results of the survey on peanut and maize drying locations are shown in Figure 5.

With regard to maize drying methods, the results showed that 8% of producers dried maize in piles in the field, 10% dried maize in piles in the village, 56% dried maize on tarpaulins and 26% dried maize on the ground. The survey showed that 6% of growers dried groundnuts in piles in the field, 6% dried them in piles in the village, 82% dried them on tarpaulins and 6% dried them on the ground (Figure 5).



Drying sites



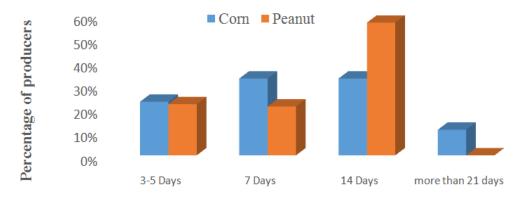






Figure 6. Peanut and maize drying times before storage

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Figure 7. Post-harvest storage location for maize and groundnuts

The results of the survey on peanut and maize drying times before storage are shown in Figure 6.

With regard to maize drying time, the survey revealed that 23% of growers dried for 3 to 5 days, 33% dried for one week, 33% dried for two weeks, and 11% dried for more than three weeks intermittently. As for groundnuts, the results showed that 22% of growers dried for 3 to 5

days, 22% dried for one week and 56% dried for two weeks (Figure 5).

The results showed that 94% of producers dried maize on the cob without spathes, and 6% dried the kernels after shelling. As for groundnuts, the survey revealed that 96% of producers dried with pods, and 4% dried with pods removed (shelled).

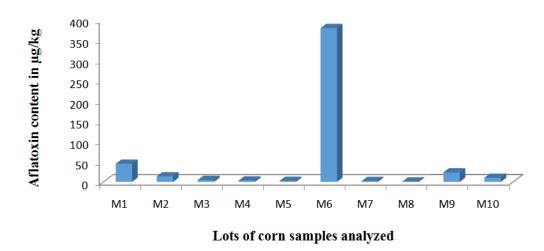


Figure 8. Analysis of aflatoxin-affected maize sample batches

The survey showed that 100% of growers store maize in kernels. As for groundnuts, the result showed that 86% of growers store with pods, and 14% store with groundnut seeds.

The results of the survey on post-harvest storage of maize and groundnuts are shown in Figure 7.

The result showed that 40% of maize was stored in warehouses, 53% in dwellings and 7% in granaries. For groundnuts, 79% were stored in warehouses, 16% in dwellings and 5% in granaries.

With regard to maize storage, the results showed that 89% of growers store maize in bags, 8% in bulk and 3% in drums. As for groundnuts, the result showed that 90% of growers store in bags, while 10% store in bulk.

The results showed that 21% of growers store maize for 1 to 3 months, 51% for 4 to 6 months, 18% for 7 to 9 months and 10% for 10 to 12 months. In the case of groundnuts, the survey revealed that 4% of producers store for two weeks, 32% store for 1 to 3 months, 50% store for 4 to 6 months, 10% store for 7 to 9 months and 4% store for 10 to 12 months.

The survey results showed that 12% of growers treated their stored corn with chemical pesticides, 3% used natural products, and 3% raised cats to chase rodents away from the house where they stored their produce. The survey also revealed that 82% of growers stored their produce without treatment. As for peanuts, the result showed that 5% of growers used chemical pesticides, 12% used natural products, 7% raised cats to chase rodents away from the house where they stored their produce, and 76% stored peanuts without treatment.

The result showed at 100% that mold attack on corn was around 2-10%. As for peanuts, the result showed that 93% of producers had recorded a mold attack of around 2-10%, and 7% had recorded no mold attack at all.

The result of the survey showed that 29% of growers produced maize for consumption, 16% that the products from their harvests were destined for marketing, and 55% that growers used their harvests for consumption and marketing at the same time. As for groundnuts, the result showed that 27% of growers produced groundnuts for consumption, 23% of growers produced groundnuts for marketing, and 50% of growers used these products for both consumption and trade. The results of the survey revealed that 32% of producers sold their maize through third parties, 32% sold their crops directly to traders and 36% sold to consumers. As for peanuts, the results showed that 24% of growers sold their crops through third parties, 30% sold to traders and 46% sold to consumers.

The result showed that 46% of customers preferred the variety, 43% preferred undamaged seeds and 11% preferred non-moldy seeds. As for peanuts, the result revealed that 56% of customers preferred the variety, 19% preferred undamaged seeds, 15% chose non-moldy seeds and 10% took the products without any requirements.

3.2. Results of Analyses of Aflatoxin-Contaminated Corn and Peanut Samples

The results of batch analyses of aflatoxin-contaminated maize samples are shown in Figure 8.

The total aflatoxin content (B1, B2, G1, and G2) of maize sample batches harvested in the study area is shown in Figure 1. Samples from maize batch M6 recorded a high total aflatoxin content of 380.83µg/kg, followed by samples from batches M1 and M9 with aflatoxin levels of 45.07µg/kg and 23.6µg/kg respectively. Samples from batches M2 and M10 had intermediate levels of 13.69µg/kg and 10.22µg/kg respectively. Low aflatoxin levels were observed in samples from batches M8 $(1.05\mu g/kg), M7 (2.10\mu g/kg), M5 (2.67\mu g/kg), M4$ (3.63µg/kg) and M3 (4.93µg/kg). In fact, the safe aflatoxin limit is less than 20µg/kg. The low aflatoxin levels were observed on samples of maize dried on tarpaulins and stored in warehouses, while the low levels were also noted on batches of maize samples that had been dried for two weeks and seeds that had been pre-treated. In fact, aflatoxin content varies according to drying method, storage location, drying time and whether or not seeds are treated. The results of the survey showed that 82% of corn is dried on tarpaulins, 6% on the ground, 33% for two weeks and 64% without fungicide or insecticide treatment. Aflatoxin levels were high in growers who dried their maize on the ground for less than two weeks and did not treat their seeds with a fungicide/insecticide mixture.

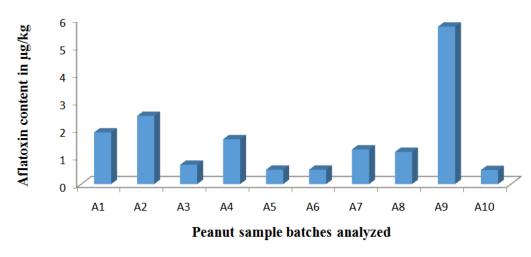


Figure 9. Analysis of aflatoxin-contaminated peanut sample batches

The results of batch analyses of peanut samples contaminated with total aflatoxins (B1, B2, G1, G2) are shown in Figure 9.

The total aflatoxin content (B1, B2, G1, and G2) of peanut samples from the study area is shown in Figure 2. The peanut lot A9 samples recorded a high aflatoxin content (\geq 5.73 µg/kg) followed by lot A2 samples with an aflatoxin content of 2.48 µg/kg. Samples from lot A1 recorded an aflatoxin content of 1.88 µg/kg, followed by those from lot A4 (1.63 µg/kg). Samples from lot A7 obtained an aflatoxin content of 1.26 µg/kg, followed by those from lot A8 ($\geq 1.17 \ \mu g/kg$). The lowest levels were observed in samples from lots A3 and A10, which recorded aflatoxin values of 0.70 µg/kg and 0.51 µg/kg respectively, followed by samples from lots A5 and A6, which obtained the same aflatoxin levels of 0.52 μ g/kg. All batches of peanut samples taken from growers and analyzed contained low levels of aflatoxins, below the limit value (<20µg/kg). The peanut growers in the study area complied with cultivation and post-harvest practices. They dried their groundnuts on tarpaulins and stocked them in their stores. They also treated their seeds with a mixture of fungicide and insecticide. In fact, the aflatoxin values obtained in the batches of peanut samples are below the limit, causing no danger to consumers.

4. Discussion

The results of the survey revealed that men are more involved in farming than women. This can be explained by the fact that women are busier with housework, they are much more involved in raising children, and they are also less hardy than men. Indeed, the low representation of women can be explained by the physical strength required by the practice to maintain the farms, which women do not have; they are also the main people responsible for domestic activities [18]. These findings do not concur with those reported by the author who noted a significant number of women involved in farm work [19]. 67% of those surveyed practice agriculture, in addition to another activity, this to diversify sources of income. The results of the survey revealed that 88% of farmers practiced harnessed farming, a figure that reflects the inaccessibility of tractors to farmers in the study area [20]. There is a correlation between high aflatoxin levels in maize and poor cultivation and post-harvest practices. In fact, high aflatoxin levels in maize samples were noted in growers who dried the maize on the ground in less than two weeks and did not treat their seeds, unlike others who dried the maize on tarpaulins for two weeks, treated the seeds with a mixture of fungicide and insecticide and stored the maize in warehouses. These results concur with those of the author, who reported that post-harvest factors such as drying and storage conditions can induce aflatoxin contamination [21]. Direct and prolonged soil contact with harvested produce increases the risk of aflatoxin contamination. Similarly, high aflatoxin levels have been observed in batches of corn samples stored outside the warehouse. Good post-harvest and drying practices can significantly reduce aflatoxin contamination [22]. Other authors have reported that fungal proliferation and aflatoxin production occur both in the field and during storage [23]. According to these authors, in the field, insects attack the surface of the grains, facilitating mold access to the internal structures that contain the nutrients and increasing the risk of contamination of the edible part. With regard to the drying time, which was two weeks, our results concur with those of many authors who did similar work, those authors reported that drying groundnuts for 14 days reduced aflatoxin contamination [23]. Furthermore, it was noted that 8% of growers harvest maize in August versus 3% of growers who harvest peanuts in August. The contamination of peanuts and maize is linked to factors such as climatic conditions [23]. Indeed, in the middle of the winter season (August), it is very difficult to dry harvested products, aflatoxin contamination during storage is due to high humidity levels [24]. This difficulty in drying leads to the high aflatoxin content observed in batches of maize samples. The results obtained that treating seeds with fungicides and harvesting at the end of wintering, drying for 14 days on tarpaulins and storing in warehouses, these practices will be able to reduce aflatoxin contamination of maize and groundnuts.

5. Conclusion

The aim of the study was to evaluate practices likely to promote mycotoxin contamination of groundnuts and maize. To determine the aflatoxins produced by fungi on maize and groundnuts in the study area, in order to design

a simplified approach to cropping practices that could reduce contamination. The results showed that aflatoxin levels were low in samples of maize and groundnuts whose seeds had been pre-treated with fungicides and harvested at the end of winter (September or October). This low aflatoxin content was also observed on maize and groundnut samples dried for 2 weeks on tarpaulins and stored in warehouses. To reduce aflatoxin contamination of maize and groundnuts, a simplified approach based on good cultivation practices (fungicide treatment of seeds and harvesting at the end of winter), drying (14 days) and storage (warehouses) is proposed. Subsequent studies on the evaluation of practices likely to promote mycotoxin contamination of groundnuts, maize and other crops throughout Barh Koh department will support the results obtained.

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