

**INVESTIGATING AFLATOXIN CONTAMINATION AND KNOWLEDGE
LEVELS IN PRODUCING SAFE PEANUT BUTTER AMONG SELECTED
LUSAKA URBAN PROCESSORS**

By

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DECLARATION

I hereby declare that this is my original work and has not been presented wholly or partly for a degree at the University of Zambia or any other University. It has been produced in accordance with the guidelines for Master of Science in Population, Health and Nutrition

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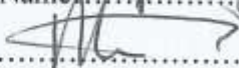
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ABSTRACT

Aflatoxin contamination is a major global public health problem especially in developing countries. Its risk effect on human consumption of peanut butter has posed a serious public health challenge by increasing morbidity and mortality in human populations. In Zambia, local and international peanut butter is consumed by the public. The objective of the study was to determine aflatoxin levels in peanut butter and factors associated with its quality production in selected urban district outlets. The study also assessed the knowledge level and practice of producing peanut butter among local processors. This study compared aflatoxin levels in peanut butter between local and international products. A cross sectional comparative study survey used quantitative and qualitative approaches from selected outlets of Lusaka, Zambia. Samples from plants, commercial stores and Soweto market of Lusaka provided the data to answer the objective. A total of 109 peanut butter samples from local and international sources were collected. Samples were tested for aflatoxin contamination levels using AccuScan Reveal Q+ test in seeking to answer the hypothesis whether there was a difference in levels of aflatoxin between local and international peanut butter based on set standards. Besides clinical assessment of aflatoxin levels, 16 key informants were interviewed using an semi structured questionnaire guided to assess the level of knowledge about aflatoxin contamination, health risks and production skills as they processed peanut butter. The findings show that only 9 (8.3%) of the 109 (100%) peanut butter samples satisfied the 0 to 4ppb European set standard as safe for public consumption regardless of its origin. It was found that 100 (91.7%) samples of peanut butter from both local and international origin were contaminated with aflatoxin. Using the European standard, there was no sufficient evidence that the level of contamination was different between local and international peanut butter, P-value 0.0768. However, using the 15ppb standard, 83 (76.1%) samples from both local and international origin were safe for consumption based on the Codex Alimentarius Commission (CAC) standard. There was a marked difference in proportions, between compared products had a p-value of less than 0.00001. Nevertheless, there was aflatoxin contamination of above 15ppb in a total of 26 (23.9%) samples of which 25 (22.9%) were locally produced and one (0.9%) was internationally produced. These samples were not safe for human ingestion.

Generally, processors seemed to have little knowledge of aflatoxin contamination and health risk. The steps of producing peanut butter were largely similar between plants and Soweto in first stages, and dissimilar in the last steps. It is recommended that government regulations be strengthened to aid processors provide quality peanut butter on the market. In order to produce peanut butter with low aflatoxin levels, there is a need to come up with a standard to follow. Further research is recommended on how to assess and improve the quality of peanut butter production as well as increasing awareness to the public about the dangers of aflatoxin contamination.

DEDICATION

This work is dedicated to the promotion of the good health of the Zambian population.

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ACRONYMS

AOAC	Association of Official Analytical Chemists- method 925.40, moisture in nuts and nut products. Method 935.53- Fiber in nuts and nut products.
CAC	Codex Alimentarius Commission- international regulatory board which sets limits on food safety commodities for consumer safety.
HACCP	Hazard Analysis Critical Control Point
HPLC	High Performance Liquid Chromatographic
EFSA	European Food Safety Authority
EU	European Union
ICRISAT	International Crops Research Institute for the Semi-Arid Tropics
IUPAC	International Union of Pure and Applied Chemists
LP	Level of processing
MTL	Maximum Tolerated Level
TLC	Thin Layer Chromatography
USAID	United States Agency for International Development
ZABS	Zambia Bureau of Standards
ng/kg	Nanogram per kilogram body weight per day
µg/kg	Microgram per kilogram
ppb	Parts per billion

CHAPTER ONE

INTRODUCTION

1.1 Background

Aflatoxin contamination is a major global Public Health problem especially in developing countries and may account for as high as 28% of all global liver cancer cases (Liu and Wu, 2010). Several epidemiological studies in Africa and Asia have demonstrated an association between dietary Aflatoxin and liver cancer, the International Agency for Research on Cancer (IARC) in 1988, placed aflatoxin B1 on the list of human carcinogens (IARC, 2002). Further, the occurrence and extent of aflatoxin contamination varies and is influenced by many environmental factors including geographical location, suboptimal agriculture and agronomic practices susceptibility of commodity to fungal invasions pre-harvest, transportation, storage and processing (Georgiadou et al. 2012, Schwartzbord and Brown, 2015 and Mutegi et al. 2013).

According to Gulchi (2015), aflatoxin is a naturally occurring toxic secondary metabolite produced by fungi *Aspergillus flavis* and *Aspergillus parasiticus*, soil borne fungi which can grow on both living and dead plant matter. Gulchi further stated that moisture, due to high heat accumulation, influence fungi populations in the soil. When temperature increases in the soil, moisture reduces causing drought stress. Fungi competition with inactivity progression lead to production of increased toxins on groundnuts. Further, aflatoxin producing strains, *Aspergillus flavis* and *A. parasiticus* can grow at optimum temperatures ranging from 32 to 33 degrees centigrade (UÇKUN and Işıl, 2014). Aflatoxin contamination development begins with fungus in the soil and dead matter in the field (Atayde et al. 2012). Since aflatoxin occurs naturally, it cannot be terminated or eradicated completely but can be reduced to low levels that are safe for human consumption (Shephard, 2008).

Historically, aflatoxins were first discovered as a result of the deaths of over 100, 000 young Turkeys in England in 1960 from a new disease termed “Turkey X disease” (Blount, 1961). Guo et al. (2009) also reported “Turkey X disease” outbreak which occurred in more than 150 villages in Western India in 1974 where 397 persons were affected and 108 persons died (cited in Krishnamachari et al. 1975). The investigations on the poultry and ducklings outbreak of “Turkey X

disease” by Blount (1961) revealed that there was an association between the feed given to these birds, a Brazilian peanut meal and the high toxins. Since then, aflatoxin has been of interest for study by scientists (Shephard, 2008). In rural Kenya, a largest outbreak happened in April 2004 where 317 cases and 125 deaths of mostly children were reported because of eating aflatoxin contaminated maize (Lewis et al. 2005). When highly aflatoxin contaminated products are consumed or just chronic consumed in low doses, health risk problems such as, liver cancers, aflatoxicosis, liver cirrhosis, stunted growth, low immunity and eventually death can occur (Guo et al. 2009, Schwartzbord and Brown, 2015, Nyikal et al. 2004). Again literature reveals that highly contaminated peanut products contribute to economic loss for the traders (N’dede et al. 2012).

Based on findings from various studies, many countries have set standards for safe tolerable limits for humans to consume in different products (Njumbe et al. 2014). Current local and international Maximum Tolerated Levels for developing countries, as imposed by the European Union (EU), are between 4µg/kg and 30µg/kg (FAO, 2003, Njumbe et al. 2014). Furthermore, legislation in the EU sets maximum aflatoxin levels for groundnuts destined for processing within the EU at 10 ppb and for those destined for direct human consumption at 4ppb (Otsuki et al. 2001, FAO, 2003). In some developing nations, including Zambia, where limits are not yet in place, 15µg/kg has been imposed as maximum acceptable levels of flatoxin for safe consumable peanut products (EFSA, 2007 and ZABS, 2008). The US Food and Drug Administration set guidelines for maximum allowable limit at 20ppb for food and feed in developing countries (Guo et al. 2009).

With regard to aflatoxin knowledge among stakeholders, Azaman et al. (2016) found that this was necessary. Processor knowledge contributed greatly to aflatoxin reduction in peanut products in their study. Little is known about processor knowledge on aflatoxin contamination and health risks associated with consumption of aflatoxin peanut butter in Lusaka urban district. Besides, skills used to produce peanut butter in these outlets is also scarce. For example, whether sorting is done to remove bad affected and poor grade groundnuts to encourage use of good groundnuts for processing peanut butter is not known. This study is the first to source such knowledge from processing plants, hence the gap to be filled. Availing aflatoxin knowledge to stakeholders like farmers, processors, traders and consumers is

necessary. This knowledge can be used in Public Health as base for interventions essential to address aflatoxin concerns. Increasing aflatoxin related research efforts can bring about vital realistic and helpful knowledge. Wild and Gong (2002) stated in their review that Toxicology of aflatoxin has paved way to provide base for decision making in Public Health on acceptable exposures and interventions to reduce aflatoxin contamination risks in human.

1.2 Statement of the Problem

Aflatoxin is a Public Health problem. Matumba et al. (2015) in Malawi in their study found that aflatoxin levels were high in the locally sold groundnuts than groundnuts destined for export. It was also stated in that study that public knowledge about aflatoxin was limited. In Zambia, ZABS has been using 15ppb as limit for aflatoxin contamination but despite this documentation, aflatoxin results have been on the high level. Bumbangi et al. (2016) found high levels of aflatoxin contamination in groundnuts sold in the supermarkets and markets. Another example is that done by Njoroge et al. (2016) who recorded aflatoxin contamination of 10,740ppb in peanut butter over a period of three years. In both studies done in Zambia, there is limited documentation on levels of aflatoxin contamination in peanut butter for both local and international products. For this reason, imported or local peanut butter in all kinds of brands and textures find their way to the market and are sold by traders without considering safety and risking the population's health. This information may not be available to the public due to inadequate information about aflatoxin contamination in peanut butter. Some processors may or may not know the reasons behind the need to produce reduced aflatoxin contaminated peanut butter. High aflatoxin contamination in processed peanut butter on Lusaka's outlets might cause health risks to humans due to accumulation of aflatoxin in the body over a period of time. Deficiency in aflatoxin contamination knowledge may lead to defects in the processing of nutritious peanut butter that would be packaged, and properly labelled using sorted suitable groundnuts. Further, lack of adherence to defined national aflatoxin set standards by processors may lead to high aflatoxin contamination levels in peanut butter in the outlets. This outcome may lead to the presence of contaminated peanut butter on the market, loss of income for traders both locally and internationally, reduced nutrients in the products, ill health to consumers and eventually deaths as reported in other nations.

Earlier studies concentrated on investigating aflatoxin levels in peanut products from the markets and stores. This study is the first to include processing plants for aflatoxin contamination level examination, processing skills, knowledge of contamination and health risks among processors which is also limited. This information will assist in detecting why aflatoxin levels continue to rise in peanut butter in the outlets. This revelation has led to the need for the current study to be undertaken.

Below is Figure 1 showing fresh groundnuts which are raw products used in processing peanut butter. Among them are bad types of peanut which sometimes can be used in processing peanut butter without noticing, thereby increasing aflatoxin in the products.



Figure 1: Aflatoxin ICRISAT & A.F Ecology Centre Farmer's Field Demonstration in Peanut Source: (Grace, 2013).

The consumer must be dependent on processors to provide safe peanut butter in their outlets. And further, cleaning of grinder is to reduce on aflatoxin contamination as Ndung'u et al. (2013) documented in their study. This knowledge is vital for processors to have in this study.

1.3 Conceptual Framework

Figure 2 is a conceptual framework showing variables that might contribute to increased aflatoxin in peanut butter.

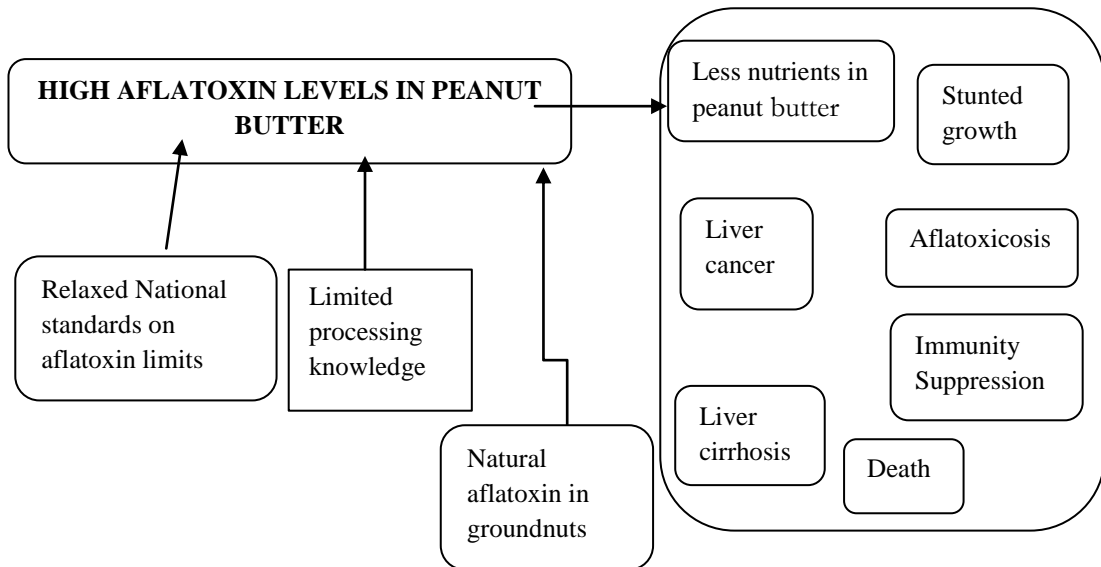


Figure 2: Conceptual Framework for Levels of Aflatoxin in Peanut Butter

The figure above shows how naturally aflatoxin from groundnuts or peanut butter can lead to high levels of aflatoxin. When we plant the seed in the soil, without clearing dead matter, high chances of aflatoxin is increased (Atayde et al. 2012). As the crop continues to grow under the soil, there is a release of carbon and nitrogen substrates by injured groundnut pegs which culminate into colonization of the pods. When the environment is hot and humid, there is release of spores on plant residues which get dispersed by wind through the field (Donor & Cole, 2002). Moisture forms moulds which increases aflatoxin in the product. Small, immature kernels are more easily infected in a shorter period of time than kernels in more mature pods. Infections of groundnut kernels at other maturity stages are relative to the survival of the fungus and not as a new infection at a later stage of maturity.

Relaxed national aflatoxin limit standards contributes to the presence of high aflatoxin contaminated peanut products. Again limited processing skills and knowledge about dangers of aflatoxin contamination contributes to increased aflatoxin in peanut butter.

In the post-harvest stage, aflatoxin continues to develop as peanuts are being prepared for storage. If peanuts are not sorted to separate quality peanut from

immature and discoloured ones, there is an increased chance of moisture creation and insect manifestation (UÇKUN and Işıl, 2014). Moisture and insects increase aflatoxin in peanut which in turn reduces the quality of peanut. When such low quality peanuts are used for peanut butter processing, aflatoxin levels are likely to be raised. Again if the storage area is not clean and well ventilated, humidity and mould develops and with it aflatoxin progression continues (Njumbe et al. 2014). When such peanut is used for processing peanut butter, the possibility of having raised aflatoxin in the product is again high.

1.4 Rationale

Aflatoxin contamination is a subject that is very important in Public Health. Aflatoxin consumption by humans through contaminated peanut butter is a Public Health hazard. Availability of information on aflatoxin is therefore crucial to stakeholders as the population's wellbeing rely on it particularly when decisions are to be made. For instance, to come up with an intervention for addressing aflatoxin contamination problems, research findings from studies like the current one would be very necessary. For as peanut butter is highly consumed by many people in Lusaka, the public is at high health risk of ingesting increased aflatoxin contaminated products in their bodies.

Results from this study will also provide knowledge to processors as well as stakeholders on aflatoxin level contamination in peanut butter and the dangers associated with the problem. The results of the study will encourage ZABS to work towards toughening regulations on processor adherence to the available standards. As a result of this study, subsequent researches in peanut butter may begin to show reduction in aflatoxin contamination levels. When that period is attained, peanut butter will have viable markets both locally and internationally. Processing plants will ensure they put into effect knowledge gained to advance market confidence but also to protect the population which they serve from contaminated peanut butter and health risks.

Further, results will help with formulation of policy and programs on clearer guidance on acceptable aflatoxin limits for peanut butter from local and international sources. This evidence underlines the need to come up with a standard to use in checking for quality imports and exports in the nation. The results will be availed to

the public through the media and further research is recommended on how to improve peanut butter production as well as increase aflatoxin contamination and health risk awareness among processors and the public. Collaborative research is encouraged with international processors to compare notes in trying to address aflatoxin contamination levels.

1.5 Research Questions

1. Are there differences in levels of aflatoxin between local and international peanut butter in relation to set regulations?
2. Are processors aware of the dangers of consuming peanut butter with high aflatoxin levels and the health risks associated with it?
3. Do processors' production skills help in reducing aflatoxin contamination levels in peanut butter?

1.5.1 General Objective

To investigate aflatoxin levels and knowledge of production skills in peanut butter among processors.

1.5.2 Specific Objectives

1. To compare aflatoxin levels between local and international peanut butter products in relation to set standards.
2. To establish whether processors are aware of the dangers and health risks associated with consumption of aflatoxin contaminated peanut butter.
3. To assess processors' production skills in relation to reducing aflatoxin levels in peanut butter.

1.6 Ethical Consideration

Permission was sought from all respondents who agreed to take part in the study. Confidentiality was observed and there was no connection between the processor and peanut butter produced throughout the study. Processors were told that the benefit from the research would be that the study would educate them on the ways of improving their peanut butter processing so that their businesses would be acceptable on the international market. The study findings would also help the customers purchase safe products for the good of their health.

CHAPTER TWO

LITERATURE REVIEW

This segment reviews what has already been completed in some parts of the world. In this section, selected studies conducted across the globe have been cited in order to help guide the current research. Aflatoxin has been a concern around the world because of the negative effects that come by ingestion of food that have high aflatoxin contamination levels.

2.1 Aflatoxin and Processing

Aflatoxin is an inevitable, undesirable food contaminant, which occurs naturally (EFSA, 2007). Aflatoxins are secondary metabolites of the common food borne fungi *Aspergillus flavus* and *A. parasiticus* which colonize crops in tropical and subtropical regions throughout the world (Njumbe et al. 2014). Food is needed by all people hence unintentional ingestion of aflatoxin, is a challenge that demands for a consensus in addressing it both locally and internationally

The presence of aflatoxin in peanut is due to suboptimal processing techniques of the raw peanuts such as drying and faulty storage conditions (Traistaru and Moldovan, 2012). Lack of sunshine leading to failure of the kernel to completely dry, Mutegi et al. (2013b), and (Traistaru and Moldovan, 2012). Water leakages, storage duration, humidity, insect activity, temperature changes are major causes of mouldy development which is associated with aflatoxin contamination during storage period (Traistaru and Moldovan, 2012). These factors contribute to high levels of aflatoxin in processed peanut.

In a study that examined the financial risk associated with sorting of peanut along the marketing chain in Benin, West Africa; findings were that the costs of sorting and storage were leading factors in reducing aflatoxin levels in peanut but the practices of drying, sorting and storing pose financial risks to market traders of peanut (N'dede et al. 2012).

Aflatoxin in processed peanut can cause harm when accidental exposures are frequent such as cross contamination by other substances (Chang et al. 2013).

Therefore, peanut processing industries must follow strict guidelines to ensure compliance in harmonisation of international safety standards (Chang et al. 2013).

In a study that was conducted in Pakistan, 198 peanut products were commercially obtained from major cities of Punjab and analysed for aflatoxins using High Performance Liquid Chromatographic (Iqbal et al. 2013). Iqbal's study revealed that the mean concentration in raw peanut with shell was 6.4 µg/kg, raw peanut without shell 9.6 µg/kg, roasted peanut with and without shell 10.4 and 12.3 µg/kg, respectively, that for peanut butter was 2.4 µg/kg, peanut cookies was 4.6 µg/kg and peanut nimko had 3.4 µg/kg, (Iqbal et al. 2013). This study influenced the Government to enact regulation that encouraged market participants to reduce contamination (Iqbal et al. 2013). Governments could be influenced with increased evidence resulting from studies done within the nation showing a true picture on the ground similar to Iqbals.

According to (Ding et al. 2012) a study conducted in China, where different peanut products were tested for aflatoxin levels from four agro-ecological zones, revealed that there was low aflatoxin contamination in raw peanuts. However, higher aflatoxin levels were detected in peanut products, such as peanut oil, fried peanuts and roasted peanuts (Ding et al. 2012). These results suggest that it is possible that contamination could have occurred somewhere within the peanut processing value chain (Mutege et al. 2013a). This indicates that processing points too could have aflatoxin levels that may be different from other points of peanut butter processing. One could find different results if samples were taken at these different points of processing.

Another study carried out in four production regions in the State of São Paulo, Brazil, analysed the soil samples to determine the occurrence of fungal species that contributed to aflatoxin contamination (Atayde et al. 2012). According to Atayde et al. (2012), it was found that although *A. flavus* and *Aspergillus parasiticus* were isolated from peanuts, few samples were contaminated with aflatoxins. Therefore, the isolation of these species from soil showed that this is the main route of peanut contamination (Atayde et al. 2012).

2.2 Aflatoxin Measurements

Aflatoxin Global limits are specified by Codex Alimentarius Commission (CAC) in: CODEX STAN 193-1995; General Standard for Contaminants and Toxins in Food and Feed (FAO, 2003). The international regulation with regards to food safety limits has been set up in the CAC); which is a joint body, managed by FAO and WHO (Chang et al. 2013, CODEX, 2001). It is a recognized global food standard-setting body which was established to protect human, animal and plant life associates CODEX (2001) and EFSA (2007). The Codex Committee on Contaminants in Foods sets standards for contaminants in food based on scientific evidence and risk analysis (FAO, 2003).

Aflatoxin threshold value used to accept or reject peanut lots is equal to the EU maximum limit for raw and consumer-ready peanuts (Adams et al. 2004). For raw peanuts, 30 kg laboratory sample has to test less than or equal to 8 ng/g BI and 15 ng/g total aflatoxin (Adams et al. 2004). For consumer-ready to eat peanuts, all three 10 kg laboratory samples have to test less than or equal to 2 ng/g BI and 4 ng/g total aflatoxin Adams et al. (2004) and FAO (2003). The shipment can fail on the basis of the results for BI or total aflatoxin as described above (Adams et al. 2004). It is necessary for a nation to come up with safe limits that will be a reference point when need demands.

Whitaker et al. (1995), conducted a study to determine the probability of accepting and rejecting a lot based on guidelines from three countries, United States, United Kingdom and the Netherlands. In this study, for the United Kingdom which sets its unit guidelines at 10ppb, they determined that the probability of accepting good lots was 0.6334. “Studies on aflatoxin concentration in peanuts are often unreliable because of large sampling errors” (Dickens and T.B, 1975); concentration of aflatoxin can vary considerably from as low as below 10ppb to as high as 5000ppb or above. Consequently, many investigators often convert the continuous concentration of aflatoxin to binary variable in order to mitigate the large variances. Therefore, the focus of a number of studies has been on determining the probability of accepting and rejecting contaminated lots in various countries that import or export peanut butter.

In Zambia, the most reliable data on aflatoxin concentration seems to be the one by Juliet Akello's presentation on Aflatoxin Awareness & Management held at the stakeholders' meeting in Lusaka on 28th April, 2015 (ICRISAT, 2015) with the proportion of 32%, obtained from Chipata, Lundazi, Katete, Mambwe, Nyimba and Petauke.

However, smallholder production systems and small–medium enterprise tend not to follow regulations in doing business in processed peanut (Lamuka, 2014). The characteristics of processing industries hinder achievement of domestic and international processed peanut safety standards (Lamuka, 2014). Nevertheless, the recent developments in developing nations include increased emphasis on processed peanut safety regulations, strict processed peanut safety standards, reorientation toward preventive quality management, and a shift toward process-based standards and mandatory Hazard Analysis Critical Control Point (HACCP), (Lamuka, 2014). Indeed if these are in place, movement towards better aflatoxin level management may be realised. This process requires team work.

2.3 Aflatoxin and Health

Aflatoxins have been linked to immune suppression, increased susceptibility to disease (like in HIV and malaria). They are also implicated in compromised vaccine efficacy and stunted child growth and development (Bankole et al. 2005).

Different health conditions are associated with high levels of aflatoxin. Acute aflatoxin poisoning has been reported in many parts of the world including Taiwan, Uganda and India. The clinical symptoms of aflatoxicosis include vomiting, abdominal pain, pulmonary oedema, convulsions, coma and death with cerebral oedema and fatty liver, kidneys and heart (Liu and Wu, 2010). In addition, exposure to high levels of aflatoxin can result in acute human aflatoxicosis leading to jaundice, oedema, hemorrhage and eventually death (Nyikal et al. 2004, Probst et al. 2007, Shephard, 2008). There have been various reported outbreaks of human aflatoxicosis in Africa (Nyikal et al. 2004, Probst et al. 2007).

The Democratic Republic of Congo is amongst African countries listed with high prevalence of liver cancer (Kamika and Takoy, 2011). Many other studies have associated aflatoxins with undesirable outcomes in human populations and livestock

(Shephard, 2008, IARC, 1993, EFSA, 2007). Some of these negative outcomes include liver cirrhosis, liver cancer, immunity suppression, stunted growth and aflatoxicosis in animals and eventually death (Njumbe et al. 2014, Ismail et al. 2014). Aflatoxin has been implicated as causing cancer, and according to (EFSA, 2007) and Shephard (2008), even very low levels of exposure. “Although it is impossible to completely eliminate aflatoxin in food worldwide, it is possible to significantly reduce levels and dramatically reduce liver cancer incidence worldwide” (Liu and Wu, 2010).

Recent studies in Benin and Togo did a cross-sectional study among 480 children aged nine months to five years conducted to identify the effect of aflatoxin exposure on child growth (Egal et al. 2005). It was highlighted that health consequence of aflatoxin exposure, led to stunting in children where growth hesitance occurs at the time of weaning. The compromised growth was attributed to consumption of aflatoxin contaminated foods (Egal et al. 2005).

2.3.1 Global

Currently, over 5 billion people worldwide are at risk of chronic exposure to aflatoxins; and many studies provide clear evidence that consumption of aflatoxin contaminated foodstuff is one of the causes of liver cancer in humans in China and sub-Saharan Africa where in certain regions, at least 250,000 deaths occur every year (CODEX, 2001, Liu and Wu, 2010, Gong et al. 2002).

Peanut is one of the most important oilseed crops and snack foods in the world agro-food trade market. Peanut is also a product that is frequently implicated in aflatoxin contamination (UÇKUN and Işıl, 2014). In Haiti, samples of raw peanuts (n ¼ 21), peanut butters (n ¼ 32), and maize (n ¼ 30) were obtained in Port-au-Prince and Cap Haitian, during 2012 and 2013 in a study to monitor aflatoxin contamination (Schwartzbord and Brown, 2015). The outcome showed the concentration of total aflatoxins being greatest in peanut butter. This ranged from 137 mg/kg and 2720 mg/kg (Schwartzbord and Brown, 2015). This information is a revelation of increased aflatoxin in peanut butter.

A study conducted in Greece followed the production chain of pistachio nuts from farm to storage. This was to determine production steps, conditions handling practices that affect aflatoxin production (Georgiadou et al. 2012). The results

showed that the most critical step for aflatoxin contamination is maturity. If kernels are immature, they tend to enclose high levels of aflatoxin. This is why sorting is necessary if aflatoxin levels are to be reduced. This was the first stage that aflatoxin was detected above permitted limits that ranged from 11 mg/kg to 1361 mg/kg among orchards (Georgiadou et al. 2012). In addition to that, this study found that at harvest time, aflatoxin concentration was even higher reaching 1420 mg/kg. the study further detected heavy insect infestations in the orchard indicating a positive correlation between aflatoxin contamination and insect infestation (Georgiadou et al. 2012). At post-harvest time, aflatoxin contamination detected in the three out of four orchards, varied from 40 mg/kg to 1200 mg/kg, at drying and 650 mg/kg to 1100 mg/kg at storage (Georgiadou et al. 2012).

Another study was conducted in Brazil to evaluate the mycoflora and occurrence of aflatoxins in stored peanut samples from Tupã, State of São Paulo, Brazil where samples were analyzed monthly over a period of one (Nakai et al. 2008). According to Nakai et al. (2008), the results showed that the growth of *Aspergillus flavus* was mainly influenced by temperature and relative humidity.

In China, a study to determine aflatoxin levels in peanut and corn was done using ultra-high-pressure limit chromatography (Fu et al. 2008). Among the 16 peanut samples tested 12.5% were contaminated with aflatoxin (Fu et al. 2008). Among 18 corn samples, 22% were contaminated using the test method which is rapid, simple and accurate for monitoring aflatoxins in corn and peanuts (Fu et al. 2008).

2.3.2 Regional

Aflatoxin is a major problem in many countries. In a study conducted in Kenya to investigate market characteristics in peanut and their association with levels of aflatoxin revealed negative impact on food security (Mutegi et al. 2013b). In this study, it was found that 37% of the samples exceeded the 10mg/kg regulatory limit for aflatoxin contamination levels set by the Kenya Bureau of Standards (KEBS). The raw podded peanuts had the lowest $c \frac{2}{4} 167.78; P < 0.001$ levels of aflatoxin, with majority having levels of less than 4mg/kg and only 4% having more than 10mg/kg (Mutegi et al. 2013b).

Furthermore, these findings established that most aflatoxin-contaminated products were peanut butter and spoilt peanuts, whose recommended KEBS levels exceeded

10mg/kg (Mutegi et al. 2013b). According to Mutegi et al. (2013b), it was found that packaging materials significantly ($c = 2\frac{1}{4}$ 73.89; $P < 0.001$) influenced the amount of aflatoxin in the product, with majority of peanut samples stored in plastic jars having $>10\text{mg/kg}$ of aflatoxin.

A study conducted in Nigeria on dry roasted groundnuts to analyzed for moisture content, fungal populations and aflatoxin contamination found that moisture content in groundnuts varied as this is because mould counts, ranged between 3.6% to 102 colony-forming units per gram (Bankole et al. 2005). Aflatoxin B1 was found in more than 50% samples, in B2, G1 and G2 detection was 26% and below. The conclusion was that regular consumption of dry roasted groundnuts by Nigerians could present potential health hazards to consumers (Bankole et al. 2005).

Another study was carried out among 480 children aged nine months to five years in four agro ecological zones from Benin, Togo on dietary exposure to aflatoxin from groundnut identified the effect of aflatoxin on child growth (Egal et al. 2005). Among the findings, it was noted that higher frequencies of groundnut consumption correlated with higher socio-economic status in most of the survey area; people with higher disposable income were more inclined to buying a groundnut snack that is notoriously contaminated with aflatoxin (Bankole et al. 2005).

According to Bankole et al. (2005), even though, the etiology of kwashiorkor is still not yet clear, much higher aflatoxins have been found in the blood, urine and livers of children in Nigeria with the disease than in similar age matched children (Hendrickse, 1983) as cited in (Bankole et al. 2005). The presence of the toxin was established in the autopsy brain tissue of some Nigerian children (Oyelami et al. 1995) as cited in (Bankole et al. 2005). Moreover, a recent epidemiological study revealed a striking association between exposure to aflatoxins and growth stunting (Gong et al. 2002). Children in Zambia who fall in this category, similarly, are given peanut butter for nutrient addition to their meals as early as the time of weaning. On the contrary, such reports are not present.

In the Democratic Republic of Congo, a study was carried out to assess natural occurrence of aflatoxin B1 in raw peanuts. Sixty peanut samples were analyzed for aflatoxin B1, using Thin Layer Chromatography (Kamika and Takoy, 2011). The study analysis showed that aflatoxin B1 levels increased from the dry season to the

rainy season with values ranging from 1.5 to 390 and 12 to 937, respectively (Kamika and Takoy, 2011). Seventy percent of the peanut samples from both seasons exceeded the maximum limit of 5 mg/kg prescribed by the World Health Organization (CODEX, 2001).

2.4 **Zambian Safe Peanut Processing Guidelines**

In Zambia, in a bid to try and promote food safety in peanut, the Zambia Bureau of Standards regulates peanut by specifying requirements for safety. Legal specification for PB processing are under ZS723 of 2008 and ICS: 67.040, are PB Specification (ZABS, 2008) and (GRZ, 2001). These specifications apply to all levels of peanut processing. A mention of aflatoxin contamination maximum limit is also indicated as 15ppb in Table 1.

Table 1: Peanut Butter Specifications

Property	Requirements		Test method
	Smooth texture	Crunchy texture	
Free Fatty Acids (as Oleic Acid)% by mass, Max	1	1	AOAC 940.28 after extract as in AOAC 948.22
Fiber, % Max	2.5	2.5	AOAC 935.53
Peroxide value, mill equiva/kg, max	5	5	IUPAC 2.501
Moisture, % Max	2.0	2.0	AOAC 925.40
Total ash, % Max	2.5	2.5	AOAC 950.49
Fatty composition, % by mass	40-55	40-55	AOAC 948.22
Aflatoxins B1, B2, G1, G2 max	15ppb	15ppb	ISO 16050

Source:ZABS (2008)

2.4.1 **Challenge of Aflatoxin Control**

In Zambia the control of aflatoxin contamination may be a challenge due to:

1. Limited information to both processor and consumer of peanut butter leading to the need for more research to compare aflatoxin levels among various points of concern.
2. Lack of adherence to defined national specific maximum aflatoxin limits by authorities.
3. There is no clear guidance by government as to how aflatoxin problem can be addressed to protect the consumer.

2.5 Aflatoxin Reduction Measures

There are several ways that can be adopted to reduce levels of aflatoxin in crops. It has been reported (IITA) that sprinkling aflasafe in a field, two to three weeks before the flowering stage of maize prevents aflatoxin contamination while the crop remains in the field, and subsequently in storage (ICRISAT, 2013, EIARD, 2013). The report further says that even if the grains are not stored properly, or get wet during or after harvest, the product continues to prevent aflatoxin contamination. With what has been revealed by studies, this idea could be adopted for tropical countries. Only that sensitisation must increase hence the need for more to be done on aflatoxin research.

In order to forge ahead with aflatoxin reduction, partnerships need to be created between local and international research institutions and departments of agriculture in various states (Atayde et al. 2012). In addition, marketing agencies, NGOs, farmer groups, consumer groups, agrochemical manufacturers, and other stakeholders need to work as a team in order to develop strategies that address the problem of aflatoxin (Atayde et al. 2012).

Aflatoxin contamination can be avoided by not keeping processed peanut for more than a few months, storing in a dry, low humidity cool environment or freezer (Mutegi et al. 2013a, Kamika and Takoy, 2011). Furthermore, aflatoxin levels can be reduced by buying from known sources where processed peanut have been handled well. In addition, physical sorting and electric colour sorting removes infected peanuts which often have high levels of aflatoxin and improves trade system (N'dede et al. 2012, UÇKUN and Işıl, 2014, Schwartzbord and Brown, 2015).

To lessen outbreaks, cooperation among all stakeholders within a peanut processing chain and regulators to implement effective peanut safety programs (Chang et al. 2013).

According to a study conducted in Turkey on Monitoring of aflatoxin in peanuts, moisture can be reduced by irrigation to relieve stress and heat created in the soil due to draught and high temperatures (Dorner and Cole, 2002, Torres et al. 2014, Dorner, 2008). Moreover, it is stated that application of non-toxic strain of *A. flavis* and or *A. parasiticus* to the soil of the developing crop increases the spoil number leading to

competition for nutrients and naturally accruing toxins for growth on peanuts (Dorner and Cole, 2002, Zanon et al. 2013).

Further ways of reducing aflatoxin levels include avoiding direct contact of peanut with the soil, using appropriate technology and minimizing damage (EIARD, 2013, Chang et al. 2013). Using appropriate packaging materials and mode of transport, pest control, storing products in airtight bags in a well-aerated store can reduce aflatoxin levels significantly (Mutegi et al. 2013a). Moreover, it is indicated that cleaning stores before loading new products will help reduce aflatoxin in peanut (Chang et al. 2013).

In the study conducted among peanut handlers in Benin to try and assess Economic Risks of aflatoxin Contamination in Marketing of Peanut, it was found that most of the handlers were unable to (N'dede et al. 2012). However, the handlers could only pick signs of spoiled peanuts such as discoloration or insect damaged (N'dede et al. 2012).

Removing discoloured and mouldy grains physically can reduce contamination because a normal peanut appearance can be among the good ones without fungal infection signs (Mutegi et al. 2013b). Sorting is another helpful technique that is coming out as being associated with reducing aflatoxin levels in a situation where mature and immature peanuts are mixed (N'dede et al. 2012, Mutegi et al. 2013b, Schwartzbord and Brown, 2015). Sorting removes damaged, immature or rotten nuts; harvesting mature kernel reduces immature grain which are prone to increasing aflatoxin (Kabak et al. 2006).

Crop rotation and use of high quality seeds of early maturing has been suggested as one of the ways to reduce aflatoxin levels in peanut (UÇKUN and Işıl, 2014). Timely planting of peanut on fertile soils must be encouraged and there must be proper control of pests and diseases pre-harvest (UÇKUN and Işıl, 2014). During post-harvest stage, ensuring that timely harvesting, fast drying of groundnuts to below 10% moisture is essential.

Good pre-harvest practices to minimize aflatoxin contamination entails use of good agricultural practices including: proper disposal of previous crop remains (Mutegi et al. 2013b).

In Zambia, use of aflasafe, drying and storage practices in the 10 districts of Central and Eastern provinces, yielded positive results in reducing aflatoxin levels in maize and groundnuts for 2013 and 2014 harvest periods (Ismail et al. 2014). According to ICRISAT (2013), sprinkling aflasafe before and after harvesting crop has proved to reduce aflatoxin levels significantly as shown in a study that covered Nigeria, Kenya and Zambia in 2011. Sorting has been used and currently, sorted peanuts from Eastern province can be sold to South Africa (Mukuka and Shipekesa, 2013).

CHAPTER THREE

METHODOLOGY

3.1 Study Site

The study site was Lusaka urban district of Zambia which has a population of 1,747,152 (CSO, 2010). This site was chosen because it receives different brands of processed peanut butter from both local and international origins. In addition, the city was chosen because of the strategic position it commands in the country in relation to peanut butter processing and consumption patterns. Outlets from which these products were purchased included: processing plants (A, B and C) and retail sites (Commercial stores and Soweto market). Soweto market was purposively selected because of its popularity among consumers in the city. Soweto also serves as a wholesale site for retail outlets for other markets in Lusaka. Figure 3 is a map from Google Earth showing places from which peanut butter was purchased.

Map for Collection of Peanut Butter Samples

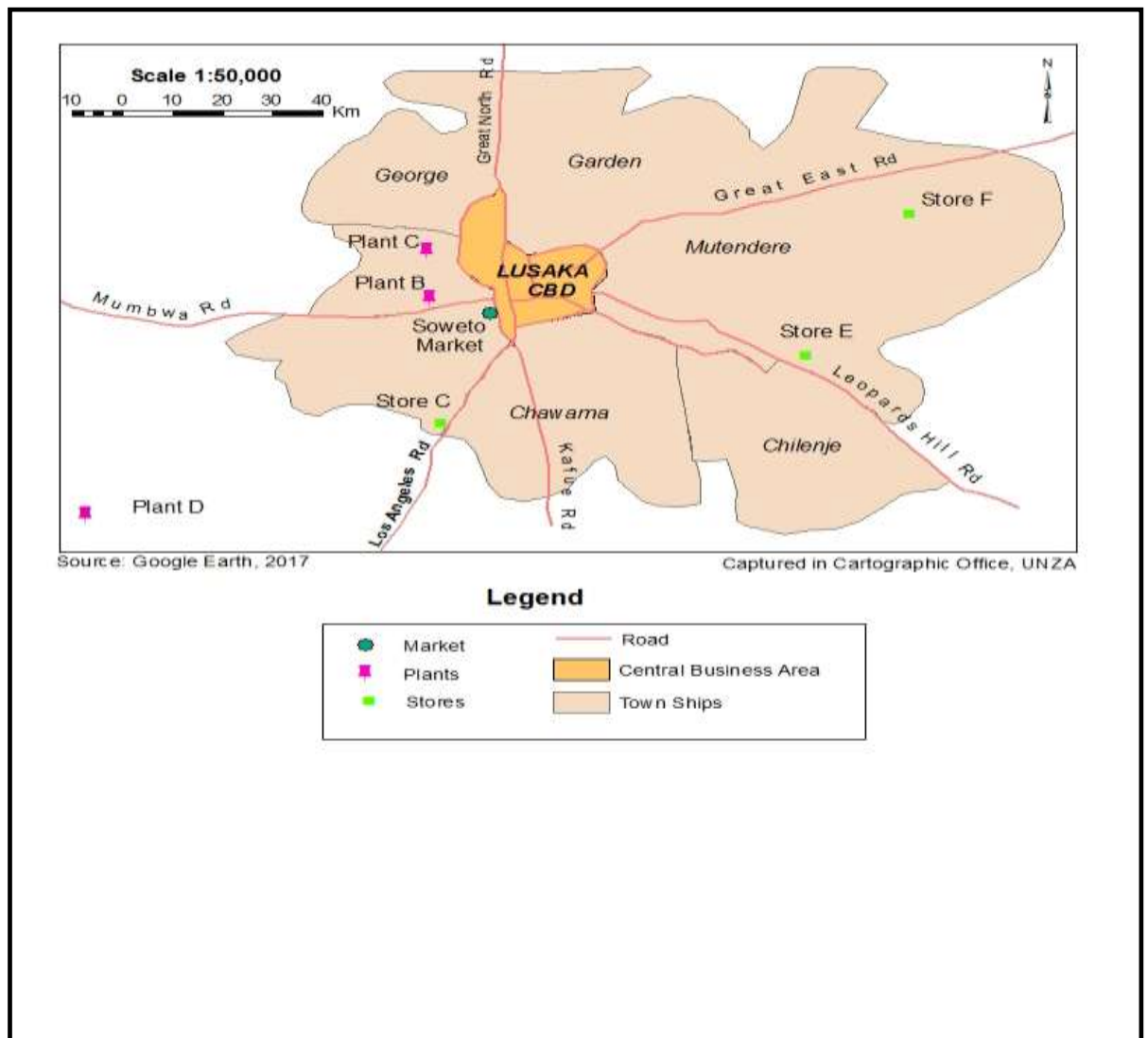


Figure 3: Map for Peanut Butter Sample Collection. Source Google Earth: Cartographic Office, UNZA (2017).

3.2 Study Design

This was a cross section survey research study between local and international peanut butter. Quantitative technique was used for canister peanut butter sample testing while for processor knowledge, qualitative method was used to help answer the questions.

3.3 Variables and Measurements

The variables included in Table 2 relate to both quantitative and qualitative data. In the quantitative are the dependent variable aflatoxin in peanut butter and the independent variables being set standards the international and local origin. For

qualitative variables, processing knowledge is the dependent variable and processor responses are the independent variables.

Table 2 illustrates the different variables and their indicators for measurements:

Table 2: Variables and Measurements

Variable Type	Operational Definition	Indicator	Measurement Scale
Dependent			
Aflatoxin in peanut butter	Presence of aflatoxin in peanut butter per ppb which may or may not be safe for human consumption	Aflatoxin presence in relation to standards: 1. European 2. CAC 3. Above 15ppb	Continuous
Processing knowledge	Ability to characterise good peanut for processing. Safe processing skills possession. Contamination and health risk awareness.	1. Characterise good and bad peanut 2. Processing skills 3. Sorting of peanut 4. Awareness of aflatoxin contamination and health risks	Responses

3.4 Sampling Procedure

A sample size was needed to compare aflatoxin contamination levels from samples of local and international peanut butter, rather difficult task. A study recorded in literature review on page 7 by Whitaker et al. (1995) will be used to help determine the probability of accepting and rejecting aflatoxin contaminated samples. In their study, for the United Kingdom which set its guidelines at 10ppb, they determined that the probability of accepting good lots was 0.6334. “Studies on aflatoxin concentration in peanuts are often unreliable because of large sampling errors” (Dickens and T.B, 1975); concentration of aflatoxin can vary considerably from as low as below 10ppb to as high as above 5000ppb. In this study, binary variables were used to compare aflatoxin contamination levels but some descriptive statistics were reported on continuous concentration of aflatoxin. In Zambia, to determination sample size, we used sample prevalence of 32% for the local and 63% for the international sample units based on cited literature on same page.

3.4.1 Sample Size

A sample size of 53 peanut butter canisters in each group (local and international), was calculated using the formula from Rosner's book and ed. (Rosner, 1990) in this way:

Let:

- P_L = probability that a randomly chosen canister of local peanut butter is accepted
- P_I = probability that a randomly chosen canister of international peanut butter is accepted
- X_1, X_2, \dots, X_n be measurements of aflatoxin levels in a random sample of canisters of size n for local peanut butter
- We assume the sample is drawn from a normal distribution with mean μ_1 and variance σ^2 .
- Y_1, Y_2, \dots, Y_n be measurements of aflatoxin levels in a random sample of size n of canisters for international peanut butter.
- We assume the sample is drawn from a normal distribution with mean μ_2 and variance σ^2 .
- We assumed variation of aflatoxin contamination levels was the same for both local and international peanut butter.

Using 10ppb as the acceptable standard for both local and international products, the values of P_L and P_I become:

$P_L = \Pr(X < 10)$ and $P_I = \Pr(Y < 10)$, where X and Y were measurements of aflatoxin levels in canisters of peanut butter for local and international samples, respectively. The sample size formula for comparing two binomial proportions is given by:

$$n_k = \frac{\left[z_{1-\alpha/2} \sqrt{\bar{p}\bar{q}\left(1+\frac{1}{k}\right)} + z_{1-\beta} \sqrt{p_1q_1 + \frac{p_2q_2}{k}} \right]^2}{\Delta^2}, \text{ general sample size formula.}$$

$$n = \frac{\left[z_{1-\alpha/2} \sqrt{2\bar{p}\bar{q}} + z_{1-\beta} \sqrt{p_1q_1 + p_2q_2} \right]^2}{\Delta^2}, \text{ specific formula for } k = 1$$

Where:

$$n_I = kn_L = n \text{ for } k = 1, \text{ i.e., equal sample sizes.}$$

$$P_I = P_L = \Pr(X < 10) = 0.32, \text{ for local}$$

$$P_2 = P_I = \Pr(Y < 10) = 0.63, \text{ for international.}$$

Initially, we assume there is no difference between local and international aflatoxin contamination levels so that $P_1 = P_2 = P$ under H_0 . The estimate of P is the average of P_1 and P_2 . Will denote the average by \bar{p} .

$$\bar{p} = \frac{P_1 + P_2}{2} = \frac{0.32 + 0.63}{2} = 0.475 \quad \bar{q} = 1 - \bar{p} = 0.525$$

$$P_1 - P_2 = \Delta = 0.31$$

$$Z_{1-\alpha/2} = Z_{0.975} = 1.96, \text{ fixing Type I error at } \alpha = 0.05 \text{ (5\%).}$$

$Z_{1-\beta} = Z_{0.80} = 0.84$, fixing Type II error at $\beta = 0.1$ (10%), i.e. power at 90%.

Substituting these values in the formula we obtained:

$$\begin{aligned} n &= \frac{[1.96\sqrt{2} * 0.475 * 0.525 + 1.28\sqrt{0.32 * 0.68 + 0.63 * 0.37}]^2}{0.31^2} \\ &= 1.384195795 + 0.859317682 = 2.243513478^2 \\ n &= \frac{5.0333527525}{0.0961} = 52.37619901 \end{aligned}$$

Therefore, $n = 53$ peanut butter canisters from each origin.

3.4.2 Sample Size Distribution

Having determined the sample size, samples were distributed to the target population consisting of one market and three plants, for the local main outlet-population and three commercial chain stores for the international main outlet- population. The choice of the plants, market and commercial stores as primary sample areas were purposively done because these were the major suppliers of peanut butter for the majority of people. The three main outlets formed the strata for the population. The strata covered one market (A), with 14 stands as sub-outlet points which provided canisters of peanut butter for testing and three processing plants (B, C and D), each plant acted as a sub-outlet point that provided canisters for peanut butter tested for local samples. And the three commercial stores (E, F and G) picked from the 20 distribution points acted as sub-outlets as well for the international canisters for peanut butter testing.

The sample size of 53 was apportioned equally to the local outlets, the three processing plants and the market. This meant that $53/4 = 13.25$ or 14 canisters were to be sampled from each sub-outlet for the local products. However, in one outlet among the plants, there was a short fall of two canisters and in another outlet we had

one extra canister. For Soweto, processor stands acted as sub-outlets in providing the 14 samples. Thus, a total of 55 instead of the calculated size of 53 samples were purchased to represent local products.

For the international products, 53 canisters were proportionally allocated to the stores based on the number of outlets each store had. Store E had nine outlets, F had six and G had five bringing the total to 20. This led to the following proportional allocation; E 24 ($9/20 \times 53 \approx 24$), F 16 and G 14, bringing the total sample size to 54. The distribution of sample size is summarised in Table 2 below.

Table 3: Actual Sample Size for Peanut Butter Canisters

Outlets	ID	Origin	Sample size
1	A	Local	14
2	B	Local	12
2	C	Local	15
2	D	Local	14
3	E	International	$(9/20 \times 53) = 23.85$ or 24
3	F	International	$(6/20 \times 53) = 15.9$ or 16
3	G	International	$(5/20 \times 53) = 13.25$ or 14

3.4.3 Canister Sample Selection Procedure

Actual stores, sub-outlets, were randomly selected from the developed list: for store E, one of the nine was selected, one out of six for store F and one out of five for store G. In these selected sub-outlets, a list of peanut butter canisters was developed to assist in selecting desired canisters using a systematic random sampling method of one in two. This selection was irrespective of texture or brand. However, data on texture or brand were recorded merely as characteristics of a canister.

All the three processing plant owners could not allow investigator to enter their warehouses to randomly sample units. Managers instead were instructed to systematically sample the number of canisters that were needed which they did. At Soweto market, samples of peanut butter were collected from processors who were available at the time of data collection using systematic sampling of processors. Purchased peanut butter samples were kept at room temperature for two weeks

before taking them to the laboratory for testing to keep samples close to the way they were at the time of purchasing. This was also a period to wait for the reagents to come from outside the country for testing samples.

3.4.4 Canister Coding and Data Collection

A checklist was used to capture sample numbers (1 to 109), brand name, date of collection, origin (local and international), main outlets assigned 1 to 3 (market, plants and commercial stores), sub-outlets for the stores (E to G) and individual stands for outlet A where each of the 14 processors supplied 1 peanut butter canister; texture (crunchy or smooth), shelf life (manufacturing and expiry dates). Proper labelling (coding) of samples was done to avoid mix up of products from different collection points and for easy identification. Each canister was identified by its main outlet and sub-outlet for commercial stores and the sampling number. For example, sub outlet D would be assigned label 2 (main outlet) and 3 (sub-outlet) and canisters number 1 (of the 14 samples collected). A canister with an identification number 2, 3 and 1 would therefore be indicated as a sample from the main outlet 2, for sub-outlet 3 (processing plant) and sample number 1. The same trend repeated itself throughout the sample unit collection.

3.5 Testing Peanut Butter Samples

At the time of testing, peanut butter samples were collected and taken to test for aflatoxin concentration levels at the University of Zambia's School of Agriculture and Microbiology Soil Laboratory using a checklist. Peanut butter produced both locally and internationally were tested using NEOGEN Reveal Q+ Test Neogen (2015) based on European standard of 4µg/kg and 15ppb Codex Alimentarius Commission (CAC) as acceptable maximum limit for safe levels (Otsuki et al. 2001, FAO, 2003). Each canister yielded two samples for testing to see variation within the canister making the actual tested peanut butter, of 20g each, totalling 218.

3.5.1 Sample Preparation and Extraction

Peanut butter was tested using Reveal Q+ Test which is a Competitive Direct Enzyme-Linked Immunosorbent Assay (CD-ELISA) in order to obtain aflatoxin concentration in parts per billion (ppb). The device is an immunoassay technique used for rapid detection and quantification of Aflatoxin levels in processed and

unprocessed foods. The tests were done by the investigator with the help of the laboratory technicians for the 109 peanut butter canisters.

Materials needed for conducting the test included, 65% ethanol solution, sample collection cups with lids, a scale, a timer, sample cup rack, Aflatoxin Stat tablet reader, graduated cylinder, filter paper, sample collection tubes with caps, 100 μ L pipettes with tips, 500 μ L pipette, tips, paper towels and gloves. In addition, Neogen test strips, sample diluents, clear sample cups and a reader were also used.

3.5.2 Testing Procedures

An appropriate number of clear sample dilution cups were labelled and placed into a rack in readiness for mixing and to avoid disorder. A total of 20 grams of peanut butter were weighed in cups and mixed with 60ml of 65% ethanol which were shaken vigorously for 3 minutes to extract Aflatoxin from the mixture. The sample was then allowed to settle for 1 minute or more depending on the thickness of the peanut butter which was later filtered. Thereafter, 500 μ L of sample diluents was put in a clear cup using a 500 μ L pipette where 100 μ L of sample extract was added and mixed by pipetting up and down for 5 times. Thus from this mixture, a 100 μ L was taken and transferred in a new clear sample cup where a test strip end was inserted into the sample cup. The timer was set for 5 minutes in readiness to test Aflatoxin concentration while being checked to ensure the strip was in contact with the liquid and had begun to wick. A timer was pressed to note the start of the 5 minutes' period for the strip to develop after which the strip was removed from the sample cup.

3.5.3 Reading Test Results

Stat reader was turned on, the test type selected and vital data entered, for instance, name of the project, which samples are being tested, if from Soweto outlet, then SO was picked from the reader tablet menu making sure that the choice matched the sample numbers (1 to 14) being tested. The strip was then fully inserted into a white cartridge adapter with the sample end first and lines facing upside-down. The cartridge was inserted into the Stat reader which automatically began analysing the strip. The reader analysed the test and the result was displayed on the screen which was saved on the tablet for storage but also recorded on a hard copy for backup. The reading of results was done within 1-minute completion of the 6-minute incubation.

Hence total aflatoxin B1 was read and used instead of the individual B1 and B2 and G1 and G2 as described in other researches. This was because the machine could not detect specific aflatoxin types.

3.6 Data Entry and Analysis

Collected quantitative data for aflatoxin concentration levels were compared and rechecked from both the tablet and the hard copy that were used. Data was coded and cleaned up before analysis. Data was entered into Excel and later transferred into SPSS version 20 which was used for statistical analysis. Descriptive summaries of the concentration of aflatoxin for both local and international peanut butter were produced. Comparisons between local and international concentration of aflatoxin were done using two sample t-test. Tests were also performed to compare proportions of samples that met specific set categories between local and international data sets. The categories considered were 0 - 4ppb and 0 - 15ppb. All tests were performed at 5% level of significance. Estimates of 95 percent confidence intervals were also reported.

3.7 Pre-testing the Tool

A pilot study was conducted on two company respondents from Zambia Milling and Quality Feeds who helped provide information that was used to refine the semi structured questionnaire in this study. The final data collection tool consisted of three parts. First part dealt with the demographic characteristics of the respondents, the second part dealt with information about aflatoxin contamination and health risks, the last part dealt with issues related to the peanut butter processing skills, inspection and distribution.

3.8 Qualitative Data Collection and Analysis

A semi structured guided questionnaire was used by the interviewer to capture qualitative data on safe processing skills, aflatoxin contamination and health risks to humans from processors. Interviews were done for triangulation of aflatoxin data between what was revealed in quantitative and what in fact was on the ground practically. All respondents were given information about the research and upon understanding and agreeing to participate, they each signed the consent form. These were interviewed and no audio recording was done. Qualitative data was analysed

using themes and texts derived from the respondents to give meaning and implications in the presentation of data. Participant and outlet identity was by use of anonymity, only the researcher and team knew who these were.

3.9 Eligibility Criteria

- Local peanut butter samples from Soweto market and processing plants were considered irrespective of brand or texture.
- Only international peanut butter available in the commercial stores at the time of the study were included.
- Processors who signed the consent form participated while, those who refused were excluded.

A total of 109 peanut butter canisters were included in the study and tested for aflatoxin contamination. Sixteen local processors from the processing plants and the market were interviewed.

CHAPTER FOUR

STUDY RESULTS

4.0 Introduction

This chapter highlights the findings of both quantitative and qualitative approaches of the survey. The first section presents the quantitative results by looking at descriptive summaries of aflatoxin contamination levels in peanut butter, and ends with inferential results. Second section presents the qualitative results of the study.

4.1 Quantitative Results

4.1.1 Descriptive Summary

In this section we looked at aflatoxin contamination in terms of the mean, variance irrespective of the characteristics of the samples. The section ends with the report on the contamination in relation to texture and shelf life.

4.1.1.1 Centrality and Variation

Table 4 below gives descriptive summaries consisting of sample size, mean, minimum, maximum and standard deviation by origin and outlet. Outlets A to D provided local products and outlets E to G provided international samples.

Table 4: Descriptive Summary of Peanut Butter

Origin	Outlets	N	Aflatoxin concentration in ppb			
			Mean	Min	Max	Std. Deviation
Local	A	14	32.50	3.30	147.20	46.848
	B	12	6.32	3.70	9.15	1.690
	C	15	68.38	42.90	122.50	25.104
	D	14	13.06	8.85	16.30	2.275
International	E	24	6.40	3.25	32.95	5.859
	F	16	5.36	3.95	8.10	1.194
	G	14	5.54	1.75	10.35	2.166
	Total	109	18.86	1.75	147.20	28.738

The results show that the amount of aflatoxin concentration in our samples had a minimum of 1.75ppb (G) and a maximum of 147.2ppb (A). Generally, high level values of aflatoxin were observed in samples from local outlets and low values in those from international outlets. Samples from outlet (F) had the lowest average amount of aflatoxin (5.3625ppb) while samples from outlet (C) had the highest average amount of aflatoxin (68.38ppb). There was a lot more variation in local samples than in international samples. For local samples, variation ranged from 1.69ppb to 46.85ppb while for international samples it was from 1.19ppb to 5.86ppb.

There were 55 (50.5%) samples from the local and 54 (49.5%) samples from international products. Ignoring outlets, the international peanut butter products yielded the lowest values with a minimum of 1.75ppb to a maximum of 32.95ppb. This data is summarized in Table 5 below.

Table 5: Descriptive Summary of Aflatoxin Concentration

Origin	No.	Minimum	Median	Maximum	Mean	Std. Deviation
International	54	1.75	4.92	32.95	5.87	4.085
Local	55	3.30	13.10	147.20	31.62	36.066
Total	109	1.75	6.95	147.20	18.86	28.738

The median and the mean are reasonably close for the data from the international sample but quite apart for the local sample as seen in Table 5. The standard deviation of aflatoxin contamination for the local sample is about nine times greater than that of the international.

4.1.1.2 Aflatoxin Contamination in Relation to Texture

In our samples, texture had two categories, crunchy and smooth. The sample yielded more of smooth canisters of peanut butter than crunchy. There were 85 (78%) smooth canisters and 24 (22%) crunchy. Table 6 shows aflatoxin concentration between local and international peanut butter in relation to crunchy and smooth textures. For the international peanut butter, 14 (12.8%) were of crunchy texture with a minimum aflatoxin concentration of 1.75ppb to a maximum of 32.95ppb. The mean for this texture was 7.59ppb with a standard deviation of 7.63ppb. For the smooth texture, there were 40 (36.7%) canisters with a minimum of 3.2ppb and a maximum of 10.35ppb and a mean of 5.2650ppb. the standard deviation was much lower than for the crunchy texture, 1.36ppb.

For the local products, 10 (9.2%) were of crunchy texture with a minimum concentration of 43.15ppb and a maximum of 83.55ppb and a mean of 59.4150ppb. For the smooth texture, 45 (41.3%) had a minimum of 3.30ppb and a maximum of 147.2ppb with a mean of 25.4467ppb. Comparing with the international products, the standard deviations were higher. This data can be examined in the table below.

Table 6: Peanut Butter Contamination in Relation to Texture

Origin	Texture	N (%)	Minimum	Maximum	Mean	Std. Deviation
International	Crunchy	14 (12.8)	1.75	32.95	7.5929	7.62695
	Smooth	40 (36.7)	3.20	10.35	5.2650	1.35747
Local	Crunchy	10 (9.2)	43.15	83.55	59.4150	12.62744
	Smooth	45 (41.3)	3.30	147.20	25.4467	36.73147
Combination Texture	Crunchy	24 (22.0)	1.75	83.55	29.1854	27.86372
	Smooth	85 (78.0)	3.20	147.20	15.9494	28.46507
Total		109 (100.0)				

Crunchy peanut butter in local products showed high values in both the minimum and maximum readings, 43.15ppb and 83.55ppb, respectively. The mean for crunchy samples was much higher than for smooth, 29.18ppb compared to 15.95ppb ignoring the origin. The difference in variation between crunchy (27.86372ppb) and smooth texture (28.46507ppb) was not much, while the mean for crunchy 29.1854ppb was higher than that for the smooth texture (15.9494ppb).

4.1.1.3 Aflatoxin Contamination in Relation to Shelf-Life

In the process of data collection, it was discovered that manufacturing and expiry dates were missing in some canisters for both local and international products. This made it difficult to calculate shelf-life for all the canisters. However, in 51 sample units, information was available on manufacturing and expiry dates. The average shelf life ranged from 274 days to 547 days which is about a year.

4.1.1.4 Aflatoxin Contamination in Relation to Standards

The results here describe percentages of aflatoxin contamination in three categories, 0 - 4ppb, 4 – 15ppb and over 15ppb as shown in. Table 7 below.

Table 7: Proportion of Contamination in Specific Standards

Origin	Range of Aflatoxin Contamination (%)			Total
	0 - 4ppb	Above 4 - 15ppb	Over 15ppb	
International	7 (6.4)	46 (42.2)	1 (0.9)	54 (49.5)
Local	2 (1.8)	28 (25.7)	25 (22.9)	55 (50.5)
Total	9 (8.3)	74 (67.9)	26 (23.8)	109 (100)

Category 0 – 4ppb

The second column, of Table 7 presents percentage of sample units that were within this category for the European standard. There were 6.4% (international) and 1.8% (local) sample units that met this standard.

Category above 4 to 15ppb

In the third column we have percentages of sample units that were above 4 to 15ppb category, which represents part of the CAC standard (0 - 15ppb). There were 42.2% (international) and 25.7% (local) sample units that met this standard.

Category above 15ppb

In the fourth column, we have percentages of sample units that were above 15ppb. This category represents products unsafe for human consumption. There were 0.9% (international) and 22.9% (local) sample units. All together, this category had 26 sample units (23.9%).

The findings show that, regardless of origin, only 9 (8.3%) of the 109 (100%) peanut butter samples satisfied the European set standard (0 - 4ppb) total aflatoxin B1 as safe for public consumption. This indicates that 100 (91.7%) samples of peanut butter from both local and international were contaminated beyond the European standards. If we use the 15ppb CAC standard (0 - 15ppb), 83 (76.1%) regardless of sample origin were safe for consumption. Of these, 53 (48.6%) were internationally produced and 30 (27.5%, locally produced).

4.1.2 Inferential Results

The preceding descriptive section has shown that there is some evidence of a difference in the levels of aflatoxin contamination between the local and international

peanut butter. In the section that follows, we determined the statistical significance of the difference seen in the descriptive section. The inferential section compared proportions of sample units that met safe standards between the local and international peanut butter irrespective of the characteristics of canisters. A parametric approach was used to carry out the test.

4.1.2.1 Parametric Test of Aflatoxin Contamination by Origin

To compare proportions of peanut butter that meet safe standards between local and international products, we carried out a two-sample t test of proportions. The test requires that data be normal and equal variance between local and international population of peanut butter be assumed to hold.

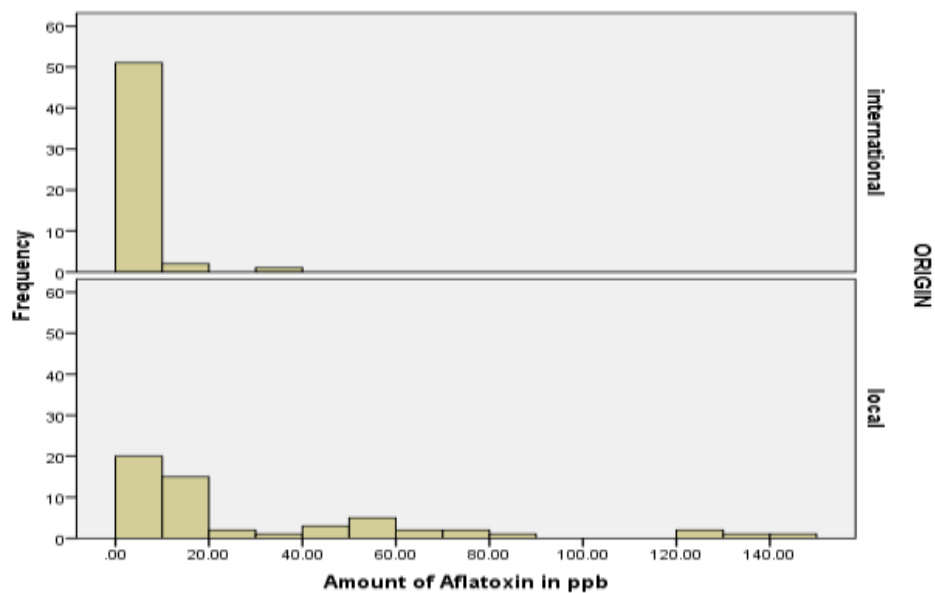


Figure 4: Aflatoxin Concentration by Origin

In Figure 4 we have histograms of aflatoxin concentration for both international and local data sets. The upper panel in the figure shows the histogram for the international data set while the lower panel shows that for the local data set. Normality assumption is not satisfied because there is skewedness to the right in both histograms. Furthermore, variation is higher in the local as compared to the international data set. For the international peanut butter, most of the values are in the range 0 to 10ppb while for the local, most of the values are between 0 and 20ppb but values go as higher as above140ppb.

4.1.2.2 Log of Aflatoxin Contamination by Origin

To achieve normality, the dependent variable was transformed by taking a natural log. The transformed dependant variable, Ln (Aflatoxin in ppb), appeared to satisfy the normality assumption and a common variance could also be assumed, as seen in Figure 5.

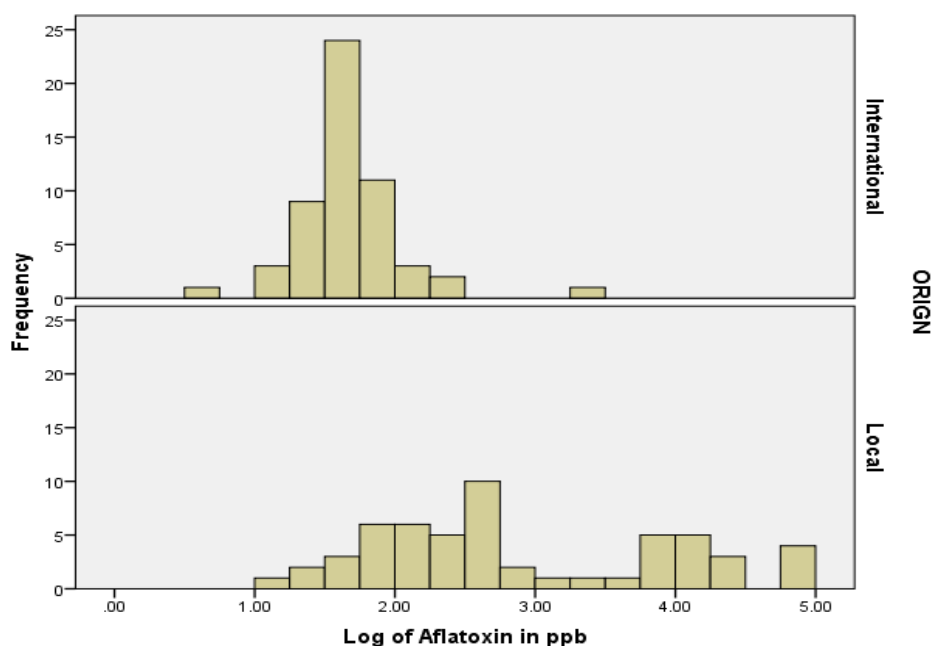


Figure 5: Log of Aflatoxin Concentration by Origin

A comparison of aflatoxin levels between local and international samples was done using the transformed variable. Details of the calculations are in Appendices 8 which shows that using the log of aflatoxin, the hypothesis that the mean values between local and international peanut butter are the same is rejected. The observed t value was -8.019 $(-1.22426/0.15266)$ which is outside the interval $(-1.98, 1.98)$, suggesting that the result is highly statistically significant with a very small p-value of less than 0.00001. The 95% confidence interval of the difference -1.22426 was estimated to be $(-1.52690, -0.92163)$. This also confirms the significant result since 0 is not contained in the interval.

4.1.2.3 Parametric Test of Aflatoxin Contamination by Set Standards

Further, tests were carried out to compare a difference in proportions between local and international peanut butter products category by category as defined in Table 7.

European Standards (0 to 4ppb)

In Table 7, for international sample units 7 out of 54 were within 0 – 4ppb while for the local, only 2 out of 55 did so. The difference in proportions was -0.094 ($2/55 - 7/55$) with an estimated standard error of 0.053. These values gave a Z-score of -1.77 with an estimated 95% confidence interval of -0.011 to 0.197. This result shows that there was no significant difference (p-value = 0.0768) in aflatoxin contamination levels between the local and international products using the European set standard of 0 – 4ppb. This means that using the 0 – 4 standard, there was no sufficient evidence that the levels of contamination are any different between local and international peanut butter.

CAC Standard (0-15ppb)

However, using the 0 to 15ppb standard, a significant result was observed with a p-value of less than 0.00001. The difference in proportions was -0.43603 with an estimated standard error of 0.07081, these values yeild a Z-score of 6.158 with an estimated 95% confidence interval of 0.29566 to 0.57640. Thus, aflatoxin concentration was significantly different between local and international products using this range. Appendix 6 gives the detailed calculations for the two categories mentioned here.

4.2 Qualitative Results

4.2.1 Introduction

This section reports the information gathered from processors regarding knowledge on aflatoxin contamination, health risks and safe processing skills of peanut butter. The first part dealt with demographic characteristics of the processors and proceeded to discuss knowledge of aflatoxin contamination, health risks and safe production skill for peanut butter.

4.2.2 Demographic Characteristics of Respondents

Overall, there were 15 males and one female participant whose ages ranged from 21 to 53 years old. Majority, seven, of the participants were aged between 40 and 44 years followed by four participants aged between 25 to 29 years. Two respondents were aged between 30 and 34 years, one was aged between 35 and 39 years. One respondent was above 50 years and one was below 21 years old. Out of 17 processors and retailers contacted, 14 were from the market while three were from processing plants. From the three plants, two key informants were interviewed while

one declined and all processors from the market were interviewed. Regarding education, majority, nine participants reported having attended up to secondary school level, four attended tertiary education while three went up to primary school. No one reported not having gone to school among processors. These characteristics are summarised in Table 8 below.

Table 8: Demographic Characteristics of Processors

Characteristic	Age Category						Total	
	< 24	25 - 29	30 - 34	35 - 39	40 - 44	Above 50		
Education	Primary	1			1	2	4	
	Secondary		3			2	1	6
	Tertiary		1	2		3		6
Sex	Female				1			1
	Male	1	4	2		7	1	15
Outlet	Soweto	1	4	2	1	5	1	14
	Market							
	Processing Plants					2		2

4.2.3 Knowledge of Aflatoxin Contamination

In terms of aflatoxin contamination awareness, the results showed that there were differences in the level of knowledge on what aflatoxin was among the respondents at plants and Soweto market. Plant processors had a higher knowledge and stated that aflatoxin was a “poison” compared with the market processors who gave varied responses. Upon being probed, some said it was a “disease” others said it was a “poison.” and some had “no idea.” The variation in responses indicated that there was little or no knowledge about aflatoxin contamination among market processors compared to those from the plants.

On the other hand, when participants were asked on how they identified or distinguished between good and bad groundnuts before processing peanut butter, their responses do not seem to vary much as evidenced by Table 9 below:

Table 9: How Processors Distinguished Good from Bad Groundnuts

Respondents	Bad Characteristics	Good Characteristics
Plants	<ul style="list-style-type: none"> - Rotten groundnuts - Affected groundnuts - Very thin groundnuts 	<ul style="list-style-type: none"> - Sorted by farmers who are asked to Follow good harvesting practice. - Good to eye - Of high quality - Broken groundnut seeds
Market	<ul style="list-style-type: none"> - Old groundnuts - Rotten - Green, black and brown type - G/nuts that attach to each other. - Bad smelling groundnuts 	<ul style="list-style-type: none"> -New groundnuts -Clean looks/good to eye -G/nuts with more fat - groundnut seeds -Small kadononga type -Makulu red -Groundnuts which can stay for nine months

At Soweto market, respondents classified bad groundnuts as those that were “rotten”, “old groundnuts”, “green, black and brown groundnuts”, ‘groundnuts that attached themselves to each other”, “bad smelling groundnuts” and chalimbana type. Plants classified bad groundnuts as “affected groundnuts”, “very thin and rotten types.” Plants characterised good groundnuts as those that “looked good to the eye”, “ones that were sorted by farmers,” “high quality groundnuts” and even “broken groundnut seeds” were listed as good upon probing. Soweto market processors characterised good groundnuts as “new groundnuts”, “looking clean and good to the eye”, “ones which could stay for nine months without getting bad”, “Makulu red”, “small Kadononga groundnuts” and “broken groundnut seeds” these were safe for use in processing peanut butter. Results show that both plants and Soweto market characterised good groundnuts as those that are good to the eye, but also on being probed, they stated that broken seeds were good for peanut butter processing. Soweto processors added more to the good groundnuts group by also mentioning “kadononga” and “Makulu red” type of groundnuts as the best for processing peanut butter since they have more fat and fry well. These types of groundnuts produce good and smooth consistent peanut butter which never hardens unlike chalimbana which has less fat.

4.2.4 Knowledge of Aflatoxin Health Risks

Crucial to the reduction of aflatoxin contamination in peanut butter, is the knowledge of health risks associated with aflatoxin. Below are the verbatim accounts of what

was reported by respondents regarding broken groundnut seeds, very thin groundnuts, health risks and sorting of groundnuts.

On broken groundnut seed one respondent had this to say:

“Broken groundnut seeds are not a problem as long as they look good, we use them to process peanut butter and some of them get broken by the machine during shelling.” (Male plant Respondent number 16)

Still another respondent contributed and mentioned that:

“Broken groundnut seeds have no problem, we use them to make peanut butter because they are okay.” (Male Respondent number 5 from Soweto).

Further, when processors were queried on what they did with very thin groundnuts that they sorted out, it was interestingly reported that these were given to those who could consume them:

“Very thin groundnuts are sorted but are not thrown away. These are given to others who can eat them.” (Male Respondent number 14 from Soweto).

The discourse above indicate that there is little or no knowledge of aflatoxin health risks in the processing plants and market. This was evident in the choices they made when given a list of possible health risks, the processors were hesitant to identify what these were. Below is what one respondent echoed:

“Health risks! I do not know what health risks can be brought about by eating contaminated peanut butter but there are people who are sick. This is why government spends a lot of money treating people in the hospitals who suffer from diseases caused by aflatoxin.” (Male Respondent number 16 from plant).

Information was solicited from the processors on how they could detect good from bad groundnuts used for processing peanut butter. Their general response was that they did so by sorting out or eliminating affected groundnuts. One of the respondents had this to say:

“By always asking farmers to follow good harvesting practices. We do sorting of groundnuts after receiving them from farmers For eliminating affected groundnuts.” (Male Respondent number 15 from plant).



Figure 6: Sorting Groundnuts before Processing Peanut Butter at a Plant
Picture by: Maureen Samson Banda (2016).

Sorting of groundnuts was particularly important among processors both in the market and the plants as Figure 6 seems to show.

4.2.5 Safe Peanut Butter Production Skills

In this part, we look at skills that would help processors produce peanut butter that has reduced aflatoxin contamination. To appreciate these safe skills, we looked at cleaning of the processing machine, peanut butter processing steps, inspection by health officers and distribution of the same.

4.2.5.1 Cleaning of the Processing Machine

Processors were asked whether they cleaned the processing machines, how they did it and how often. This study found that plant processors were in a habit of routinely cleaning the machine every time after processing while among Soweto processors, there were varied responses. The findings demonstrate that the level of cleaning processing machines is poor among Soweto market processors compared with the

plant processors. Frequency of cleaning included daily, every other day, three times per week due to busy schedules, twice per day and still others did it every they used the machine. Some processors never cleaned their machines at all for fear that they might get damaged since grinders were electrical. One of the respondents said:

“I clean the machine sometimes but not every day because of the busy schedules and one cannot use water to clean for fear that the machine would be damaged.” (Male respondent number 11 from Soweto).

Another respondent stated that:

“I clean the machine every day because if it is dirty, customers will not buy peanut butter. I clean by wiping with a damp cloth.” (Male Respondent number 2 from Soweto).

4.2.5.2 Processing Steps of Peanut Butter

Another interesting aspect of the findings was that a respondent was asked to state what steps he took to process peanut butter with reduced aflatoxin contamination. After stating the steps, he wanted to know the consequences for those whose peanut butter would be found with high aflatoxin contamination levels. In his words this is what he wondered about:

“What will happen to processors whose peanut butter will be found to have high aflatoxin contamination? And what precautions will those making laws put to ensure peanut butter that is made here has low aflatoxin levels if they are not going to be around to see what we do?” (Male Respondent number 12 of Soweto).

There are about 12 steps of processing raw groundnuts into peanut butter among which include: storage, sorting, roasting, cooling, blanching, grinding, mixing, and filling in canisters before distributing. There were some variations between plants in some stages of processing. Clearly plants are highly mechanised than the processing in Soweto market so that some stages are more elaborate in plants. Pictorially, the processes can be viewed as shown in the flow chart in Figure 7.

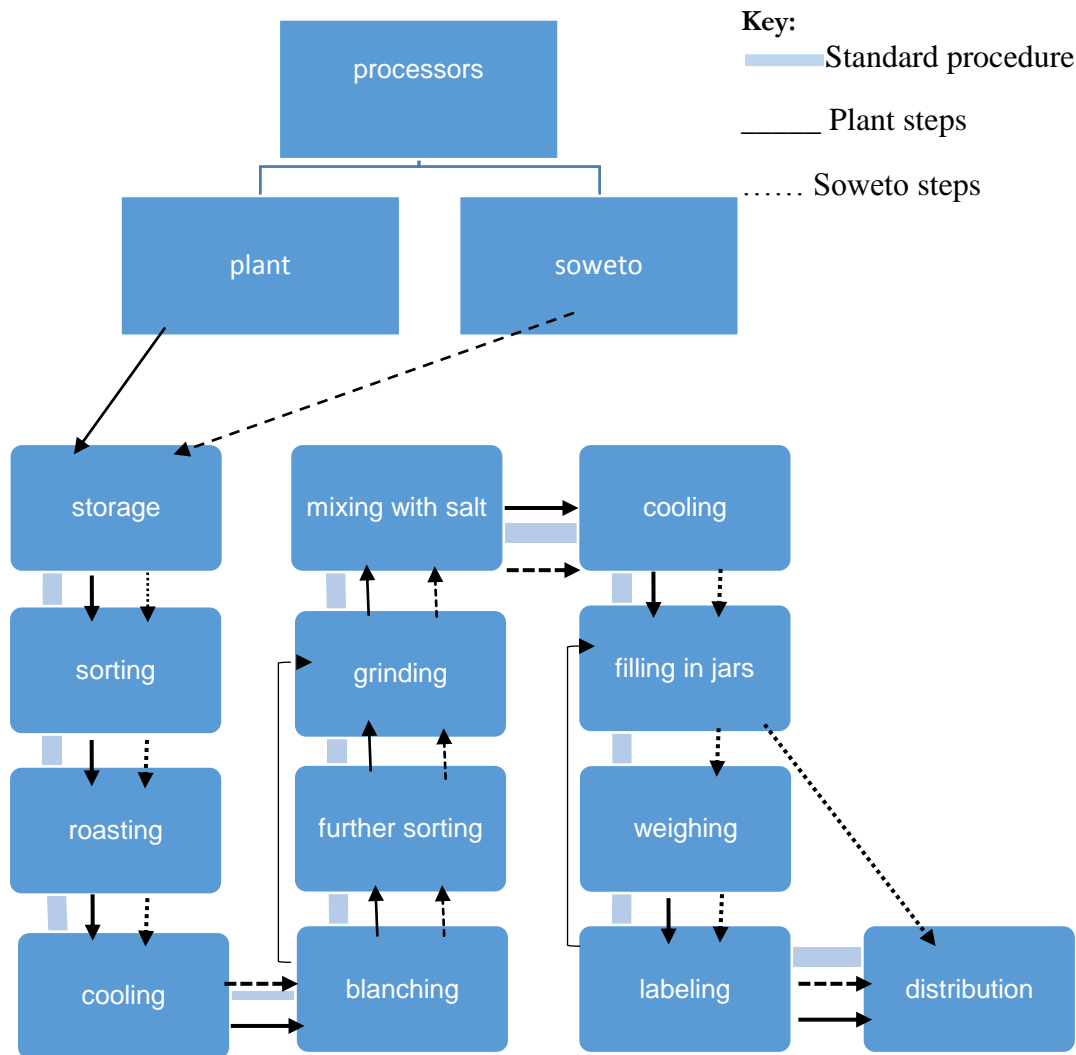


Figure 7: Flow Chart for Processing Peanut Butter at the Sub-Outlets

In response to the steps, the results were as shown in Figure 7; where one plant followed all the steps for processing from “storage to distribution.” The other plant began from “storage,” and then “sorting” of groundnuts before “roasting,” “Cooling” followed after roasting, and then “blanching” (removal of skin). There was no “further sorting” done and the next step was “grinding” groundnuts and “mixing it with salt.” After “mixing,” another step “cooling” was done in readiness for packaging or “filling in jars.” Again this plant skipped weighing of peanut butter and went on to “labelling” the canisters and finally, “distribution” to consumers and other outlets.

In Soweto, many respondents followed processing steps from “storage to filling in the jars” as indicated in Figure 7, while they skipped weighing and labelling, going

straight into distributing to consumers. Some skipped “further sorting” after blanching. Comparing peanut butter production between plant and Soweto market, it was concluded that there were differences in the steps. The similarities were shown at the beginning but later, dissimilarities were seen.

4.2.5.4 Inspection by Health Officers

Inspection is an important aspect of ensuring that standards are upheld especially in issues affecting Public Health. The findings in our study show that processors’ responses regarding being visited by the health team from both the processing plants and Soweto market were rare. Public Health officers never visited producers nor did they pass through to see how processors produced peanut butter. This was cemented by one respondent who made it clear by stating:

“No Public Health officers don’t come here. It is us who go to them especially

when we want to sell our products elsewhere because officers need to inspect the place where we make peanut butter to see if the place is good.” (Male Respondent 1 from Soweto)

Another respondent proposed the following:

“Environmental health workers must be going around checking and seeing that good processing of peanut butter is maintained in the outlets. Educate processors and sensitise them so they know how to produce reduced aflatoxin contaminated peanut butter.” (Male Respondent number 12 from Soweto).

Yet another respondent demanded inspection:

“Regular reviews of Aflatoxin standards by ZABS is required. Aflatoxin level is at 20ppb, in Zambia but outside they require 0 Aflatoxin in Europe, South Africa requires 5ppb. So we need this checked so that we can sell our peanut butter outside because kwacha has been unstable.” (Male Respondent number 15 Plant).

4.2.5.5 Distribution of Peanut Butter

Whether or not peanut butter is contaminated, processors must sell what they produce. This led to the researcher to enquire where processors distributed their products to. Market processors stated that they distributed theirs to passers-by and

people from compounds who wanted to consume it as well as for resell. The plants distributed their peanut butter to passers-by, retailers, commercial shops and to international markets. In comparing data from the two main outlets, both distributed to passers-by and retail shops but the plants had the liberty to sell internationally as well as to commercial shops. Another respondent revealed that his peanut butter was sold to the Chinese nationals:

“Some Chinese and other processors come to order for sell in their shops. Others bring their own peanuts to process for consumption.”
(Male Respondent number 11 of Soweto).

Yet another respondent added that:

“Some come to buy our peanut butter that we make and put labels on them to go and sell in their shops.” (Male Respondent number 8 from Soweto).

And another respondent mentioned:

“I sell to orphans. Three people come every two weeks to buy for them.” (Male Respondent number 2 Soweto)

One of the outlets testified selling the products to neighbouring countries and mentioned:

“We sell to all people. We sell to neighbouring countries such as Malawi, Zimbabwe, Namibia, Tanzania and DRC.” (Male Respondent number 15 Plant).

CHAPTER FIVE

DISCUSSION OF FINDINGS

The study sought to compare and investigate differences in aflatoxin contamination levels of peanut butter between local and international origin from selected outlets of Lusaka. The research also evaluated processor knowledge of aflatoxin contamination, health risks and good production skills that would help in lowering

aflatoxin levels in peanut butter among local processors. The results were presented in chapter four and in this chapter, a discussion of the findings is made. These outcomes were established in both quantitative and qualitative results section revealing that there was little or no knowledge of aflatoxin contamination and health risks among processors. Processing plant producers had better knowledge on the skills that would help reduce aflatoxin contamination in peanut butter compared to processors from the market. Triangulation of responses were included to consolidate the outcome of the whole research work.

5.1 Aflatoxin Contamination in General

Study results revealed that there was no significant evidence to conclude that there was a difference in the proportions of aflatoxin contamination levels between local and international origin peanut butter. Local peanut butter samples, however, showed higher aflatoxin levels compared with that of the international samples as indicated in the results section. These findings are comparable to those done in Malawi by Matumba et al. (2014), where local and imported peanut butter products were compared. It was discovered in that study that locally processed peanut butter had significantly higher aflatoxin contamination levels than the imported type. This was true with our findings when we use 15ppb. The median for imported products was 2.7µg/kg and maximum 4.3µg/kg. Locally processed peanut butter revealed results that ranged from 34.2µg/kg to 115.6µg/kg and were labelled unhealthy for human consumption globally as recorded in FAO, 2004 (Matumba et al. 2014). This result differs from the current study which found 30 local samples safe for public consumption and 25 were unhealthy for human consumption

Schwartzbord and Brown (2015) in the Haiti recorded 32 peanut butter samples presenting advanced total aflatoxin contamination of median 137µg/kg and maximum of 2720µg/kg, more than that found in Lusaka with the median of 6.95ppb and maximum of 147.2ppb. In Pakistan, 198 peanut products were commercially obtained from major cities of Punjab Iqbal et al. (2013). Analysis of aflatoxin contamination revealed that 16 (50%) tested peanut butter samples had mean concentration of 2.4 µg/kg. In Lusaka's selected outlets study, 100 (91.8%) samples were contaminated with high aflatoxin levels of overall mean value 18.8638 when

European standard was used. The local peanut butter mean was 31.6227ppb while that of the international was 5.8685ppb.

In India, a study to find out aflatoxin levels in common foods by Koirala et al. (2005) found that one-third of the samples had high levels of aflatoxin of above 30ppb with the highest values detected from peanut butter (42.5%). The information continues to tell the information that aflatoxin contamination is a real problem in many parts of the world, hence the need for more research and sharing of information with the stakeholders to address it.

Contrasting with peanut butter from Soweto market and processing plants where very high aflatoxin contamination levels than that from commercial stores were recorded, Mupunga et al. (2014) in Zimbabwe found very high aflatoxin levels in commercial peanut butter samples than those from the markets.

5.2 Aflatoxin Contamination in Relation to Set Standards

The findings were that using the 0 to 4ppb European standard, 9 out of 109 total samples, regardless of origin were safe for human consumption. The range included samples from 1.75ppb to 3.95ppb. The second category using CAC of 15ppb included the 9 (8.3%) samples from the European standard, totalling 83 (76.1%) samples as safe for public consumption. The remainder, 26 (23.9%) samples were unsafe for human consumption. These results are comparable to those done by Mutegi et al. (2013) where 71.8% of the peanut butter products satisfied both the European and Kenyan Bureau of standard (KBS).

The study results were further comparable to the findings reported by Njoroge et al. (2016) from the survey undertaken in the years (2012, 2013 and 2014) on regular monitoring of peanut butter survey in Zambia. Njoroge recorded extremely higher aflatoxin contamination values in peanut butter of upto 130 μ g/kg, 1000 μ g/kg and 10,740 μ g/kg. Aflatoxin contamination among the eight brands they tested were 73%, ranging upto 130 μ g/kg, 80% ranging up to 10,740 μ g/kg and 53% ranging up to 1000 μ g/kg respectively. Our study recorded contamination of up to 147.5ppb which is higher than 130 μ g/kg recorded in the first year of the survey. Using 15ppb approved (CAC, 2014) to judge how the public is exposed to aflatoxin contamination in peanut butter, 53 (48.9%) international samples were safe for consumption compared to only 30 (27.5%) from the local products. The local peanut butter had highest

aflatoxin contamination levels compared to the international peanut butter. This contrasted with Njoronge's who recorded that South African peanut butter had the highest aflatoxin contamination than that of the local products.

In Taiwan, Chen et al. (2013) detected aflatoxin contamination in 32.7% peanut products where levels ranged from 0.2 to 513.4 mg/kg. The highest contamination among the products was peanut butter of 142 samples, with a mean value of 52.8% exceeding 15 mg/kg Taiwanese regulatory limit. In our study, samples that exceeded 15ppb were 26 (23.9%). These findings imply that aflatoxin is a problem and addressing it by finding ways of reducing it might be the way to go in all these outlets to protect the population. Much good would come about which might include improved health, good profit realization by processors and well informed processors being established in the outlets when aflatoxin levels in peanut butter is reduced.

5.2.1 Aflatoxin Contamination Knowledge

The findings on the understanding of aflatoxin contamination among Soweto market and plant processors revealed that respondents did not know what aflatoxin contamination was. The variation in responses to what aflatoxin was informed the study that there was little or no knowledge of aflatoxin contamination in the sub-outlets. Further, processors' general classification of good groundnuts for use in processing peanut butter revealed in the result section leaves much to be desired. For instance, groundnuts of good looks may be deceiving because some of such kinds have been found to be of poor quality when closely examined. They do carry high levels of aflatoxin contamination levels and use of such groundnuts for processing peanut butter may be unsafe to the consumer which later may translate into customer's poor health. Another signal was given when processors stated that they used broken groundnut seeds to process peanut butter as recorded on page 38 in the results section. This result explains why aflatoxin contamination was high in most of the local peanut butter products. On the contrary, the results are dissimilar to the study by Azaman et al. (2016) who showed that participants had adequate knowledge on aflatoxin contamination.

Naturally, business people do not want to make a loss in doing business. Similarly in this study processors did not want to suffer loss by sorting out broken groundnut seeds and so they used the eye to judge good seed to process peanut butter. One

respondent in the current study indicated the need to sell peanut butter outside Zambia so he can make profit since the currency was low compared to dollar. The findings are similar to West African study by N'dede et al. (2012) in Benin who examined the financial risk associated with sorting and storing of peanut along the marketing chain. It was found that even though the costs of sorting and storing peanut could aid in reducing aflatoxin levels in peanut products, the practice posed financial risk to peanut traders. This implied that the quantity of peanut is reduced and less money is paid to the processors. This might explain again why peanut butter in the current study had increased aflatoxin contamination despite processors stating that they sorted out their groundnuts before processing. Such unfit groundnuts would be added to rather maximize profit in processed products because they were seen as having no problems. Even thin ground nuts could have been added to the seeds destined for peanut butter processing as noted by other researchers that we do not see what actually happened during processing. In our results, thin groundnuts were used for others who can eat them, as stated by one of the participants. This profit making revelation may have gratified processors with cash received while retarding aflatoxin level reduction efforts in peanut butter. There is cause to look at how this problem can be mitigated.

Literature has shown that not sorting out broken groundnut seeds lead to increasing aflatoxin contamination levels. A study in Greece by Georgioudou et al. (2012) analysed aflatoxin in nuts and discovered that damaged nuts contained 60% more aflatoxin in them than normal. Therefore, if processors could sort broken groundnuts seeds before processing their peanut butter, aflatoxin contamination may be greatly reduced thereby improving the aflatoxin contamination levels in peanut butter on Lusaka's urban outlets. On one hand, removing bad groundnuts among good ones lead to having more aflatoxin contaminated products on the market that will supply local poor people with contaminated products. With this contaminated product, the health of consumers is at stake. Indeed, such kind of groundnuts are bound to be sold at lower prices which the poor can easily access due to their economic constraints as some authors have noted. On the other hand, such unfit groundnuts end up being used in peanut butter processing still putting the consumers at great health risk through consumption of contaminated product.

Knowledge that broken groundnut seeds and just good to the eye looking groundnuts can increase contamination is necessary for Lusaka's producers and consumers to attain. If processors' understand the importance of sorting and what to sort from groundnuts before processing, through sensitisation and campaigns, reduction of aflatoxin levels in peanut butter can be possible. Indeed, Mutegi et al. also quoted Park, 2002 who stated that sorting out physically damaged and infected grains with strange shapes and sizes could result in 40-80% reduction of aflatoxin levels. This is a very good reduction percentage to fight for in both Soweto market and plant outlets but even so internationally. Again parallel to these study findings, Schwatzbord and Brown (2015) cited that although peanut processors removed kernels with visible mould, sorting was often uncommon before peanut butter processing in Haiti where contamination in 94% samples was detected.

5.2.2 Health Risk Knowledge

Regarding aflatoxin health risks to humans due to consumption of contaminated peanut butter, both respondents from the local sub-outlets expressed their ignorance of such. This was evidenced by responses given by participants as some of them stated that they did not know what health risks there were. A mention by one of the processors in the results section on page 38 and 39 that he did not know what health risks aflatoxin contaminated peanut butter would bring about but was certain that it caused diseases, is an example. These results were similar to a study in Ghana by Jolly et al. (2009) who examined quality assurance institutions and farmers' practice and perception on health effects awareness of aflatoxin in the peanut value chain among the public. The conclusion was that there was low awareness of health effects among them. Again our results are similar to Wild & Gong, (2010) who found comparable results that in regions where there is greater contamination, mycotoxin knowledge and understanding of adverse health effects was lacking and the known risks were poorly communicated to governments. In Zambia, aflatoxin knowledge is not a subject that most people know about and hence the need to improve communication of such important information to the public and stakeholders at large. This move can begin with investigator's passion for research accumulation of evidence enough to convince other interested parties of the need for knowledge on aflatoxin subject and health risks to enlighten the population. Consumption of high aflatoxin contaminated peanut products, like Liu and Wu in their review (2010)

observed, bring about consequences such as liver and kidney cancers and other ailments. Liu and Wu in 2010, further stated that although it is impossible to completely eliminate aflatoxin in food worldwide, it is possible to significantly reduce the levels and so reduce liver cancer incidence worldwide. The Democratic Republic of Congo is amongst African countries listed with high prevalence of liver cancer as cited in Kamika and Takoy, (2011). These health issues might be prevailing in Zambia where little knowledge regarding contamination and health risks in the sub-outlets has been revealed. The population is unaware due to lack of research to expose such information. Congo for example, is Zambia's neighbor and there is need for quick precautions in the direction of increasing awareness through the media and whatever means would be necessary to take.

5.2.3 Safe Peanut Butter Production Skills

In our study, both plants and Soweto market respondents stated how they try to reduce aflatoxin levels in peanut butter for consumer safety. Sorting of groundnuts before processing peanut butter was one of the ways as already mentioned above. On page 40, a flow chart for peanut butter processing was given with various steps for use in processing peanut butter. Some steps were omitted and others were considered by the processors in local sub-outlet. Omitted steps on further sorting of groundnuts could have been crucial and critical in trying to reduce aflatoxin in peanut butter. This omission may have contributed to high accumulation of aflatoxin contamination in peanut butter as given in the results.

Desire to maximise profit by processors could have been the reason causing some steps to be omitted in processing. The steps given by the processors are close to the example given in the GMP, (2009) document which included purchasing of shelled peanut, receiving, storing, cleaning, cooling, grinding, adding ingredients, grinding, DE aeration, cooling, filling in jars, packaging and labelling. There is a branch to purchasing jars and cleaning them. All together, the steps add between 13 and 15 whereas in our study processors mentioned 12 steps.

5.2.3.1 Cleanliness of the Machine

Observance of hygiene is vital and must be considered and practiced if processors are to reduce aflatoxin contamination in peanut butter and provide safe peanut butter to consumers. Processors stated that they clean the machine by wiping and not all the

time but rarely due to busy schedules. They cleaned by wiping as the machines were electrical and use of water to clean would cause damage. Variation in responses between the plant and market respondents in cleaning and how often this was done led to a conclusion that no proper procedure was followed. Thus, not observing hygiene regularly could have contributed to high aflatoxin contamination in peanut butter samples.

Further observation on cleanliness was made and some retailers in Soweto market appeared quite unkempt at their stands. All respondents had no dress code to help in their appearance and to attract customers. Smart appearance of processors would translate into them desiring to produce quality peanut butter to match their standard of cleanliness. The gesture could help in reducing aflatoxin contamination in peanut butter but also in attracting customers to buy their product. Accumulation of sand in peanut butter, which most consumers have been experiencing and complaining about upon chewing food spread with peanut butter could be associated with failure to keep up with personal and machine hygiene among processors in these outlets. Our findings are similar to Ndung'u et al. (2013) who found in their study that most practices by processors such as failure to clean the grinder after peanut butter production, storing roasted peanut longer before grinding and cooling roasted nuts on the floor, in the open could have contributed to increased aflatoxin contamination in peanut butter. Processors who do not observe hygiene when producing peanut butter contribute to creation of high aflatoxin levels in the product. To curb the situation, there is need to impart knowledge in the processors regarding aflatoxin contamination and health risk effects to humans associated with peanut butter processing. Azaman et al. (2016), respondents had favourable attitude and high hygiene practices towards aflatoxin reduction efforts. Such safe practices if acquired and applied, might aid Lusaka processors in that direction.

Another study in Coted'voire, by Boli et al. (2013) established that Good Manufacturing Practices and good hygiene could help reduce contamination in processed products. In addition, if proper hygiene practices and personal sanitation are not applied, Azaman et al. (2016) reported, public health crisis can result from aflatoxin contamination. In the same study, hygiene practices between small-scale manufacturers and retailers showed no differences and citing Walker, Pritchard and

Forsythe (2003) and Bas, Ersun and Kıvanç (2006); who reported that small and medium sized businesses had knowledge deficiency on personal hygiene and food safety due to time and expert constraints. In our study, lack of time was one of the reasons for not cleaning the processing machine regularly.

5.2.3.2 Inspection by Officers

Learning from the results section, whether health officers visited the outlets to inspect the processing of peanut butter attracted variations in answers from processors. Some of the producers stated that officers did not visit them while some said officers did random visits and still others even put times when they were visited. This variation could possibly mean that respondents feared that investigators would report them to the public health officers' higher authorities who would in turn come and close their businesses if they discovered the truth. Over all, the conclusion was that they did not visit the outlets in the way processors expected. One of the respondents thought if ZABS could be reviewing aflatoxin standards regularly to check levels in peanut butter, his products could be sold outside the nation to make profit since there is uncertainty of the currency's stability in the nation. The need for profit making by processors and willingness to do whatever it may take for the product's safety is a good opportunity to begin to increase awareness of aflatoxin harm to humans among stakeholders. Another respondent in our study suggested that processors be educated and sensitised on how they can reduce contamination in peanut butter. Again this shows that authorities need to check and ensure aflatoxin levels are in reduced measures and to act on the suggestions by respondents in order to achieving this goal. Our results are similar to Boli et al. (2013) who mentioned in their study that, good manufacturing processing and good hygiene practices would help to minimize fungal contamination in to obtain safe peanut butters. This can be done better if inspection and demanding adherence to the practice by responsible people is observed.

5.2.3.3 Distribution of Peanut Butter

Regarding peanut butter distribution, it was interesting to note that they sold their peanut butter to other retailers in Lusaka and to some customers from the compounds and to orphans who are vulnerable children. This information tells us that contaminated local peanut butter is distributed not only in Lusaka but also to other

shops open to the public including the vulnerable children. Within the country, this peanut butter is bound to go to all provinces of Zambia as business men and women travel to Lusaka to purchase orders for their outlets in the rural areas. Lusaka is a central place for business and almost all things concerning business begin from Lusaka. Business people buy peanut butter from Soweto market and put labels on those canisters, with no shelf life attached, as revealed in our qualitative results. Sales of such peanut butter are done in shops as if the distributors in those shops manufactured the product. Soweto market is a loophole since things are much cheaper there and so products easily penetrate through to borders by passers-by, cyclist, bus passengers and even those who travel by air.

This finding reveals how far and wide peanut butter that is processed in Lusaka urban outlets can go. Truly, some of the peanut butter finds its way out of the country to other nations through people or business men and women who come to visit Zambia. There is a danger of exposing the wider population to health risk of aflatoxin contamination through this means.

With high aflatoxin levels in peanut butter, the product cannot easily sell outside the nation and so will end up being bought and consumed by locals and neighbouring countries who may not question the standard of the products that they purchase. Certainly, consumers have a challenge in that they are not able to see what is contained in peanut butter by merely looking at the canister. Often times, peanut butter appears very good and attractive to the eye yet it is contaminated. Since Lusaka, Zambia, does not have a guide to check how much aflatoxin level in peanut butter is safe for the public to consume, “imports” and even “exports,” come in and go out of the nation to the detriment of the population’s health. Without proper guidance, our local products cannot get to compete with the outside market where better reward can be realized by those whose products meet certain requirements for sale.

This outcome is comparable to a review carried out by Bui-Klimke, et al. (2014) which looked at Nut–Pasticcio trade where, due to increased number of countries with regulations on allowable mycotoxin levels in imported foods; an interest to find out whether there was an association between regulation and trade that existed was created. It was revealed in this review that nations tended to trade with other nations

that had identical aflatoxin standards. Countries without regulations were importing highly contaminated products from other countries with unrestricted laws or no regulations. The findings are similar to the current study report conclusion since responses revealed that peanut butter from their outlet was sold to other nations. Due to the lack of monitoring of peanut butter that comes in by the receiving nation and how peanut butter goes out through the distributing processor or company, there is a Public Health threat. Such revelations could be true with peanut butter processed in Lusaka, which is bound to reach many a population in and outside the nation, exposing them to contaminated peanut butter and its health risks.

Regarding aflatoxin contamination and health risks in Lusaka's outlets, further exploration is required to protect this population and also that of the neighbouring countries. It has been said elsewhere in public health that prevention, from health risks, is better than curing a disease. In addition to training and sensitising of processors, a provision of a mechanism for accepting good and rejecting bad peanut butter on the market would be a starting point of addressing this problem.

5.4 Limitations

The results could not be generalised to all of Lusaka, but it was limited to selected outlets in Lusaka urban district in the province. Future studies may require taking into consideration more processing plants not included in this study to see variations in responses and the experience and attitudes of processors before and during data collection. Another observation was that aflatoxin was a topic that few people knew about, several visits were made to try and convince the processors to provide their products for testing and to be interviewed. This experience inherently affirms the inadequate knowledge about the implications of aflatoxin contamination among processors.

CHAPTER SIX

CONCLUSION AND RECOMMENDATIONS

6.1 Conclusion

The study compared aflatoxin levels between local and international peanut butter totalling 109 samples from selected outlets of Lusaka urban city, Zambia. The study also explored knowledge on aflatoxin contamination, health risks and safe production skills to see if what is practiced would contribute to reducing aflatoxin contamination in peanut butter among processors. The rates of aflatoxin concentration levels sold in these outlets were not significantly different using the European set standard of 0 to 4ppb. Only nine (8.3%) sample units of which seven (6.4%) were from the international and two (1.8%) from local origin, were safe for consumption. Using 15ppb approved CAC, 83 (76.1%) of all products, with 53 (48.6%) from international and 30 (27.5%) from local peanut butter sample units satisfied the standard. The high rates of aflatoxin contamination found in peanut butter samples sold in these outlets implies that Lusaka's population is at risk of consuming contaminated peanut butter which presents a threat to the population wellbeing. Generally, processors seemed to have little or no knowledge of aflatoxin contamination and health risks to human beings through consuming contaminated peanut butter. Regarding safe production skills, processors had similar steps in the outlets but divergences were observed in some parts. Cleaning of the processing machines was poor among Soweto respondents compared to those of the processing plants. Inspection was rarely done.

The presence of high aflatoxin levels in peanut butter must be a concern for both the general public and government. Ways of reducing contamination need to be explored. This is possible through government intervention who must encourage research and enforce available regulations of standards to be followed. Processors and retailers can be held accountable by ensuring that they adhere to regulations and for those who fail to obey, punishment is to be exercised.

6.2 Recommendations

In promoting set standards for the benefit of the population, the public should be well informed about the health risks that aflatoxin contamination in peanut butter can bring to humans. Emphasis on compulsory sorting or removing of broken seeds,

good to eye and very thin groundnut seeds before processing peanut butter should be encouraged to reduce aflatoxin contamination. It is important that the Zambia Bureau of Standards work towards toughening inspection, monitoring and ensuring compliance to available means of standards for quality maintenance of the products. Even though based on CAC standards, aflatoxin levels from international peanut butter were lower than the locally produced, inspection must still be carried out to ensure products on the markets are safe for public consumption.

Sensitising the local population through the media and training of processors on the health risk of consuming aflatoxin contaminated peanut butter is recommended. Health officers need to make inspection visits to processing plants and retail markets to check and give guidance on safe processing of peanut butter. Practices that contribute to reducing aflatoxin contamination in peanut butter such as hygiene observance through use of dress code and cleaning of the processing machine should be encouraged.

Areas of future investigations should focus on ensuring that imported and other locally produced peanut butter that have not been captured in this research be also studied. It is also recommended that researchers be provided with opportunities to conduct collaborative research with international processors to keep abreast with current trends and works on aflatoxin. This would contribute to upholding acceptable standards and practices for reducing aflatoxin contamination in peanut butter processing. Teamwork must be considered seriously, not only among peanut butter processors in the outlets but also among all involved in the groundnut production string; from seed planter, to harvester, to the one who stores and the processor of peanut butter to ensure reduction in aflatoxin contamination. There is need to improve the quality of peanut butter and increase awareness on the dangers of aflatoxin contaminated products in the outlets in order to protect the public.

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APPENDICES

APPENDIX 1: Information Sheet and Consent Form

The University of Zambia
School Public Health
Department of Population Studies

Information Sheet and Consent for Peanut Butter Processors and Retailers in Lusaka

TITLE: Investigating Aflatoxin Contamination and Knowledge levels in producing safe Peanut Butter among selected Lusaka Urban Processors.

SECTION A: Information Sheet

This information sheet is for processors and retailers in peanut butter that are going to be invited to participate in the study, “**Investigating Aflatoxin Contamination and Knowledge levels in producing safe Peanut Butter among selected Lusaka Urban Processors.**” Aflatoxin is a chemical produced by fungi in peanuts. It is mould seen as dark, brown, white, grey coloured peanuts and in its high levels of product consumption; it causes ill health to consumers.

Introduction

To Participant,

My names are Maureen Samson Banda, a student at the University of Zambia undertaking a master’s degree in public health. I am conducting a study in Lusaka urban to determine and compare Aflatoxin levels between peanut butter produced in Lusaka (Zambia) and that coming from outside Zambia. You are invited to take part in this study because you can help with the information that is needed. If there is anything you do not understand please feel free to ask as we go through the information sheet and I will explain it to you.

The Purpose of the Study

- The main purpose of this study is to investigate and compare Aflatoxin levels in local and international peanut butter in relation to acceptable international set standards.
- The study would also like to assess the level of Aflatoxin contamination knowledge and production of safe peanut butter among local processors and retailers in selected Lusaka Urban outlets.

Participant Selection

You are being invited to take part in the study because you have been processing and retailing peanut butter over a period of time and you know a lot concerning Aflatoxin that can help in answering the questions.

Voluntary Participation

Your participation in this study is voluntary. You are free to choose to take part or stop taking part at any time without any punishment.

Duration

The interview will be given only once and will take about 15 or 20 minutes.

Risks

Risks in this study are somewhat minimal. Nonetheless, participant may be affected psychologically as their business could get associated with issues of Aflatoxin. Nevertheless, you are free not to take part in this study.

Benefits

Even though the findings of this study may not benefit you right away, it will help regulatory agencies such as Public Health Authorities and Ministry of Health in decision making with regards to public health policy on Aflatoxin harm as it relates to public exposure. The study findings will be vital for public safety and hence the need for it to be done.

Confidentiality

The information that you provide will not be exposed and confidentiality will be observed. Anything you say will be confidential and it will not be associated personally to you in any reports after this study. You are included in this study because you may have knowledge on the possible health effects of aflatoxin in PB that may create on consumers. Information given will be used to prepare reports after analysis, without including any specific names.

Findings from the Study

The information that will be collected will be used to find solutions to the problem of high Aflatoxin levels in peanut butter. Further, information will quicken public health workers to check for acceptable Aflatoxin levels in peanut butter that the public is consuming from Lusaka outlets.

Sharing of Results

The information collected will not be shared with or given to anyone except among the research team and University of Zambia. The information will also be shared with you on conclusion of the study by the investigator. Information will also be shared with the Ministry of Health, School of Agriculture laboratory and Zambia Bureau of standards.

Right to Refuse or Withdraw

You have the right to refuse to participate or to withdraw from the study at any time.

SECTION B: Consent Form

The purpose of this study has been adequately explained to me, and I understand the aim, risks, benefits and confidentiality of this study. I further understand that if I agree to participate, in this study, I can withdraw at any time without having to give an explanation and that taking part in this study is purely voluntary.

If you are willing to participate in the study, please do so by signing below.

I (Name) agree to participate in this study.

Participant signature/ thumb print.....Date.....

Consent provider sign..... Date.....

If you need any further clarifications even after our discussions, you can contact the Principle Investigator or the chairperson of the ethics committee on the following addresses:

WHO TO CONTACT IN CASE OF QUESTIONS:

Maureen Samson Banda
UNZA School Public Health
Department of Population Studies

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Lusaka.
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You may also contact:
The Chairperson Biomedical Ethics Committee
Telephone: +260211256067
E-mail: unzarec@zamtel.zm

APPENDIX 2: Nyanja Translation of the Information Sheet and Consent Form

The University of Zambia
School of Public Health
Department of Population Studies

CIPEPALA ca Nkhani ca Opanga ndi Ogulitsa Pinati Bata m'malo Osankhidwa
mu Lusaka

CHAPAMUTU: Kufuna Kuwona Muyeso wa Cuku mu Pinati Bata Mogwirizana
ndi kutetezedwa kwake m'malo osankhidwa ogulitsilamo mu
Lusaka.

Gawo A: Pepala ya Nkhani Zounikila

Cipepala ca nkhani ici ndi ca opanga ndi ogulitsa pinati bata amene azaitanidwa kutengako mbali mu maphunziro a “Kufuna kuwona muyeso wa cuku ndi kutetezedwa kwa pinati bata m'malo osankhidwa kupanga ndi kugulitsilamo a mu Lusaka.”

Ciyambi

Kwa otengako mbali,

Maureen Samson Banda ndiyo maina anga, wa mu sukulu la maphunziro akuya pa University of Zambia mugawo la za umoyo wa anthu ambiri. Ndicita maphunziro mu Lusaka akufuna kupeza komanso kusiyantsa ndi kulinganiza kwake kwa muyeso wa cuku mu pinati bata yopangidwa mu Lusaka ndi pinati bata yocokela kunja kwa dziko. Muitanidwa kutengako mbali mu maphunziro amenewa cifukwa mukhoza kuthandiza pa nkhani yofunikila iyi. Ngati pali ciliconse cimene inu simumvetsa, conde khalani omasuka kufunsa pamene tipita mu cipepala ca nkhani ici ndipo ndizayesa kumasulila.

Cilingo cha Maphunziro awa Ndicho:

- i. Kufuna kupeza komanso kulinganiza ndi kusiyantsa muyesowa cuku copezeka mu pinati bata yopangidwa mu Lusaka ndi pinati bata yocokela kunja kwa dziko kulingana ndi malamulo oikidwa obvomelezedwa a maiko ambiri.

- ii. Kuwona pa muyeso wakudziwa kuipa kwa cuku ndi mapangidwe a pinati bata yotetezedwa pakati pa opanga ndi ogulitsa pinati bata a m'malo osankhidwa mu Lusaka.

Kusankha kwa Otengako Mbali

Muitanidwa kutengako mbali mu maphunziro amenewa cifukwa cakuti mwakhala opanga ndi kugulitsa pinati bata mwa kanthawi ndipo mudziwa zambiri zimene zingathandize kuyankha mafunso pa nkhani yofunikila pa zinthu zokhuzana ndi cuku mu pinati bata.

Kuzipeleka pakutengako Mbali

Kutengako mbali m'maphunziro awa ndikozipeleka. Ndinu omasuka kuleka kutengako mbali kopanda kulandila cilangociliconse.

Nthawi

Mafunso azapatsidwa kamodzi cabe ndipo azangotenga mphindi monga 15 kapena 20 cabe.

Ziopsyezo

Ziopsyezo potengako mbali m'maphunziro awa ndizazing'ono kwambiri. Otengako mbali akhoza kubvutidwa m'maganizo cifukwa bizinesi yao ikhoza kukhudzidwa mwakupezeka ndi cuku cambiri mu pinati bata. Otengako mbali akhoza kuganiza kuti mwina zotulukamo za cuku zikhoza kukhala zoipa. Ndinu omasuka kusatengako mbali m'maphunziro amenewa.

Zothandiza

Angakhale zili tero, zotulukamo za m'maphunziro aye ndizofunikila kuteteza anthu akudya pinati bata kuti akhale ndi umoyo wabwino. Kulibe malipilo aliwonse kwa inu pakutengako mbali mu maphunziro aya. Angakhale kuti zotulukamo m'maphunziro aya sizizakuthandizani inu kwatsopano apa, zimenezi zizathandiza aulamuliro monga aza umoyo wa anthu ambiri ndi a boma (Ministry of Health) kuti apange ciganizo pa za umoyo pakupanga mapulogramu ya kucepetsa cuku mu pinati bata. Maphunziro ndiyofunikila kucitika cifukwa cuku cibweretsa mabvuto pathupi la anthu akudya pinati bata imene ili ndi cuku copitilila muyeso.

Cisinsi

Nkhani zimene muzapatsa sizizaululika kwa aliyense ndipo cisinsi ceni-ceni cizasungidwa kwathunthu. Zilizonse zimene inu muzakamba zizakhala za cisinsi ndipo sizizaphatikizidwa kwa inu mu ma lipoti titatha maphunziro awa. Inu mwasankhidwa kutengako mbali m'maphunziro amenewa cifukwa cakuti mwina mukhoza kudziwa za kuculuka kwa cuku mu pinati bata ndi kukhudzidwa kwacha kwa za umoyo wa iwo akudya.

Zopezeka mu Maphunziro aya ziza:

- Thandiza akulu a zaumoyo wa anthu kufuna kudziwa muyeso wa cuku woyenela mu pinati bata imene anthu akudya mu Lusaka.
- Zizaunikila opanga pinati bata pa njira zabwino zopangilamo pinati bata.

Kugawana Zotuluka m'maphunziro

Nkhani zotuluka m'maphunziro sizizagawanitsidwa ndi munthu wina aliyense kucoselako akulu ocita maphunziro ndi a ku University of Zambia. Nkhanizi zizapatsidwanso kwa inu kupyolela mwa oyang'anila pa maphunziro amenewa titatha kupeza mayankho m'maphunziro amenewa.

Danga Lokana kapena kuleka Kutengako mbali

Otengako mbali ndiomasuka kufunsa mafunso aliwonse ndipo ngati awona kuti sakhutira, ali ndi danga loleka kutengako mbali mu maphunziro nthawi iliyonse.

GAWO B: Cipepala ca Cibvomekezo

Cilingo ca maphunziro aya camasulidwa kwa ine, ndipo ndamvetsa za colinga, ziopsyezo, thandizo ndi cisinsi ca maphunziro amenewa. Mopitiliza, ndamvetsanso kuti ngati ndibvomela kutengako mbali m'maphunziro amenewa, ndikhoza kuleka panthawi iliyonse kosapeleka lonjezo ndikuti kutengako mbali m'maphunziro aya, ndikozipeleka kwathunthu.

Ngati mwazipeleka kutengako mbali mumaphunziro aya, Conde citani cimenezi pakusaina cipepala ici pansipa.

Ine (Dzina) ndibvomela kutengako mbali m'maphunziro aya.

Wotengako mbali kusaina /Kudinda cala (Tsiku).....

Wopatsa cibvomelezo kusaina (Tsiku).....

Ngati mufuna zomasulila zilizonse ngakhale titatha zokambilana zathu, mukhoza kuwona mkulu wamaphunziro kapena a cheya a zocititsa maphuziro a mtundu umenewu pansipa.

Ngati muli ndi mafunso kapena bvuto iliyonse pa maphunziro amenewa- Onani:

OWONA NGATI PALI MAFUNSO

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Mukhozanso kuwona

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**APPENDIX 3: Semi-Structured Questionnaire Guide for Processors and Retailers in
Lusaka Urban District**

Questionnaire number.....

Interview Date.....
Day Month Year

Interviewer initial

SECTION A. Participant Identification and Demographic

No.	Please answer these questions	Response
1	Outlet name 1) Soweto (1-12) Specify 1,2,3,4,5,6,7,8,9,10,11,12 [] 2) Plant JB [] 3) Plant FP [] 4) Plant SAZ []	Official use <input type="text"/>
2	Respondent title: 1) Manager/Director [] 2) Assistant [] 3) Other specify.....	<input type="text"/>
3	Sex: 1) Male [] 2) Female []	<input type="text"/>
4	When were you born? <input type="text"/> <input type="text"/> <input type="text"/> Day Month Year	<input type="text"/>
5	What is your highest level of education? 1) No education [] 2) Primary [] 3) Secondary [] 4) Tertiary []	<input type="text"/> <input type="text"/>

SECTION B. Aflatoxin Health Risk, Processor Production Skills, Inspection and Regulation in Zambia

Aflatoxin is something that is produced by fungi. It is present in peanuts and other crops. Peanuts that looks mouldy, shrivelled, manifested with insects, contain high levels of Aflatoxin. Frequent consumption of such products has been associated with ill health and eventual death. Now I am going to ask you about Aflatoxin and its health issues. Please feel free to answer these questions. Your name will not appear in the results. The information that you give is meant to help improve peanut butter processing in Lusaka.

Q6. What do you understand by the word Aflatoxin? List of probe responses:

Aflatoxin is:	
1	A disease
2	Poison
3	Medicine
4	Do not know

Q7. Are you aware that high Aflatoxin can cause health risk to humans if consumed?

- 1) Yes [] go to Q7b
 2) No [] go to Q8

Q7b. What health risk can be caused by Aflatoxin? List of responses and probes:

Aflatoxin causes:	
1	Stunted growth
2	Liver cancer
3	Immunity suppression
4	Aflatoxicosis
5	Do not know

Q8a. Are you aware of what currently Zambia uses as acceptable Aflatoxin limit for peanut butter?

- 1) Yes [] go to Q8b
 2) No [] go to Q9

Q8b. What is Zambia using for Aflatoxin acceptable peanut butter limit? Probe with:

Zambia's acceptable standard	
1	15µg/kg
2	1000µg/kg
3	2500µg/kg
4	Do not know

Q8c. Other

Q9. How do you know that the groundnuts you are using to make Peanut butter has less aflatoxin?

Q10. Do public health officers come to inspect your peanut butter processing?

1) Yes [] go to Q10b

2) No [] go to Q11

Q10b. How often do public health officers come to inspect your peanut butter processing?
.....

Q11. There are some helpful requirements which processors can use to produce quality peanut butter. Please indicate your opinion how much you agree or disagree with the requirement as helping. Probe list

1 = Strongly disagree, 2 = Disagree, 3= Neutral, 4= Agree or 5= Strongly agree

Possible helpful activity	Strongly disagree 1	Disagree 2	Neutral 3	Agree 4	Strongly agree 5
Training of processors					
Sensitisation of processors					
Increase profit on PB sales					
Law enforcement					
Encourage Team work					
Use of Lab tests					
In-process quality control					

Q12a. As a processor, have you received any training in peanut butter processing?

1) Yes go to Q12b

2) No go to Q13

Q12b. Where did you train?

Training Source	
1	College
2	Workshop
3	Conference
4	By observing others do it

SECTION C. Peanut Butter Processing Skills

Now I am going to ask you questions regarding peanut butter processing that you may be familiar with. By 'processing', I mean step by step activities that you undertake before you come up with a finished packaged container to distribute.

Q13a. Tell me step by step what you do to produce your peanut butter from start to end when it is ready for sale? List of responses and probes

Processing steps		Response
1	Storage	
2	Sorting	
3	Roasting	
4	Cooling	
5	Blanching remove husks	
6	Further sorting	
7	Grinding	
8	Mixing salt/stabilizer	
9	Cooling	
10	Filling in sterilised jars	
11	Weighing	
12	Labelling	
13	Distributing to customers	

Q13b. Do you clean your peanut butter processing machine?

1) Yes go to Q13c

2) No go to Q14

Q13c. How often do you clean your peanut butter processing machine?

.....

Q14. Before processing your peanut butter, what do you sort out as bad in the groundnuts? Probe with:

Sorting:	
1	Broken kernels
2	Insect manifestation
3	Rotten peanut
4	High moisture peanut
5	Mixed coloured peanut

Q15. Based on your experience or what you have heard, what are the things that Show that this groundnut is good to use for peanut butter processing?

Q16. How much peanut butter do you produce per day?

Amount..... Date.....

Q17. Where do you sell your peanut butter?.....

Q18. Do you have any questions or additions from what we have discussed in this interview?

Thank you

APPENDIX 4: Nyanja Translation, Mafunso a Mapangidwe a Pinati Bata a Opanga ndi Ogulitsa m'malo a Lusaka District:

Nambala ya mafunso.....

Tsiku la kukambisana.....

Tsiku Mwezi

Caka

Dzina la ofunsa mafunso muufupi

Conde yankhani mafunso aya:	
1	<p>Dzina la ogawanitsa1) Malo oyamba Soweto (1-12) osankha1, 2, 3,4,5,6... []</p> <p>1) Malo oyamba SO []</p> <p>2) Malo aciwiri JB [] <input type="text"/></p> <p>3) Malo acitatu FP []</p> <p>4) Malo acinai SAZ []</p>
2	<p>Woyankha:1) Wamkulu wa nchito/Mtsogoleri []</p> <p>2)Wamkulu wa nchito wa ciwiri [] <input type="text"/></p> <p>3) Winawache</p>
3	<p>Munthu: 1) Mwamuna [] <input type="text"/></p> <p>2) Mkazi []</p>
4	<p>Zaka: Munabadwa liti? <input type="text"/> <input type="text"/> <input type="text"/></p> <p style="text-align: center;">Tsiku Mwezi Caka</p>
5	<p>Kodi sukulu munafika patali bwanji? <input type="text"/></p> <p>1) Sindinapite kusukulu []</p> <p>2) Pulaimale []</p> <p>3) Sekondale []</p> <p>4) Maphunzilo apamwamba []</p>

GAWO B: Cuku, umoyo, Kuyang'anira Opanga Pinati Bata ndi lamulo m'Zambia

Cuku ndi cinthu cina cimene cipangidwa ndi turombo cimene cimapezeka munshawa. Nshawa zimene ziwoneka zakuda, zamankwinya, zili ndi tudoyo

zimakhala ndi cuku copitilila malire. Kupezeka kawiri-kawiri akudya nshawa zoterezi kumabweretsa umoyo wa matenda pa thump la munthu ndipo pambuyo pake imfa.

Tsopano ndizakufunsani mafunso pa za cuku ndizina zaumoyo. Conde khalani omasuka kuyankha mafunso amenewa. Dzina lanu silizawoneka pa zotulukamo m'maphunziro aya. Nkhani zimene muzapatsa zizathandiza kukometsa mapangidwe a pinati bata mu Lusaka.

Q6. Kodi munvetsa ciyani pa liu lakuti 'cuku'? Conde nenani cimene muganiza.

Cuku ndi		Responses
1	Matenda	
2	Paizoni	
3	Mankhwala	
4	Sindidziwa	

Q7a. Kodi mudziwa kuti Cuku cikhoza kubweretsa bvuto pazaumoyo ngati cidyedwa mopitilila muyeso?

1) Inde [] pitani ku Q7b

2) Iyai [] pitani ku Q8

Q7b. Kodi ndizaumoyo zobvuta zotani pathupi zimene zibwera cifukwa ca cuku? Conde nenani zimene muganiza. Mndandanda wakufunsitsitsa.

Cuku cibweretsa	
1	Kusakula msinkhu bwino
2	Kansa ya mciwindi
3	Kucepa kwacitetezo ca matenda mthupi
4	Thupi kutentha kamba ka ciwindi codwalila
5	Sindidziwa

Q8a. Kodi mudziwa za muyesowa wa cuku cololedwa mu pinati bata cimene cigwiritsidwa nchito palipano mu Zambia?

1) Inde [] pitani ku Q8b

2) Iyai [] pitani ku Q9

Q8b. Kodi cuku cololedwa pa mapangidwe a pinati bata panthawi ino ndi mpimo uti? Kufunsitsitsa:

Q8c. Zina.....

Q9. Kodi mudziwa bwanji kuti nshawa zimene mugwiritsa nchito kupanga pinati bata zili ndi cuku cocepa?

Q10. Kodi akulu akulu a zaumoyo wa anthu a mboma amabwera kukawona m'mene mupangila pinati bata?

1) Inde [] pitani ku Q10b

2) Iyai [] pitani ku Q11

Q10b. Kodi amabwera kangati pa caka?.....

Q11. Pali zina zofunikila zimene zingathandize opanga pinati bata kuti apange pinati bata yabwino kwambiri. Muthebulo pansipa, pali mndandanda wa zina mwazinthuzo. Conde, onetsani ciganizo canu pakuchula zinthuzi:

1 = Kusabvomeleza kwambiri 2 = Kusabvomeleza 3 = Pakati-kati 4 = Kubvomeleza5
= Kubvomeleza kwambiri.

Zocitika zingathandize	Kusabvomelez a kwambiri 1	. Kusabvom eleza 2	Pakati- kati 3	Kubvo meleza 4	Kubvomelez a kwambiri. 5	
Kuphunzitsa wopanga pinati bata						
Kuunikila ogulitsa onse						
Kuikilako phindu pogulitsa pinati bata						
Kuika lamulo limodzi lopangilamo PB						
Kugwira nchito mogwirizana						
Kufufuza bwino mulabu yamayeso						
Kasamalidwe ka mapangidwe a PB						

Q12a. Monga opanga pinati Bata, kodi munalandilako maphunziro aliwonse kulingana ndi mapangidwe a pinati bata?

1) Inde [] pitani ku Q12b

2) Iyai [] pitani ku Q13

Q12b. Kodi manaphunzira kuti?

Kumene munaphunzira	
1	Kukoleji
2	Kuwekishopo
3	Kukonfalensi
4	Pakuwonelela anzanga opanga

GAWO C: Mapangidwe a Pinati Bata

Tsopano ndizakufunsani mafunso pa zimene muziwa popanga pinati bata zimene mungakumbukile bwino. Pa liu lakuti ‘kupanga’ nditanthauza zimene mumacita kuti mupange pinati bata.

Q13. Ndiuzeni mwa tsatane-tsatane zimene mucita pakupanga pinati bata kucokera poyamba kufikila pothela pamene mulonga mucoikamo kukagulitsa. Mndandanda wakufunsitsitsa.

Zocita		Yankho
1	Kosungila	
2	Kusankha	
3	Kukanzinga	
4	Kuzizilitsa	
5	Kucotsa mamba	
6	Kusankhanso	
7	Kugaya	
8	Kuika mcere/sauti	
9	Kuziziritsanso	
10	Kulongeza moika	
11	Kupima kulemela	
12	Kuika malemba	
13	Kupeleka kwaogula	

Q13b. Kodi mumatsuka mashini yopangila pinati bata yanu?

1) Inde [] pitani ku Q13c

2) Iyai [] pitani ku Q14

Q13c. Mumatsuka kangati pa sabata?.....

Q14. Musanapange pinati bata yanu kodi mumasankha nshawa zanu? Conde nenani mtundu wa nshawa zimene musankha zoipa. Mndandanda wakufunsitsitsa.

Nshawa zoipa	
1	Nshawa zong'ambika
2	Nshawa zili ndi tudoyo
3	Nshawa zowola/ zodwala
4	Nshawa zamnanira kwambiri
5	Nshawa zamitundu-mitundu

Q15. Kulingana ndi zimene mwapitamo kapena zimene mwamvapo, kodi ndi zinthu zotani zimene ziwonetsa kuti nshawa iyi ndi yabwino? Conde nenani nshawa zimene zili bwino mupangila pinati bata.....

Q16. Ndi pinati bata yocuruka bwanji imene mupange pa Tsiku?
Kukuluka...../mpimo.....

Q17. Kodi kugulitsa kuti pinati bata yanu? Conde nenani kina ciliconse Kumene mugulitsa pinati bata. Mndandanda wakufunsitsitsa.
.....

Q18. Kodi muli ndi zowonjezerapo zilizonse kapena mafunso aliwonse pa zimene takambitsana pamaphunziro aya?.....

Zikomo.

APPENDIX 5: Two Sample T test Calculations

Hypotheses: $H_0: \mu_L - \mu_I = 0$ vs $H_1: \mu_L - \mu_I \neq 0$, and tested statistics using

Test used:
$$t = \frac{\bar{x} - \bar{y}}{S_p \sqrt{\frac{1}{n_L} + \frac{1}{n_I}}} \sim t_{n_L + n_I - 2} = t_{54 + 55 - 2} = t_{107}$$

Decision used:

The Null hypothesis is rejected if the t value observed is outside the interval:

$(-t_{107,0.975}, t_{107,0.975}) = (-1.98, 1.98)$.

Results from SPSS:

Log of Aflatoxin concentration in ppb: Group Statistics

	ORIGIN	N	Mean	Std. Deviation	Std. Error Mean
Log of amount of Aflatoxin in ppb	International	54	1.6694	.39177	.05331
	Local	55	2.8937	1.05244	.14191

	t-test for Equality of Means				
	t	df	Sig. (2-tailed)	Mean Difference	Std. Error Difference
Log of amount of Aflatoxin in ppb	-8.019	107	.000	-1.22426	.15266

APPENDIX 6: Test of Proportions

Evidence that proportions are different

Let X_L be the number of observations which are below 4ppb in a random sample size n_L from the local population. Let X_I be the number of observations which are below 4ppb in a random sample size n_I from the international population. Let P_L, P_I be the true proportions of acceptable peanut butter by European standards for the local and international populations, respectively.

Hypotheses: $H_0: P_L - P_I = 0$ vs $H_1: P_L - P_I \neq 0$, and using test statistic

Test used: $Z = \frac{\hat{p}_L - \hat{p}_I}{\sqrt{pq\left(\frac{1}{n_L} + \frac{1}{n_I}\right)}} = \frac{\hat{p}_L - \hat{p}_I}{SE_{Error}} \sim N(0, 1)$, \hat{p}_L and \hat{p}_I are estimates of

observations below 4ppb for local and international samples, respectively. $p = \frac{x_L + x_I}{n_L + n_I}$ and $q = 1 - p$, where x_L, x_I are the number of observations below 4ppb for local and international samples, respectively.

Decision used: We reject H_0 in favour of H_1 with $\alpha = 0.05$ if the values are outside

$$(-Z_{0.975}, Z_{0.975}) = (-1.96, 1.96). \quad \hat{p}_L = \frac{2}{55} = 0.0364$$

$$\hat{p}_I = \frac{7}{54} = 0.1296$$

$$p = \frac{9}{109} = 0.0826$$

Difference in proportions for 0- 4ppb European set standard

Difference of proportion	Std. Error Difference	$Z = \frac{\hat{p}_L - \hat{p}_I}{SE_{Error}}$	95% Confidence Interval of the Difference of proportions	
			Lower	Upper
-0.094	0.053	-1.77	-0.011	0.197

Difference in proportions for 0- 15ppb CAC standard

Difference of proportion	Std. Error Difference	$Z = \frac{\hat{p}_L - \hat{p}_I}{SE_{Error}}$	95% Confidence Interval of the Difference of proportions	
			Lower	Upper
.43603	.07081	6.158	.29566	.57640

APPENDIX 7: Summary Statistics of Aflatoxin Brands

Brand name	N	Mean	Minimum	Maximum	Std deviation
Black cat crunchy	3	5.7833	4.55	8.10	2.00769
Black cat smooth	5	6.1300	4.65	7.15	1.26.521
Chocolate crunchy	5	54.8500	47.10	59.80	5.30990
Extra crunchy	14	63.9800	43.15	83.55	16.68756
Jungle Beat plain	12	6.3167	3.70	9.15	1.69039
Plain smooth	5	86.3100	42.90	122.50	35.27206
Extra smooth	14	13.0571	8.85	16.30	2.27468
PnP smooth	2	8.0250	5.70	10.35	3.28805
Pot o'ogold smooth	2	4.7250	4.40	5.05	0.45962
Ritebrand crunchy	2	5.9750	4.85	7.10	1.59099
SaveMor crunchy	2	21.7250	10.50	32.95	15.87455
Spar crunchy	1	4.3500	4.35	4.35	.0
Unclassified	14	32.4964	3.30	147.20	46.84809
Yum caramel crunchy	2	2.3000	1.75	2.85	.77782
Yum creamy	4	5.1250	3.20	6.35	1.42741
Yum crunchy	4	6.1500	4.60	7.20	1.10303
Yum no added sugar	2	4.4250	3.95	4.90	.67175
Yum smooth	25	5.0040	3.25	8.05	1.02316
Total	109	18.8638	1.75	147.20	28.73854