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Aflatoxin concentrations in locally processed Groundnut products in the Tamale Metropolis

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ABSTRACT

This study determined the aflatoxin levels in processed groundnut products in the Tamale metropolis. A simple random sampling technique was used to obtain roasted groundnuts, groundnut paste, kulikuli, and kulikulizim samples from three markets in the Metropolis. A total of 36 locally processed groundnut products were obtained and analyzed for total aflatoxin concentration, using the Reveal Q+ aflatoxin test method. All the groundnut products sampled tested positive with values ranging from 1.7 to 519 ppb. The most aflatoxin-contaminated product was kulikuli followed by kulikulizim. The roasted groundnut recorded the lowest mean total aflatoxin concentration of 21 ppb which was the closest to the acceptable limit of total aflatoxin (20 ppb) in Ghana. The high aflatoxin levels in kulikuli is a health concern that needs urgent attention.

Keywords: *Aflatoxin; Kulikuli; Kulikulizim; Groundnut paste and Groundnut*

INTRODUCTION

Aflatoxins are toxic secondary metabolites that are produced predominantly by *Aspergillus* fungal species such as *Aspergillus flavus*, *A. nomius,* and *A. parasiticus* (Granados-Chinchilla et al., 2017; Juan et al., 2008; Shukla et al., 2017). They are low molecular weight (312 g/mol-328 g/mol) lipophilic compounds that are made up of alternating groups of ketone and methylene called polyketides which are carcinogenic and immunosuppressive to mammals (Amaike & Keller, 2011; Pandey et al., 2019; Sarmento Amoras & Pena Costa, 2020). Exposure to aflatoxins through the consumption of contaminated foods may lead to acute poisoning (aflatoxicosis) which is characterized by a weakening immune system, impaired growth, and other health issues such as liver cancer in both livestock and humans (Alenyorege et al., 2015; Awuah & Kpodo, 1996; Coulombe et al., 2005; Hong et al., 2010). There are four naturally occurring aflatoxins which are B1 and B2 (produced by *A. flavus*), and G1 and G2 (produced by *A. parasiticus*). Among these, B1 is classified as a group 1, and the most potent carcinogen and genotoxic that affects all mammalian species even at low concentrations (Pandey et al., 2019; Pitt, 2014). The prevalence of aflatoxin has been reported in various food commodities from many parts of the world (Juan et al., 2008). Their contamination can occur at pre-harvest, harvest, and post-harvest stages in the food chain which is facilitated by various conditions including insect damage, grain physical injury, high temperature, water activity, and humidity (Kebede et al., 2012; Marin et al., 2013). They have also been reported in processed foods such as cereal legume blends in Ghana (Opoku et. al., 2018). Aflatoxins affect the nutritional quality of products and cause economic loss to farmers and traders. Groundnut is one of the most susceptible substrates for aflatoxin production.

Groundnuts (*Arachis hypogaea* L.) also known as peanuts are one of the most important oilseed and food crops that are cultivated in the world (Prasad et al., 2010). Groundnuts can be used directly for food, or processed for cooking oil and a range of products (Shanahan et al., 2003; Singh & Singh, 1992). Due to its vitamins and mineral content groundnut products are utilized globally for the treatment and prevention of malnutrition in infants and young children, especially in less privileged homes that cannot afford animal protein (Achaglinkame et al., 2017; Asare et al., 2019; Masters et al., 2015). In Ghana, groundnuts are widely cultivated on small scale and consumed in almost every part of the country (Naab et al., 2005). It plays a very significant role in the lives of marginal farmers with respect to income and nutrition (Carlberg et al., 2014). In northern Ghana especially, groundnuts are eaten in roasted forms or processed into forms such as paste, *kulikuli,* and *kulikulizim* (Florkowski & Kolavalli, 2013). The impact of aflatoxin contamination in groundnuts along the food chain has been a significant threat to trade, food security, and livelihoods in the country (Kumar et al., 2008). The stability of aflatoxin to varying processing conditions such as cooking, roasting, and extrusion makes it difficult to eliminate it from contaminated food and food products. Therefore, maximum limits have been set in various countries for foods and feeds (García & Heredia, 2014). The Ghana Standards Authority, for instance, considers 10 ppb as the acceptable level of aflatoxin in groundnuts (Omari et al., 2022), while the European Union (EU), Codex Alimentarius Commission (Codex), and United States (USA) set 4 ppb, 15 ppb and 20 ppb, respectively, as the permissible limits (Agyekum & Jolly, 2017; Omari et al., 2022; WHO, 2004). Akullo et al. (2023) reported high levels of aflatoxin in groundnut products with 41.8% of the values higher than 20 ppb in the eastern and northern regions of Uganda. Similar reports have been made by different authors, in Zimbabwe (Dube & Maphosa, 2014), Nigeria (Ajeigbe et al., 2015), and Mali (Waliyar et al., 2015).

This study evaluated the occurrence of aflatoxins in groundnut-based products in the Tamale metropolis in the northern region of Ghana. The outcome of this study will help to create public awareness most especially for the safety and wellbeing of consumers.

MATERIALS AND METHODS

Sample collection

A simple random sampling technique was used to obtain samples from the markets. Three of each groundnut product was purchased from each market. A total of 36 groundnut products (roasted groundnut, groundnut paste, kulikuli, and kulikulizim) were purchased from 3 different markets (Aboabo, Central, and Lamashegu) in Tamale metropolis and stored in a refrigerator (at -20ºC).

Sample preparation and extraction of groundnut samples

The samples were ground using a waring blender (HGBRWTS3-USA) and sieved using 20 mesh sieves. The sieved sample was extracted using 65% ethanol in distilled water with a sample-to-solvent ratio of 1:5. The resulting mixture was vigorously shaken in a well-covered glass bottle for 3 minutes. The mixture was then filtered with filter paper into a glass jar to obtain groundnut extract for analysis.

Total Aflatoxin Analysis

The samples were analyzed as described in the Reveal Q+ Test kit protocol for aflatoxin analysis. The Reveal Q+ method is a single-step lateral flow immunochromatographic assay based on a competitive immunoassay format, designed for the quantification of aflatoxin in cereals, grain, and their products (Le et al., 2019). Briefly, the red sample dilution cups and white sample cups were placed into a sample cup rack and 500 µL of sample diluent was pipetted into each red sample solution cup. 100 µL of sample extract was then added to each red sample dilution cup containing the sample diluent. The mixing was done by pipetting up and down five times. Subsequently, 100 µL of each diluted sample extract was transferred into the white sample cups, and a new Reveal Q+ for Aflatoxin test strip was placed in the sample cup and a timer was set for 6 minutes to ensure wicking. After 6 minutes, it was removed and inserted into the cartridge. The cartridge with the test strip was then inserted into an mReader application one after the other. The mReader automatically analyzed the test strip and the results were displayed in part per billion (ppb).

Statistical Analysis

Data were entered into an EXCEL spreadsheet and analyzed by one-way analysis of variance (ANOVA) at a 5% level of significance using GenStat (12th edition) statistical software.

RESULTS AND DISCUSSION

Groundnut is a major dietary staple crop that is highly nutritious and high in energy owing to its high fat and protein content. It is widely cultivated and consumed in Ghana, Africa, and the world as a whole. However, it is one of the most susceptible crops to aflatoxin contamination (Jallow et al., 2021; Kumar et al., 2021). The impact of aflatoxin contamination along the food chain has been a major threat to trade, food security, and livelihoods. About two-thirds of farmers in the northern part of Ghana cultivate groundnuts on a relatively medium scale for sale, which is usually on a need basis and for home consumption. Groundnut serves as an imperative ingredient in local foods. Groundnut can be eaten roasted or processed into other forms such as paste which is used for soups and stews or into snacks such as kulikuli. Data on aflatoxin levels in locally processed groundnut products is limited in Ghana.

There was a highly significant difference ($p = 0.004$) within the locally processed groundnut products obtained from the markets. *Kulikuli* recorded the highest level of aflatoxin concentration (319 ppb), while roasted groundnut recorded the lowest (21 ppb) (Figure 1). The results showed that all the groundnut products had some levels of aflatoxin contamination. On average, only 17% of the products were below the various acceptable limits (4-20 ppb) for total aflatoxin in groundnut products. The high level of aflatoxin in three out of four of the products could be attributed to both primary and secondary factors. Primary contamination arises from pre-harvest and post-harvest practices (Abass et al., 2014; Gachara et al., 2022; Mahuku et al., 2019) whereas secondary contamination normally occurs due to poor handling that allows inoculation of the product with the fungi during the production process (Chang et al., 2013).

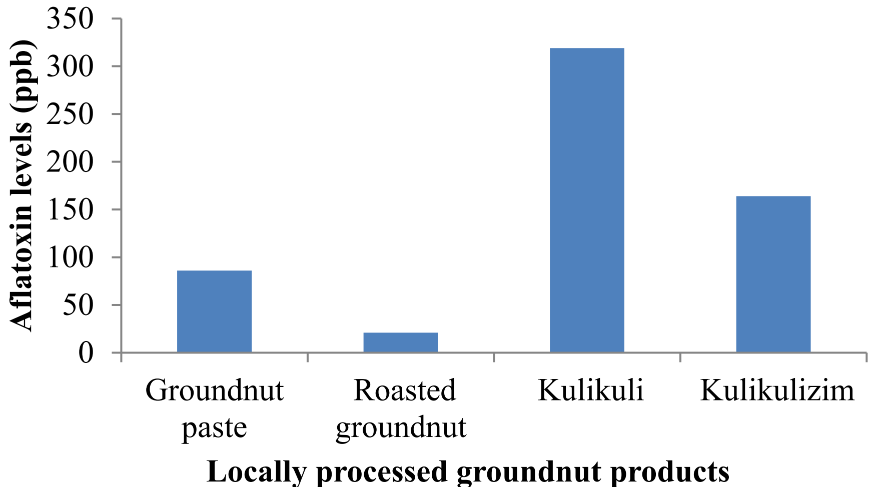

Figure 1: Average aflatoxin levels in various locally processed groundnut products

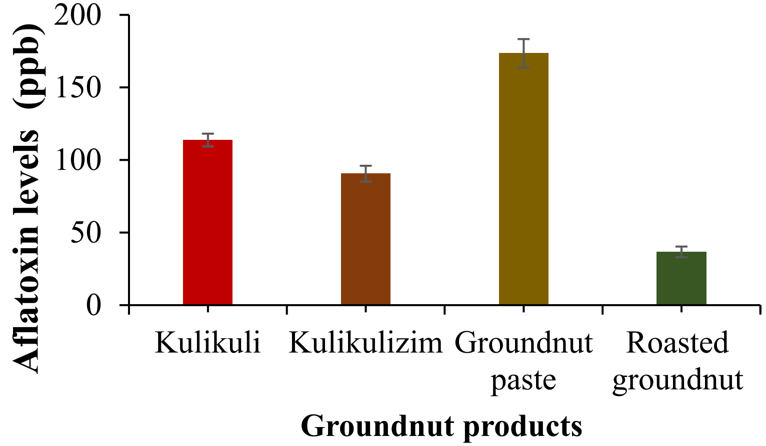

Figure 2. Aflatoxin level of processed groundnut product from Lamashegu market

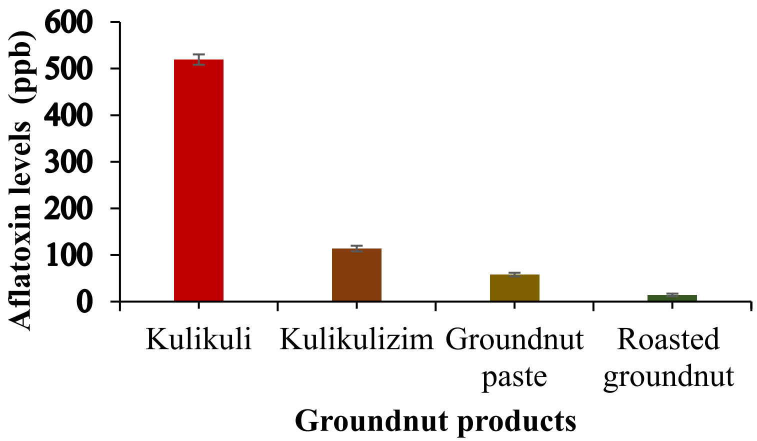

Figure 3: Average aflatoxin levels in the various locally processed groundnut products from the Central Market (error bars = SEM)

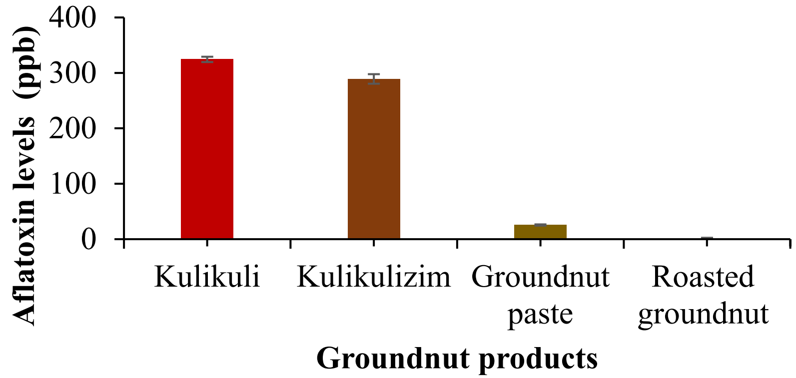

Figure 4: Average aflatoxin levels in the locally processed groundnut products in the Aboabo market (error bars = SEM)

Aflatoxin levels in locally processed groundnut products in various markets

Lamashegu market

In this market, groundnut paste had the highest mean value of 173 ppb, while roasted groundnut had the lowest aflatoxin concentration of 36 ppb (Figure 2).

Central Market and Aboabo Market

In the central market, kulikuli had the highest level of aflatoxin with a mean value of 519 ppb (Figure 3) while in the Aboabo market, kulikuli had the highest mean aflatoxin of 324 ppb. This was closely followed by kulikulizim (Figure 4).

Primarily, contamination may arise from pre-harvest and post-harvest practices (Abass et al., 2014; Gachara et al., 2022; Mahuku et al., 2019). Drought stress is one of the major contributing factors to pre-harvest aflatoxin contamination in groundnuts (Guo et al., 2008) and this is exacerbated when coupled with high temperature and low atmospheric humidity (Achaglinkame et al., 2017; Molnár et al., 2023). Inappropriate post-harvest practices such as poor shelling can lead to mechanically damaged kernels which increases the ease of fungal attack and colonization and thus aflatoxin contamination. Poor storage is a critical postharvest practice that has the potential to increase aflatoxin buildup in groundnuts during storage. When nuts are not properly dried before storage or when there is reintroduction of moisture into nuts as a result of rewetting and activities such as respiration of rodents and insects, aflatoxin builds up rapidly.

The high level of aflatoxin in *kulikuli* can be attributed to the use of high-risk aflatoxin groundnuts. Currently in the Northern parts of Ghana, the practice among groundnut farmers and traders is that, when bad nuts (high-risk aflatoxin groundnuts (HRAG)) are sorted from the lot, the good lot is sold and the HRAG are either mixed with relatively good ones and consumed by the household or sold at low prices to retailers. The HRAGs thus find their way back into the food value chain when producers of *kulikuli* and *kulikulizim* purchase them due to their low price, and mix with relatively better nuts but of higher production price, to reduce production cost. HRAG, normally, would have a rancid to bitter taste. However, when used in relatively low amounts in products like *kulikuli*, *kulikulizim,* and groundnut paste, the bitterness is masked, most especially in kulikuli and kulikulizim due to the addition of spices to these two products. This may explain the higher aflatoxin levels in *kulikuli* and *kulikulizim*. Again, *kulikuli*, *kulikulizim* and groundnut paste are all milled in commercial milling machines during processing.

In all three markets, roasted groundnuts consistently recorded lower aflatoxin levels compared to the other groundnut products. Roasted groundnut, is normally processed by sorting before roasting. Sorting is known to reduce aflatoxin by about 40-80% (Park, 2002). Thus, the low levels of aflatoxin in roasted groundnuts from all the markets suggest that sorting was done before roasting. This is understandable because a single HRAG can easily be noticed in packaged roasted nuts or taste bad once chewed on, which can lead to reduced acceptability by consumers.

CONCLUSION

This study evaluated the aflatoxin concentration in groundnut products. Out of the 36 groundnut products collected from the three markets, only 17% which happened to be roasted groundnut samples had aflatoxin levels (1.7 -20 ppb) within the permissible limits in Ghana. The extremely high levels of kulikuli raise serious health concerns especially when it is used as a snack for children. This calls for education and sensitization of stakeholders on the necessary value chain management approach in reducing aflatoxin levels. The efforts by food processors and vendors in controlling aflatoxin levels and measures by farmers to control pre- and post-harvest contamination are essential to ensure the quality of these products.

Conflicts of interest

The authors declare that there are no conflicts of interest.

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