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Determination of Aflatoxin Levels in Groundnuts: A Comparative Study between Domestic and Imported Seed Supplies in Libya

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Abstract:

The production of high-quality seeds presents a significant challenge for small-scale farmers in Libya. These farmers often face difficulties with storing their own seeds, encountering issues such as storage fungi and aflatoxin contamination. Aflatoxins, known to cause various human and animal health problems, are prevalent contaminants in groundnut seeds during storage, primarily produced by Aspergillusflavus and Aspergillusparasiticus fungi. This study compares aflatoxin levels between locally sourced and imported groundnut seeds.

Samples were procured from the local market in Tripoli, Libya, and aflatoxins were extracted and identified using thin-layer chromatography (TLC) with Silica gel. The findings reveal higher aflatoxin levels in local samples compared to imported groundnut seeds. Various factors influence aflatoxin levels, and the detected levels surpass European Union standards. To address this, regulatory measures or awareness campaigns among local farmers are recommended to ensure quality and sustainability in groundnut cultivation in Libya.

Keywords:

Groundnut, aflatoxin, moisture stress, Aspergillusflavus

1. INTRODUCTION

Groundnut(*Arachishypogaea* L.) isone of the most importantoilseed cropsin world agriculturaltrade. It contributes to satisfying the protein needs of many households who cannot afford animal protein.This seed quality is changed by the Aflatoxins which are produced by *Aspergillus flavus*, *Aspergillus parasiticus* andrarely by*Aspergillus nomius* and*Aspergillus tamarii*.[1,2] Aflatoxin contamination can occur in the field before harvest and after harvest during curing, storage and transportation. Contamination due to Aflatoxin is indeed a serious food safety and security concern Sub-Saharan region of Africa including Libya.

Co-exposure to aflatoxins and hepatitis B virus (HBV) greatly increases hepatocellular cancer (HCC) risk[3].During aflatoxins contaminated seed is eaten, the aflatoxins are then transformed into aflatoxin-8,9- epoxide metabolite in the liver which has been implicated in the numerous hazardous consequences in the body [4]. The seriousness in human health affects the local trade and farmers wealth. So the awareness must be givento the farmers for the quality and sustainable development of Groundnut and other crops cultivation. Here the authors concentrate on the comparison of the presence of Aflatoxins in local and imported Groundnut seeds.

2. MATERIALS AND METHOD

Study Place:

The samples were collected fromTripoli market, Libya.

SampleSize:

3 Samples from local (L1-L3) and 3 samples from an Imported (I1-I3) Groundnut seeds (available in the local Tripoli market).Seeds were checked.

Extraction of samples:

Methanol with 25% and 50% concentration was used for the extraction of Aflatoxins from the Groundnut seeds. Seeds were collected from the local market and checked the moisture content and grounded with the above solvent and centrifuged at 3000rpm and collected the supernatant for the presence of Aflatoxin study.

Thin Layer Chromatography:

Methodsofaflatoxinanalysisare mainlybased ontheir solubilitycharacteristics in methanol as wellas on their characteristic fluorescent properties. Their characteristic fluorescence and absorption in the long wavelength range have visible and UV light aids their detection and estimation.Since, 1990 it has been considered the AOAC official method and the method of choice to identify and quantitative aflatoxins at levels as low as 1 ng/g[5].

Requirements:

Reagents:

- Petroleumether(40-60ºC)
- Chloroform
- Anhydroussodiumsulphate

- Silicagel
- Methanol
- AflatoxinBstandard

Apparatus:

- Ambercoloredbeaker
- Ambercoloredconicalflask
- Rotaryvacuumevaporator
- Mechanicalshaker
- TLCplate
- Centrifuge

Procedure:

Weighed accurately 50 g of the well mixed sample in a 500 ml glass stoppered conical flask. Add 150 ml of petroleum ether, mix and keep overnight. Filter out the ether using an ordinary filter paper. To defatted sample in the flask, add 250 ml of methanol mix and place in the mechanical shaker for 30 min. Centrifuged at 3000rpm for 10minutes and collected the supernatant and dried it.

Prepared TLC plates of silica gel G (8-10 g with about 20 ml of water on a plate of 20 x 20 cm size) using an applicator (0.40 mm thickness). Air-dry the plates overnight. Activate the plates in oven at 110 ± 1ºC for 1 h. Diluted the dried extract to a fixed volume (e.g. 200 µl) with benzene: acetonitrile mixture (98:2, v/v). Applied 5 or 10 µl of the diluted extract on the marked TLC plates (3 cm above the bottom edge of the plate) along with standard aflatoxin solution representing 2.5, 5.0, 7.5 and 10 µg concentrations. Ran the plates to about 15 cm height in a solvent system (chloroform: acetone, 90:10 v/v). Removed the plates and air dry. Observe the plates under UV light at 360 nm in a UV chromatography view cabinet. Compare the intensities of the fluorescent of the spots produced by the sample with those produced by the standard for quantifying the aflatoxin content in the sample.

3. RESULTS AND DISCUSSION

Food safety and security are among the major problems inthe current climate of increasing population. Levels of aflatoxin contamination varied from 0.8 to 2200 µg per kg in groundnut kernel. The European Union standard for the total Aflatoxin limit in the Groundnut is 4 µg per kg [(6)].

Table1: Moisture level of local and Imported seed Samples.

S. No.	Sample	Moisture(%)	
		Local	Imported
1	S1	8.0	6.0
2	S2	11.0	5.0
3	S3	9.0	7.0

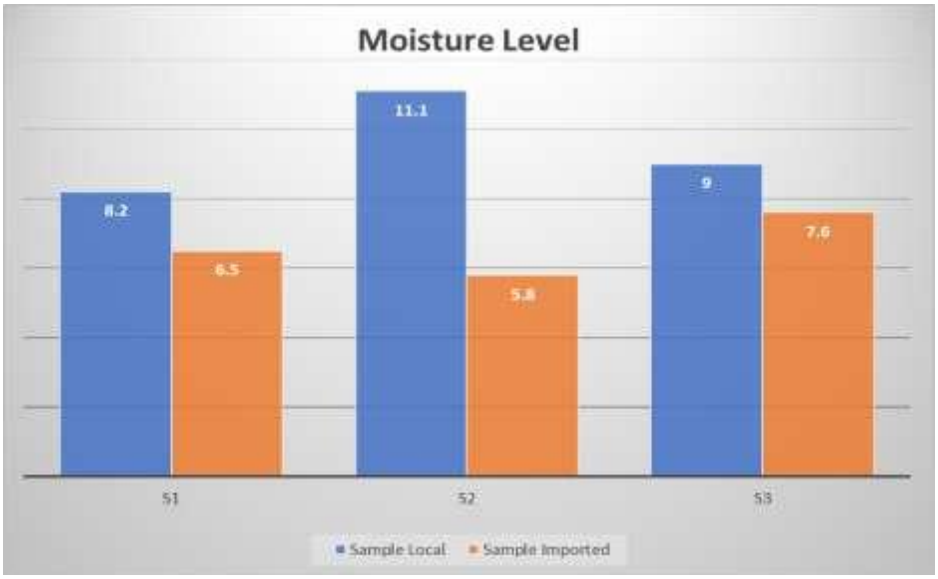

Figure1: Moisture level of local and Imported seed Samples.

Moisture content of a seed determines the nutritional quality of seed..Table 1 and the Figure 1 illustrate the moisture level (%)of local (L1-L3) andimported (I1-I3) groundnut seed samples. In this, Sample 2 of the local seed (L2) have the highest moisture level and it may deteriorate the seed quality. The food contamination by an Aflatoxins may be due to one of the main factor that the presence of moisture contentabove 10% in general.This contamination spoil the nutritional quality and increases the food insecurity in a country.

Table 2:Total Aflatoxin level in Local and Imported samples of both 25% and 50% of Methanol extract

S. No.	Sample	Total Aflatoxin (µgperkg)			
		Local		Imported	
		25% Methanol	50% Methanol	25% Methanol	50% Methanol
1	S1	13.6	15.2	7.5	8.2
2	S2	23.3	47.1	6,2	8.0
3	S3	15.6	17.2	8.1	9.4

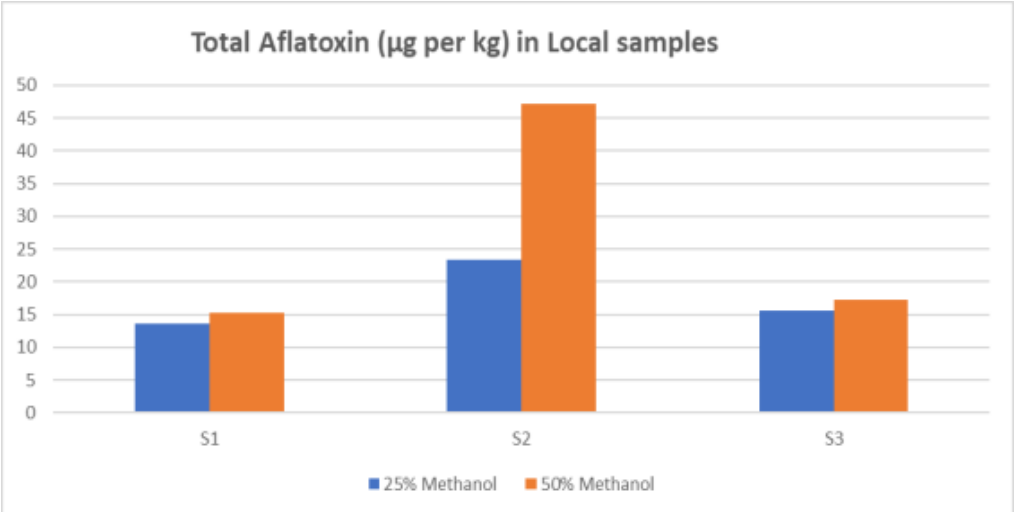

Figure 2:Total Aflatoxin level in Local samples of both 25% and 50% of Methanol extract

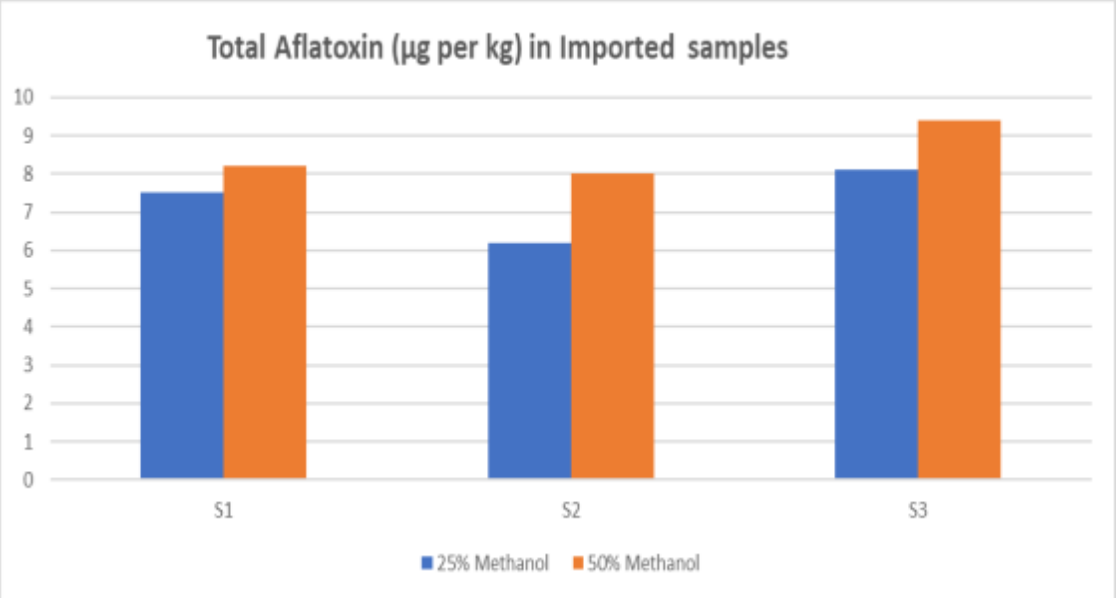

Figure3:TotalAflatoxinlevelinImportedsamplesofboth25%and50%ofMethanolextract

Table 2 and the Figure 2 and 3 indicates the total Aflatox9inlevel in Local and anImported samples of both 25%and 50% of Methanol extract. Higher concentration of methanol(50%) has extracted more quantity of aflatoxins than the 25% of methanol.The local samples of Groundnut contain larger amount of Aflatoxin level thanthe imported one.Thishigher level inthe local samples maybe due to higher moisture content in the local samples and shows the level of total aflatoxins is directly proportional to the moisture content of the seed. This moisture content may favour the growth of *Aspergillus sp*. and induces the production of Aflatoxins.This Aflatoxin contamination may reduce the dietary intake and nutritional properties of the Groundnut seeds [7,8].The storage practice of local farmers, the size of the seed quality and variety may contribute the quality of Groundnut by influencing the aflatoxins production. Based on our results, interventions are needed to reduce aflatoxin levels, which would lead to minimize consumer dietary exposure and prevent disease.

4. CONCLUSION

Local samples have more levelof Aflatoxins thananimported Groundnutseed.Many factorsinfluence the level of aflatoxins.This level also look higher than the European union standards. This level must be maintained by recommending some regulations or creating awareness among the local farmers. Agronomic practices such as crop rotation, use of resistant varieties, insect control, timely planting and harvesting, weed control, adequate fertilization and late season irrigation can reduce pre-harvest aflatoxin production. Additionally, atoxigenic fungi can be applied in the field to competitivelydisplace toxigenic fungi to reduce the population of toxigenic fungi in the soil. Post-harvest aflatoxin contamination of groundnuts can be minimized by rapid and proper drying following harvesting, proper transportation and packaging, sorting and post-harvest insect control.

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