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Evaluation of aflatoxin levels in selected varieties of cowpea in the Greater Accra Region, Ghana

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ABSTRACT

Cowpea, a major source of food in Ghana is known for its nutritional benefits and is highly recommended for consumption to tackle malnutrition. Its production and distribution however are faced with challenges such as fungal and mycotoxin contamination. The aim of this study was to assess aflatoxin levels of common cowpea varieties in the Nima and Agbogbloshie markets and to explore if these levels fall within the acceptable consumption ranges. The aflatoxin levels of all the procured samples were analyzed following the Reveal Q+ protocol. All the samples analyzed contained aflatoxin at levels ranging from 2.1 to 12.6 ppb. Red beans cowpea variety from Nima market had the highest mean aflatoxin concentration (9.1 ppb) while Agbogbloshie Red beans variety had the lowest mean aflatoxin concentration (3.6 ppb). There was no significant difference between the aflatoxin level of both markets ($p = 0.610$) nor was there a significant difference between the aflatoxin levels in the cowpea varieties ($p = 0.950$). The results revealed in this study compromise food safety and could lead to serious health implications for consumers. Authorities in charge of food ensuring safety must provide important education and training to food crop vendors which will gradually lead to the total elimination of fungi and aflatoxin in food crops. Also, there should be periodic checks on the condition of cowpea and cowpea products in markets to boost food safety.

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1.Introduction

Cowpea [*Vigna unguiculata* (L.) Walp] is an economically essential grain legume crop produced as food in the Guinea and Sudan savannah zones such as the Northern, Upper East and West, the Savannah and a portion of the Brong Ahafo Regions of Ghana (1).

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The Guinea savannah zone (Northern and Upper West Regions) is reported as the major cowpea production area in Ghana (2). Cowpea comes second after groundnut in Ghana when ranked in terms of area of cultivation, quantity produced and annual consumption rate (3).

According to a study by the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT), Ghana is ranked as the fifth highest producer of cowpea in Africa because the country produces an average of 143,00 MT on about 156,000 ha (4). Per Kirse and Karklina (5), cowpea grain is estimated to have a fat content of as low as 1%, about 60% carbohydrate content and above 23% total protein concentration. Cowpea is as well made up of essential minerals and vitamins (6).

Due to cowpea's nutritional components, it is considered to have numerous health benefits and hence the potential to tackle malnutrition. In addition to these nutritious benefits, cowpea is a source of income and has a positive impact on the livelihood of millions of people living in developing countries who depend on agriculture as a source of livelihood. Cowpea, therefore, has the potentials for boosting food security, nutrition and local incomes and economies.

Despite the benefits received from cowpea, its production is associated with challenges including contamination by biological and chemical agents of food poisoning. Cowpea is highly susceptible to fungal and mycotoxin contamination and there have been reports of such contamination in various countries (7 – 9). Certain fungi after contaminating food crops may produce mycotoxins.

Aspergillus species produce aflatoxins in food crops like cowpea which poses dangerous and severe health implications to humans and animals. Aflatoxins in humans and animals have resulted in immunosuppression, impaired growth, cancer and death (10). Also, fungal and mycotoxin contamination impacts negatively on trade and the overall economy (7).

Although agricultural products like cowpea have been identified as targets of Aspergillus species, there is a lack of information on fungal and mycotoxin evaluation of cowpeas sold in Ghana. This is alarming and dangerous due to the high rate of consumption of cowpea by Ghanaians. A recent survey showed that 52.17 % of Ghanaian households consume cowpea produced foods at least once a week (11). This exposes consumers to harmful mycotoxins like aflatoxin which Manafi and Khosravinia (12) describe as the most poisonous and lethal mycotoxin present in grains. To boost safe food consumption and enhance trade of food crops, it is essential to periodically assess commercial food samples for harmful microbes and their toxins therefore this work is carried out to compare the levels of aflatoxin in selected varieties of cowpea and to assess if these levels fall within the acceptable consumption levels.

2. Materials and methods

2.1. Sample collection

A survey was first conducted to reveal the available and the most patronized varieties of cowpea in the various markets. It was revealed that "Nigeria" and "Red beans" varieties were the most available and purchased varieties so the study focused on

cowpea varieties. A total of 500 g of Nigeria and Red beans cowpea grains were then obtained from two different sellers each in Abgogbloshie and Nima markets. The samples were aseptically packaged into well-labelled paper bags and then transported to the Spanish Laboratory Complex of the University for Development Studies, Nyankpala for aflatoxin analysis. Samples collected from Agbogbloshie markets were identified as An (Agbogbloshie Nigeria) and Ar (Agbogbloshie Red beans) while those collected from Nima markets were labelled as Nn (Nima Nigeria) and Nr (Nima Red beans).

2.2. Aflatoxin detection

The aflatoxin level of all the procured samples was analyzed following the Reveal Q+ protocol (13). The Reveal Q+ aflatoxin test kit used to detect the aflatoxins in this study had an LOD and LOQ of 2 ppb and 3 ppb respectively. The test kit uses 6 calibration standards (0, 2, 4, 8, 16 and 32 ppb) and an aflatoxin recovery percentage range of 75 – 110% (13).

The analysis was carried out using analytical grade diluents and reagents. The cowpea samples were ground into a powdery form. Then, 10 g of the powdered sample was weighed into a glass bottle and 30 ml of 65% ethanol was added to the weighed sample. After, the glass bottle was covered and shaken for 5 min to homogenize. The sample extract was then filtered into a jar. 500 mL of the diluent was pipetted into the red sample cup. After, 100 mL of the sample extract was then added to the diluent in the red sample cup.

The mixture obtained was mixed by pipetting up and down five times after which 100 mL of the mixture obtained was further transferred into the clear sample cup.

A test strip was placed in the solution in the clear sample cup and allowed to develop for about six min. The strip was analyzed using m-Reader and the results were recorded and stored on the m-Reader.

2.3. Data analysis

After all the aflatoxin analyses were carried out, results were recorded and analyzed using Microsoft Office Excel 2016 and Genstat edition 12.

3. Results

The aflatoxin analysis conducted revealed a successful detection of aflatoxin in all cowpea grains collected from the retailers in the Agbogbloshie and Nima markets of the Greater Accra Region. Nigeria cowpea seeds variety collected from Agbogbloshie market recorded the highest level of aflatoxin with 12.6 ppb while Nigeria cowpea seeds variety from Nima market also recorded the lowest aflatoxin amount at 2.1 ppb (Table 1).

Table1. Aflatoxin concentration in cowpea varieties from Abgogbloshie and Nima markets

Variety	Aflatoxin levels (ppb)	
	Minimum	Maximum
Ar	3.0	4.3
An	2.8	12.6
Nr	7.5	10.7
Nn	2.1	8.8

Furthermore, the results showed that Nima red beans had the highest mean aflatoxin concentration of 9.1 ppb followed by Agbogbloshie Nigeria variety (7.7 ppb) whiles Agbogbloshie red beans variety had the lowest mean aflatoxin concentration (Fig. 1).

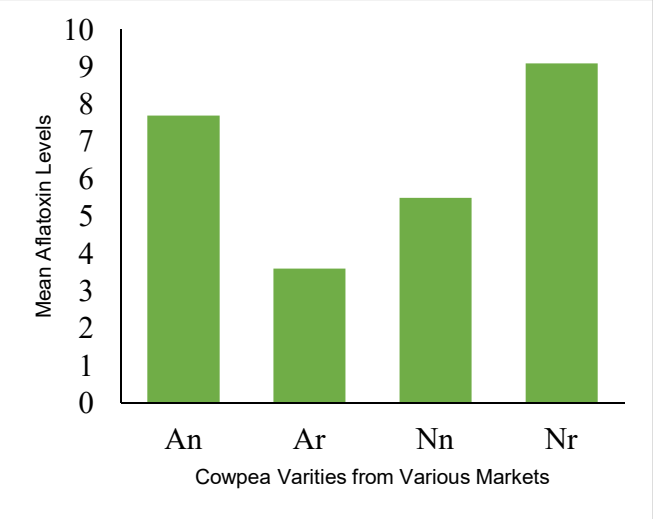

Figure 1. Mean Aflatoxin concentration in cowpea varieties from Abgogbloshie and Nima markets

Cowpea samples in the Nima market (7.3 ppb) showed a relatively higher aflatoxin mean concentration than those in Agbogbloshie markets (5.7 ppb) (Table 2). Results of the study revealed that there is no significant difference between the aflatoxin level of both cowpea varieties considered in this study as well as the markets (Table 2).

Table 2. Descriptive statistics of aflatoxin levels

Sample	Mean Aflatoxins (ppb)	P-value
Varieties		
Nigeria	6.6	0.950
Red Beans	6.4	
Markets		
Agbogbloshie	5.7	0.610
Nima	7.3	

4. Discussion

The aflatoxin analyses in cowpea grains collected from retailers in the Agbogbloshie and Nima markets of the Greater Accra Region successfully detected aflatoxin in all samples. Nigeria cowpea variety collected from the Agbogbloshie market showed the highest level of aflatoxin with 12.6 ppb while the Nima Nigeria cowpea variety contained the lowest aflatoxin concentration at 2.1 ppb. Additionally, the findings indicated that Nima red beans had the highest average concentration of aflatoxin, measuring at 9.1 ppb, followed by the Agbogbloshie Nigeria variety with 7.7 ppb, whereas the Agbogbloshie red beans variety had the lowest average aflatoxin concentration. The findings of this study is supported by that of Achaghinkame et al. (14) which reported incidences of aflatoxin contaimination on several legumes and cereals retailed in Ghana.

The results of this study project a disturbing and worrisome future for cowpea consumers in Ghana because of the presence of aflatoxin in the most patronized and consumed cowpea variety samples.

This is similar to the findings of Ogungbemile et al. (10) who reported aflatoxin presence in stored cowpea samples from markets in Ibadan, Nigeria.

The presence of aflatoxin in the sampled varieties from both Nima and Agbogbloshie markets could be attributed to various factors such as poor handling during harvest and packaging for sale, poor storage conditions and favourable environmental conditions that promote the growth of fungi. Fuseini (15) reported that sandy soils promote rapid proliferation of Aspergillus flavus, especially, in adverse dry conditions. Most cereals and legumes in Ghana such as cowpea are grown in this type of soil hence a possible reason for the presence of aflatoxin in some of the samples analysed in this study.

Although both varieties of cowpea from the markets contained levels of aflatoxin, it is relevant to note that the levels fall below 15 ppb and 20 ppb which are the acceptable or tolerable consumption range standards set by Ghana Standards Authority and US food safety authorities. This find is corroborated by that of Ahmadi et al. (16) who reported that all their legume samples were in the aflatoxin allowable limits in foods and feeds set by the US FDA.

Unlike the finding of Ahmadi et al. (16) where all the legume samples analysed for aflatoxin contained aflatoxins acceptable by the Codex Alimentarious standard, 25% of the cowpea sampled varieties in in this study were above that limit. It may seem good news that the level of aflatoxin in most (75%) of the samples are below the acceptable consumption levels but that still does not guarantee the safety of consumers from the deleterious effects of aflatoxin.

Foods that have contaminants below the acceptable limits do not mean a consumer will be safe totally from foodborne illnesses and possible complications but they assure reduced intake of a particular food toxin or contaminant hence reduced chances of complications. There are possible health effects that can arise in the long run due to the possible accumulation of toxins in body organs and systems. To prevent foodborne illnesses and prevent health hazards, foods that are free from fungi and aflatoxin are those that have to be traded and consumed (17). According to Santini and Ritieni (18), ingesting small quantities of aflatoxin over the long term could result in cancers and mutation. The recommended total aflatoxin level in food products by the World Health Organization is therefore 0 ppb for children.

Cowpea samples in the Nima market (7.3 ppb) showed a relatively higher aflatoxin mean concentration than those in Agbogbloshie markets (5.7 ppb) (Table 2). Results of the study revealed that there is no significant difference between the aflatoxin level of both cowpea varieties considered in this study as well as the markets (Table 2). Per this finding, a direct relationship may not be drawn between the aflatoxin level and markets as well as aflatoxin level and cowpea variety so handling and storage by the cowpea vendors can be pointed as one of the main factors that influenced aflatoxin concentration in the cowpea samples. Post-harvest storage conditions of agricultural produce are generally poor in Ghana and most African countries (19). Achaghinkame et al. (14) reported that some of the storage conditions used by farmers and retailers in

Ghana do not provide the right internal condition t give adequate protection from insects, water and rodents which does facilitate aflatoxin contamination. Poor handlng habits influence aflatoxin contamination in cowpea as supported by the findings of Samari et al. (20) and Ahmed and Asgar (21).

In the Agbogbloshie market, the Nigeria cowpea variety had the highest mean aflatoxin concentration (7.7 ppb) while in the Nima market, the red beans cowpea variety rather recorded the highest mean aflatoxin concentration (9.1 ppb) and not the Nigeria cowpea variety. The activities of food handlers such as food vendors hence are crucial to contamination of food by contaminants.

The presence of aflatoxin in all varieties and across both markets is alarming as it compromises food safety. This finding calls for an investigation into the available cowpea storage facilities used by the cowpea vendors for storing cowpea samples in the market as well as the handling practices of cowpea vendors across these markets. Also, the presence of aflatoxin in the cowpea samples could lead to a significant economic burden for both individuals and the government. Foods containing harmful toxins such as aflatoxin are at risk of being rejected for exportation and called off from the market because of the strict acceptable limits set by several regulators around the world. For instance, 25% of the cowpea samples from this study would not make it into areas that rely only on Codex Alimentarius acceptable aflatoxin limit.

It is important to acknowledge the limitations of this study. The small sample size used in this study is one key limitation. The small sample size may limit the statistical power and the generalizability of the findings.

Despite these limitations, the study provides valuable insights into the aflatoxin contamination levels of cowpea and the consequence of such, especially considering the limited available information on cowpea aflatoxin contamination in Ghana.

5. Conclusion

The study revealed a presence of aflatoxin in various cowpea samples across the markets indicating a food safety concern although the aflatoxin levels were below the recommended limits set by the Ghana Standards Authority. The study suggests that food vendors' perception and habits also influence the level of contamination by aflatoxin in foods. A similar study should be replicated in other areas of the country to give a better insight into the country's aflatoxin challenge. Authorities in charge of ensuring food safety should provide important education and training to food crop vendors which will gradually lead to the total elimination of fungi and aflatoxin in food crops. Similarly, authorities should conduct random checks on the condition of cowpea and cowpea products in markets to boost food safety. Further research should be conducted to characterize the various aflatoxins in food products to understand the extent of the damages that can be imposed on human health by these toxins.

Conflict of Interest

The authors declare that there is no competing interest.

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