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EFFECTS OF PACKAGING AND PRE-STORAGE TREATMENTS ON AFLATOXIN PRODUCTION AND QUALITY IN STORAGE OF SHELLED AND IN-SHELL PEANUTS IN GHANA.

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ABSTRACT

This study was designed to determine the effect of pre-storage treatments and packaging to control aflatoxin production and quality degradation during storage of shelled under tropical conditions. Sorted shelled peanuts were subjected to the following four pre-storage treatments: raw-clean (Raw-Cl), raw-inoculated-with-A. flavus (Raw-Inf), inoculated-partially-roasted-not-blanch (PRN-blanch), and inoculated-partially-roasted-blanch-with-discolored-nuts-sorted-out (PR-blanch). They were stored for 26 weeks under ambient environmental conditions in Ghana (i.e., Northern and Ashanti region). These samples were analyzed approximately every 4 weeks for aflatoxin levels and quality (peroxide and p- Anisidine values). The results showed that There was no significant increase of aflatoxin levels for partially roasting, and blanching peanuts over the 26 weeks period of storage but for the raw infested peanuts in polypropylene sacks increased slightly from about 30ppb to about 60 ppb for the storage period because of low peanut moisture levels and low humidity

Reduction in quality as determined by p-Anisidine values was 49% for raw-clean, 29.88% for partially-roasted-blanch and 17.6% for partially-roasted-not-blanch samples in hermetic bags.

Partially roasting and blanching peanuts can increase the effectiveness of sorting, and hence aid in reducing aflatoxin along the peanut value-chain. Also, hermetically storing peanuts can

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suppress the growth of aflatoxigenic fungi and the production of aflatoxin under tropical ambient conditions.

Keywords: Aflatoxin, Lipid Oxidation, Peanuts, Hermetic Storage, Polypropylene woven sacks, Ghana

1. INTRODUCTION

Peanut is a leguminous crop that belongs to the family of *Fabaceae*, genus *Arachis*, and botanically named *Arachis hypogaea*. Peanuts are rich in calories and contain many nutrients, vitamins, antioxidants and minerals that are essential for maintaining optimum health (Settaluri, Kandala, Puppala, & Sundaram , 2012). Peanuts also contain a high amount of fats and proteins. According to Eshun , Amankwah, & Barimah, (2013), post-harvest losses of major staple crops like maize and peanuts in Ghana and other countries in Sub-Saharan Africa are mostly due to fungi infestation; particularly by *Aspergillus* fungi mostly *Aspergillus flavus* and *Aspergillus parasiticus* which metabolize to produce aflatoxins.

Aflatoxin contamination of peanuts is a worldwide problem and when injected can cause ailments, for example liver cancer and in addition to its effect on our health, its economic losses are significant (Dorner, 2008). A recent World Bank study indicated that the European Union (EU) regulation on aflatoxins costs Africa $750 million annually in the export of cereals, dried fruits and nuts (Agyei, 2013). Kaaya & Kyamuhangire (2006) reported that most food contamination occurs during post-harvest storage of agricultural products and aflatoxin contamination of foods has been found to increase in storage. Aflatoxigenic fungi grow exponentially in conventional storage for a longer duration as a result of prevalent heat and high humidity (Hell et al., 2010; Guchi, 2015), ranging from 26.7-43.3°C, with a relative humidity of 62–99%, and a moisture content (seed) of 13-20% (Sumner & Lee, 2012). Ideal storage conditions for shelled peanuts around the recommended moisture content of 7-8 % (w.b.), temperature 10 °C and relative humidity conditions 55%, (Sablani & Mujumdar, 2006).

Shelled peanuts in Ghana and most parts of Sub-Saharan African countries are generally stored in jute sacks or polyethylene or polypropylene woven sacks. These sacks are not airtight, and there is evidence that these methods of storage facilitate fungal contamination and aflatoxin development (Hell et al., 2000; Udoh et al., 2000). Mutegi Wagacha, Christie, Kimani, & Karanja, (2013) reported that peanuts stored in polypropylene woven and polyethylene bags were 5.6% and 13.4% more contaminated with total aflatoxin than samples stored in jute bags, respectively. Jute bags easily absorb moisture but allow good airflow while polypropylene woven and polyethylene are non-absorptive but trap heat (Kennedy & Devereau 1994). Results

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from Darko et al, 2018 showed that hermetic packaging controls aflatoxigenic fungi and hence aflatoxin production.

Aflatoxigenic fungi can grow and produce aflatoxins if the oxygen in the headspace is not removed. As reported by Navarro (2012), aflatoxigenic fungi may take as long as thirty (30) days to reach a 3% oxygen level in ultra-hermetic storage (Navarro et al., 2012) and this is too long a period to prevent aflatoxin production under optimum conditions for storage. This means it is best to remove oxygen from peanut surroundings before storage. Oxygen scavengers are capable of removing or reducing the oxygen concentration in a package (Miltz & Perry, 2005) in storage.

Oxygen and other factors, e.g., heat and high humidity, do not only aid in fungi growth and aflatoxin production but also help in lipid oxidation (Nkagawa & Rosolem, 2011). Lipid oxidation is considered as a major cause of food quality deterioration and generation of undesirable odors and flavors. In addition to altering texture and color, lipid oxidation may also decrease the nutritional value of food (Alamed Chaiyasit, McClements, & Decker, 2009).

A study by Pattee and Sessoms (1967) reported that *A. flavus* growth and aflatoxin production was highly correlated with increase in fat acidity, which are major substrates of oil or lipid oxidation. There has been considerable research on aflatoxin production in peanuts during storage and some few examples are Dorner & Cole 2002; Navarro 2012; Mutegi et al 2013. However, studies dealing with the control of aflatoxin production levels in hermetically stored peanuts have been limited. There is a need to fill in this gap and also find ways of maintaining quality of peanuts in Ghana and other Sub-Saharan African countries as a whole. The objective of this study was to find appropriate pre-storage treatments and packaging to reduce aflatoxin production and maintain quality during the storage of shelled peanuts under tropical conditions.

2.0 MATERIALS AND METHODOLOGY

2.1 Study Area

The study was conducted in Ashanti and Northern regions of Ghana. Ghana has ten political regions, but studies were conducted in only these two mentioned regions. The Northern region is one of the leading producers of peanuts and the Ashanti region also produces a sizeable amount of the commodity and has a big market for peanuts. In the Ashanti region, shelled peanuts were stored in Kumasi Central Market and Asuoyeboah Seed Company in Kumasi. The Northern region samples were stored in Aboabo Market and Agric-Ridge Seed Company in Tamale.

2.2 Experimental Design

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Four by two factorial design was used for shelled peanut samples. There were four pre-treatment methods (raw-clean [Raw-Cl], raw-inoculated-with-*A. Flavus* [Raw-Inf], inoculated-partially-roasted-not blanched [PRN-blanch], inoculated-partially-roasted-and-blanched-with-discolored-nuts-sorted-out [PR-blanched]. All the four pre-storage treatments were packed in two packaging systems (i.e., polypropylene woven sacks–PS, and ultra-hermetic pack with oxygen absorber–HPO). One of each treatment combination was randomly sampled from each region for analysis in 0, 2, 6, 10, 14, 18, 22 & 26 weeks.

Aspergillus spp. Spore Suspension Preparation

A. flavus (Aflatoxingenic-producing fungi) was applied to potato dextrose agar and incubated at room temperature for 5 days to enable significant sporulation to take place. After incubation, 10-15ml of sterile distilled water was added to each plate. A sterile plastic inoculation loop was used to loosen the conidia from the potato dextrose agar plates. The suspension created was then filtered through sterile cheesecloth into a sterile 50 ml capacity Falcon tube. Spores were enumerated using a hemocytometer (Neubauer, Hausser Scientific, Horsham, USA), and appropriate dilutions were made from the stock spore solution.

Peanut Inoculation

Peanuts variety (Chinese) were obtained from Tamale market in the Northern region of Ghana. Shelled peanuts were visually inspected for defects, e.g., discoloration and mold growth, and defective peanuts were removed from the batch. The amounts of peanuts needed to be inoculated were infected by spraying 10^5-10^6 CFU/ml of *A. flavus* on the surface. Infected peanuts were then incubated at a temperature of 30 ±1 ^{0}C and water activity of 0.85± 0.01 for 7 days before being applied to the samples and packaged.

Sample Preparation and Packaging

Thirty-three (33) sacks of raw shelled peanuts, each weighing about 80kg, and 25 bags of in-shell peanuts, also weighing about 45kg, were sorted manually (removing discolored, moldy, and defective peanuts by hand). After sorting, mixtures with a ratio of 100 g of inoculated peanuts to 50 kg of clean peanuts were then partially roasted in batches in a pot over fire at an average pot temperature of 190 ºC for about 50 minutes in batches for 17 bags. Half of the partially roasted peanuts were blanched (removing the testa manually) and the other half not blanched after cooling. The blanched portion was then hand-sorted to remove burnt, defective and discolored grains.

The remaining 16 bags of raw shelled peanuts did not receive any further pre-storage treatment, and 8 bags out of these 16 bags was regarded as Raw-clean (Raw-Cl). The other half was

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processed for "raw-inoculated-with-*A. flavus*" - (Raw-Inf) samples; with each pack containing 10kg of clean peanuts and 20g of inoculated peanuts. All peanut treatments were packaged in the hermetic bags (special polyethylene storage bags Super Grain-bag III, Grain Pro. Inc., Concord, MA, USA) with oxygen absorber (Oxyfree®, Marietta, GA, and U.S.A) and polypropylene woven sacks (from the local market). Each pack contained 10 kg peanuts. The four shelled treatment samples were stored in the market places and seed companies in Ashanti and Northern regions of Ghana under normal environmental conditions for 26 weeks. The various treatment combinations were analyzed for aflatoxin, peroxide value, and p-Anisidine value.

Sampling (Quartering Method)

After samples taken out of storage, each bag was poured onto a clean, non-absorbent surface and mixed thoroughly and gathered together into a cone-shaped pile. The top surface was flattened out and then the pile was divided into quarters, A, B, C, and D. Incremental samples of 125g were picked from each quarter and the procedure was repeated to achieve a laboratory sample of 2 kg.

Moisture Content Analysis

About twenty grams of peanuts sample was placed in moisture (Adam Moisture Meter Pmb 53) after samples taken out of storage. Then the moisture content was recorded after each sampling event with the digital moisture meter.

Aflatoxin Extraction

Aflatoxin was extracted using methods described by Sirhan, Tan, Al-Shunnaq, Abdulra'uf, & Wong, (2014), with a few modifications. Using an IKA homogenizer, slurry of peanut and water (1:1 ratio) was prepared, 2g of slurry weighed into a 15ml centrifuge tube and 3ml of 60:40 (v/v) methanol acetronitrile solution was added. The resultant mixture was vortexed using a Genie Vortex machine for 3mins. 1.32g of anhydrous MgSO4 and 0.2g of NaCl were added to the mixture and then vortexed for 1min. The tube was centrifuged for 5min at 4000rmp and the upper organic layer filtered through a 0.45□m nylon syringe prior to injection. A volume of 100 µl of the filtered extract was injected into the HPLC for analysis.

Aflatoxin Testing (High Performance Liquid Chromatography)

A Cecil-Adept Binary Pump HPLC coupled with Shimadzu 10AxL fluorescence detector (Ex: 360nm, Em: 440nm) with Phenomenex HyperClone BDS C18 Column (150 x 4.60mm, 5µm) was used for the aflatoxin analysis. The mobile phase used was methanol (HPLC grade): deionized water solution (40:60, v/v) at a flow rate of 1ml/min with column temperature

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maintained at 40°C. To 1 liter of mobile phase were added 119 mg of potassium bromide and 350ul of 4M nitric acid (required for post column electrochemical derivatization with Kobra Cell, R-Biopharm Rhone). Aflatoxin Mix (G_1, G_2, B_1, B_2) standards ng/g (ppb) were prepared from Supelco® aflatoxin standard of 2.6ng/µL in methanol. The concentrations of B1 and G1 were 0.5, 1, 2, 8, 16 ppb per 100µl injection. The concentrations of B2 and G2 were 0.15, 0.3, 0.6, 2.4, 4.8 ppb per 100µl injection. Limit of Detection and Limit of Quantification of total aflatoxin were established at 0.5ng/g (ppb) and 1ng/g (ppb).

Aflatoxin Calculation

Aflatoxin level was calculated by:

$$\frac{ng}{g}(ppb) = A\left(\frac{T}{I}\right) * \left(\frac{1}{W}\right) \text{---------- (1)}$$

Where A= Nano gram of aflatoxin as eluate injected, T=final test solution eluate volume (ul), I=volume eluate injected into LC (ul), W=mass (g) of commodity represented by final extract.

Oil Extraction

Oil was extracted from 600g of peanuts sampled by using a hand cranked oil press (Piteba, brand). The peanuts were put in the hob of the oil press and the crank turned to get the lipids out.

Peroxide Value

Peroxide value (PV) is one of the most widely used chemical tests for the determination of the quality of fat and oil. The peroxide values of the oil extracted from the different samples were determined using the American Oil Chemists' Society (AOCS) official method Cd 8-53 (1998). Briefly, about 5g oil was put in a 125ml glass Erlenmeyer flask, 30ml acetic acid-chloroform solution (3:2, v/v) was added and shaken. Then 0.5ml saturated potassium iodide solution was added swirled gently for exactly one minute, and the flask stoppered and shaken vigorously to liberate the iodine from the organic layer. Starch indicator (1 ml) was added into the mixture and then titrated with 0.1 N sodium thiosulfate until the blue grey color disappeared. The volume of the titrant was recorded to the nearest 0.01 ml.

The peroxide value (milliequivalents peroxide/ 1,000g) sample) was determined using the formula:

$$PV = \frac{(S - B) * N * 1000}{m} \text{---------- (2)}$$

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where B is the titration of blank (ml), S is the titration of sample (ml), N is the normality of the sodium thiosulfate solution and m is the weight of the sample

P-Anisidine Value

Oil was extracted from the treatment combinations of peanuts after storage and p-Anisidine value was determined by following AOCS official method Cd 18-90 (1998). About 3g of the oil sample was weighed into a volumetric flask and mixed with 22ml iso-octane. The absorbance of this solution (Ab) was measured at 350nm with a spectrophotometer, using iso-octane as the blank. Then 5ml of the mixture was measured and the blank was transferred separately into a new test tube. 1ml of p-Anisidine was added to each test tube and incubated for 10 minutes. The absorbance (As) was recorded and the p-Anisidine value (p-AV) were given by the formula:

$$p\text{-}AV = 25\left(1.2 * As - Ab\right)/m \text{ ---------- (3)}$$

Where As is the absorbance of the fat solution after reaction with the p–Anisidine reagent, Ab is the absorbance of the solution of the fat and m is the mass, in grams, of the test portion.

Statistical Analysis

Data were analyzed using Analysis of Covariance (ANCOVA) in JMP-PRO version 13 (SAS Institute Inc., Cary, N.C., U.S.A.) treating time (weeks) as a continuous variable to adjust for least square estimates of treatment and interaction means of pre-storage treatments and packaging system. Least square means of all the treatment combinations were separated using Turkey's Kramer Test with corrected α=0.05 when a significant F-value was obtained.

3.0 RESULTS AND DISCUSSION

Aflatoxin

The production of aflatoxin of the four shelled peanut treatments in the conventional polypropylene woven sacks and hermetic bags with oxygen absorbers in Tamale in the Northern region and Kumasi in the Ashanti region of Ghana is as shown in figures 1 a, b, c, & d. The partially-roasted-blanched samples had relatively low mean aflatoxin levels of 4.13 ppb, followed by 18.8 ppb for partially roasted not blanch samples, 21.25 ppb for raw clean samples, and 39.91ppb for raw inoculated samples (Table 1). Roasting can reduce aflatoxin levels (Yazdanapanah et al., 2005), blanching and sorting can further reduce them (Cole et al., 1995). The *A. flavus* fungi introduced into the raw inoculated samples did not grow and produce aflatoxin as expected, because the moisture content of the peanuts used in the experiment was relatively low, in the range of 5-11%.

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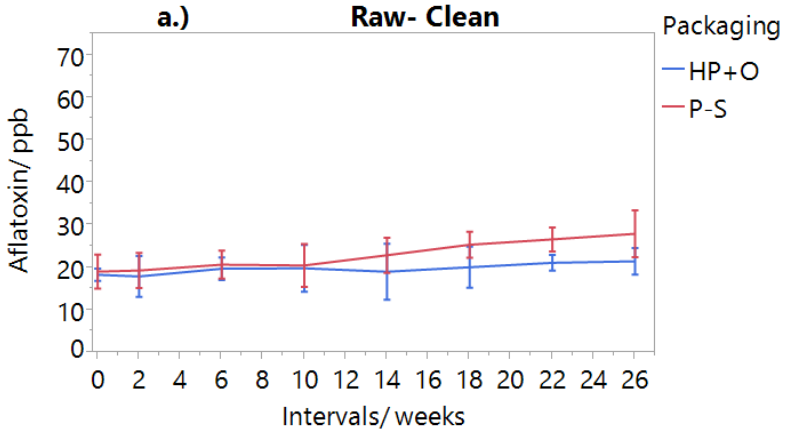

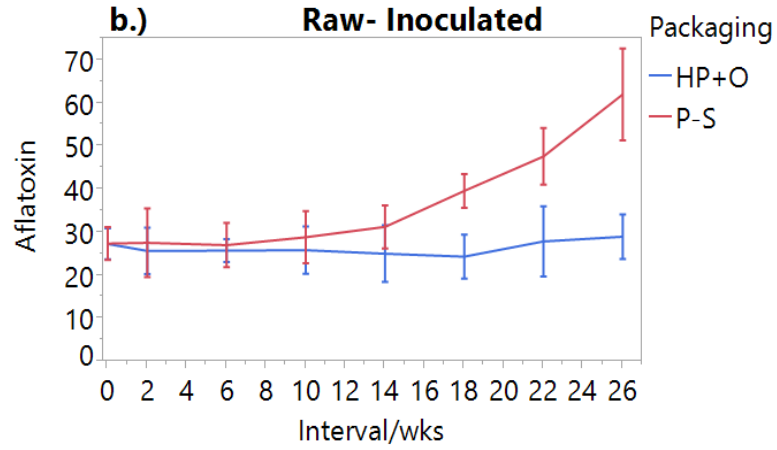

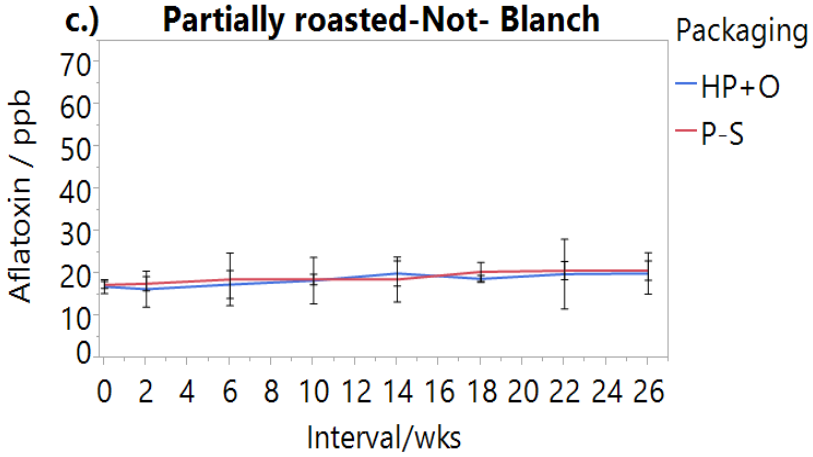

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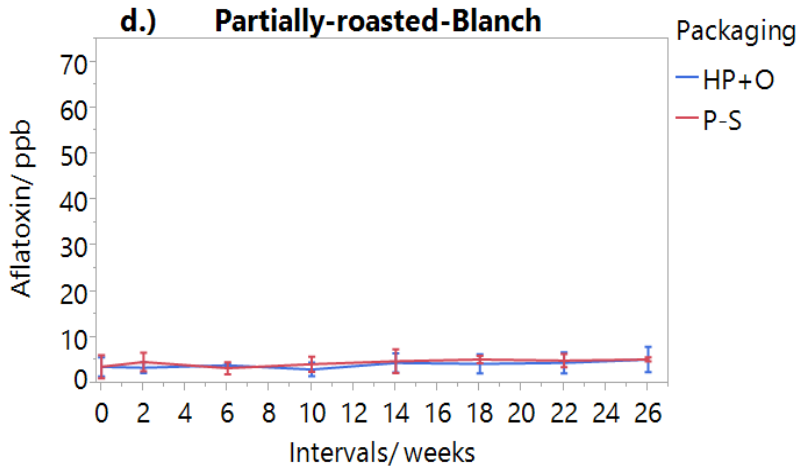

Fig. 1- Aflatoxin production results of the raw clean (a), raw inoculated (b), partially roasted not blanched (c) and partially-roasted-blanched (d) peanut treatments in the two packaging systems (PS, and HPO) over 26 weeks of storage.

Moisture level is also critical for fungi growth, sporulation and mycotoxin production (Sanchis & Magan, 2004; Magan 2007). There were slight increases in aflatoxin levels for the raw clean samples in polypropylene woven sacks. This shows that although peanuts were sorted there was evidence of small doses of aflatoxin-producing fungi, which produced small amounts of aflatoxin during the twenty-six (26) weeks of storage, just as in the raw inoculated samples. The aflatoxin production of the samples in the hermetic bags with oxygen absorbers was quite stable for the whole storage period, because of the deprivation of oxygen in the package. Reduced oxygen levels were able to reduce aflatoxin production of *A. flavus* to safe and acceptable levels <20 ng/g - ppb (Ellis, Smith, Simpson, Khanizadeh, & Oldham, 1993).

The fungi were dormant from week zero (0) even for the PS packages which was in December, to week ten (10) in February which was the harmattan (Dry-Season) season in Ghana. During this season humidity was relatively low ranging from 16.1-76.2% for Ashanti region and 11.0-55.0% for Northern region (Table 2) as recorded. There was an increase in aflatoxin level from week 18 onwards when the weather was relatively humid; but before the 18th week, the aflatoxin levels did not increase. Although aflatoxn levels increased slightly from about 30ppb to about 60 ppb for the storage period. Fungi can survive under adverse conditions when they produce microscopic spores (Stajich et al, 2009). Also, Coley-Smith (1971) reported that fungi such as aflatoxigenic ones can produce dense aggregations of fungal tissue called scelerotia, and these structures help them survive challenging conditions such as freezing temperatures and long term absence of a host. These Aspergillus flavus fungi can survive until favorable growth conditions are available (Sumner & Lee, 2012).

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Regardless of the package type, there was no increase of aflatoxin levels of the partially roasted blanched and non-blanched samples. It was known that *A. flavus* cannot survive temperatures above 80 °C (H. Mehl, personal communication, May 22 2015). Therefore, partially roasting peanuts at a temperature of 190 °C for 50 mins must have killed all fungi on the inoculated peanuts. This observation probably explains why there was no increase in aflatoxin level during the storage period.

Blanching the peanuts and sorting out infested and discolored ones was very effective in reducing the aflatoxin levels by about 90 %, compared to raw-inoculated samples in polypropylene woven sacks. Aflatoxin levels in Kumasi and Tamale were significantly different from each other at $p<0.05$, (23.17 ppb) at Kumasi where the environment was more humid than Tamale, which recorded 18.89 ppb during the 26 weeks of storage. Overall, at 95% confidence level, there was a significant difference in aflatoxin levels for all the four pre-storage treatments as well as a significant difference between the two packaging systems.

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Table 1: Mean aflatoxin, peroxide and p-Anisidine values of Raw-Clean, raw-inoculated, Partial-roast-blanch and Partial-roast- not-blanch samples in the Polypropylene woven sacks- PS, and hermetic pack +02 -HPO, packaging systems.

Aflatoxin/ppb			
Treatment/ Package	HPO	PS	Overall Mean
Raw-Clean	19.71 ± 1.13^{c}	22.80 ± 1.13^{c}	21.25 ± 0.80 ^{b}
Raw- inoculated	35.12 ± 1.13 ^{b}	44.70 ± 1.13^{a}	39.91 ± 0.80 ^{a}
Partial-roast-blanch	4.13 ± 1.13^{d}	4.14 ± 1.13^{d}	4.13± 0.80 ^{d}
Partial-roast-not-blanch	18.53 ± 1.13 ^{c}	19.08 ± 1.13^{c}	18.8 ± 0.80 ^{c}
Overall Mean	19.37 ± 0.56 ^{b}	22.68± 0.56 ^{a}	
Peroxide Value/meq/kg			
Raw-Clean	8.87 ± 0.19 ^{e}	10.54 ± 0.19^{d}	7.71 ± 0.13^{c}
Raw- inoculated	11.77 ± 0.19 ^{c}	15.29 ± 0.19 ^{a}	13.53± 0.13 ^{a}
Partial-roast-blanch	10.69± 0.19 ^{d}	13.72 ± 0.19^{b}	12.21± 0.13 ^{b}
Partial-roast- not-blanch	10.99 ± 0.19 ^{cd}	15.24± 0.19 ^{a}	13.12± 0.13 ^{a}
Overall Mean	10.58 ± 0.10 ^{b}	13.70 ± 0.10^{a}	
P-Anisidine Value /meq/Kg			
Raw-Clean	5.12 ± 0.18^{e}	6.18± 0.18 ^{d}	5.65 ± 0.13 ^{c}
Raw- inoculated	8.49 ± 0.18^{c}	10.04± 0.18 ^{b}	9.26 ± 0.13 ^{b}
Partial-roast-blanch	7.04± 0.18 ^{c}	11.26± 0.18 ^{a}	9.65 ± 0.13 ^{ab}
Partial-roast- not-blanch	8.27± 0.18 ^{c}	11.68 ± 0.18^{a}	9.98 ± 0.13 ^{a}
Overall Mean	7.49± 0.09 ^{b}	9.79± 0.09 ^{a}	
Levels not connected by the same letter are significantly different			

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Table 2: Relative humidity and temperatures in Ashanti and Northern regions of Ghana during the storage period. Minimum-maximum (Average)

Interval/ weeks (Shelled &InShell)	Relative Humidity/%		Temperature/°C	
	Ashanti Region Range (Average)	Northern Region Range (Average)	Ashanti Region Range (Average)	Northern Region Range (Average)
Week 0	30-46(39.6)	14-38(21.5)	24.2-31.9(27.6)	17.6-42 (30.0)
Week 2	30.3-47.1(37.2)	12-47(20.2)	24.2-31.9(28.6)	18-42.5 (31.9)
Week 6	16.2-76.2(39.3)	13-55(21.3) 28.5	24.8-36.1(30.0)	20- 41.8 (29.5)
Week 10	16.1-71.9 (34.7)	11-49(22.5) 24.3	27.1-39(31.9)	17.9 – 42 (31.0)
Week 14	36.0-82.6(62.5)	14-83(48.8) 54.3	25.6- 37.9(32.2)	22- 42.5 (33)
Week 18	47-86.5 (70.4)	36-88(59.5)	25.4-35.2(30.9)	22- 39.8 (32.3)
Week 22	52- 94.0 (75.8)	33-87.1 (65.3)	24.6 – 36.5(30.9)	21.5 – 40.5 (30.9)
Week 26	62.5 – 90.7 (79.2)	44.0-96.0 (71.25)	24.7 – 34.6 (29.0)	21.50 – 36.50 (29.3)

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Table 3: Percentage reduction of aflatoxin, peroxide value and P-Anisidine values of the four pre-treatment (raw clean, raw inoculated, Partial-roast blanch and Partial-roast-not-blanch) in Polypropylene woven sacks- PS, hermetic pack +O_2 -HPO packaging system with respect to the raw- inoculated samples in polypropylene woven sacks.

Treatment/ Package	Aflatoxin/ %	Peroxide Value/ %	P- Anisidine Value/ %
Raw-Cl/HPO	55.9	44.0	49.0
Raw-Cl/PS	49.00	31.1	38.5
Raw-Inf/HPO	21.4	23.0	15.4
Raw-Inf/PS	0.0	0.0	0.0
PR-B/HPO	90.8	30.1	29.9
PR-B/PS	90.7	10.3	-12.2
PR-NB/HPO	58.6	28.1	17.6
PR-NB/PS	57.3	0.3	-16.3

Peroxide Value

Figures 2 (a, b, c, & d) present the trend of peroxide values (primary oxidation parameter) for the peanuts stored under Ghanaian ambient conditions. All the four treatments experienced only minor increases in peroxide levels during storage. This may be due to the fact that external factors such as light, elevated temperatures and oxygen may have contributed to the generation of oxidative degradation products from their precursor fatty acids (Kamal-Eldin, 2006; Merrill et al, 2008). Additionally, peroxide values from samples in the polypropylene woven sacks plateaued after week 18. As reported by Hill (1994), the use of peroxide value as flavor quality indicator is only reliable during the initial stages of lipid oxidation because the peroxide value increases to a maximum and then decreases as storage time increases. All four-treatment samples stored in the polypropylene woven sacks exhibited the highest values of peroxide although they were stored under the same environmental conditions. One possible reason for higher peroxide reading is that the sacks had oxygen and this facilitated lipid oxidation as compared to hermetic packs.

Statistical analysis (Table 1) of quality (Peroxide value) of the peanut samples for the two storage systems HPO and PS were significantly different from each other at $P< 0.0001$, indicating that the storage system utilized may have influenced the peroxide levels in the

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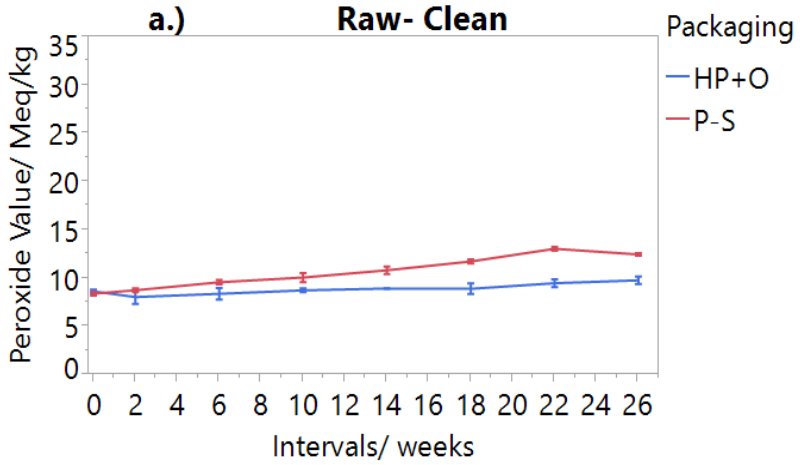
samples. There were no significant differences in peroxide values between samples stored in Kumasi and Tamale (P-value=0.56), although it was expected that samples from the Northern region would be higher because of higher temperatures, compared to Kumasi (Table 2). Kumasi samples had relatively higher aflatoxin levels; and it is known that aflatoxin is highly correlated with peroxide value (Darko, Mallikarjunan, Kaya-Celiker, Frimpong, & Dizisi, 2018), hence high peroxide values for the Kumasi samples.

Furthermore, there was a very good correlation (r= 0.84, r^2 =0.70, RMSE-=1.34, P-value <0.0001) between aflatoxin production and quality (peroxide value) for inoculated samples in polypropylene woven sacks (fig. 3). As reported by Pattee and Sessoms (1967), increase in fat acidity was highly correlated with visible A. flavus growth and aflatoxin production. Overall, there was no significant difference in peroxide values of both raw inoculated samples and partially roasted non-blanched peanut samples (P-value = 0.13). This similarity in peroxide values may be due to the fact fungi growth aids in quality deterioration and elevated heat also helps in lipid oxidation. This was confirmed by Nakagawa and Rosolem (2011), as cited in Santo et al (2016) that high temperatures and relative humidity can also contribute to the deterioration of seeds because of lipid peroxidation. The rest of the pre-treatment samples were significantly different from one another at P <0.0001. Comparing peroxide values with respect to the raw-inoculated samples in polypropylene woven sacks, the raw clean treatment samples in both polypropylene woven sacks and hermetic bags were best in reducing oxidation by 31% and 42%, respectively. This was followed by partially roasted blanched samples with 30%, and 28.1% for partially roasted non-blanched samples in hermetic packages.



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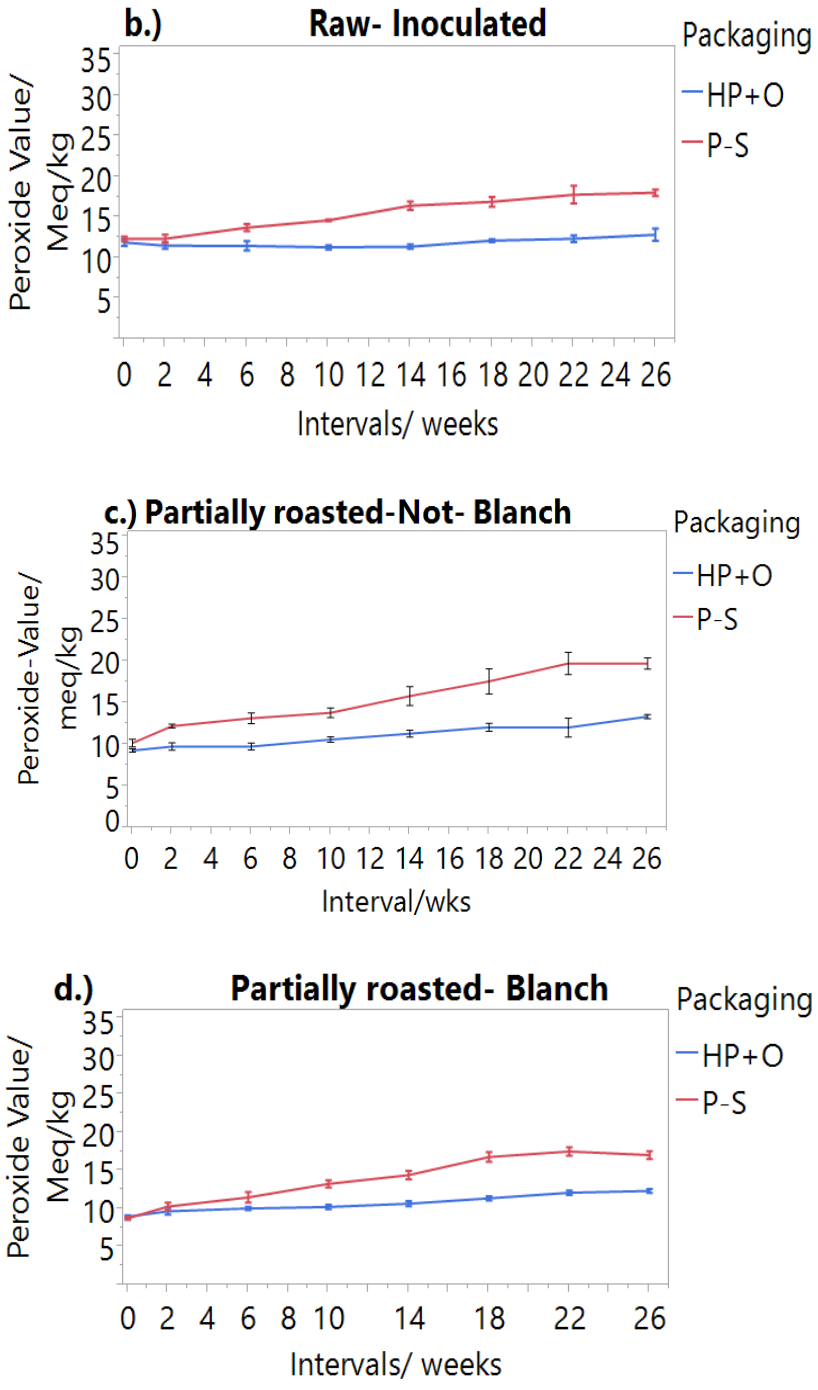

Fig. 2- Peroxide Value results of the raw clean (a), raw inoculated(b), partially roasted not blanched (c) and partially roasted blanched (d) peanut treatments in the two packaging (PS, and HPO) systems

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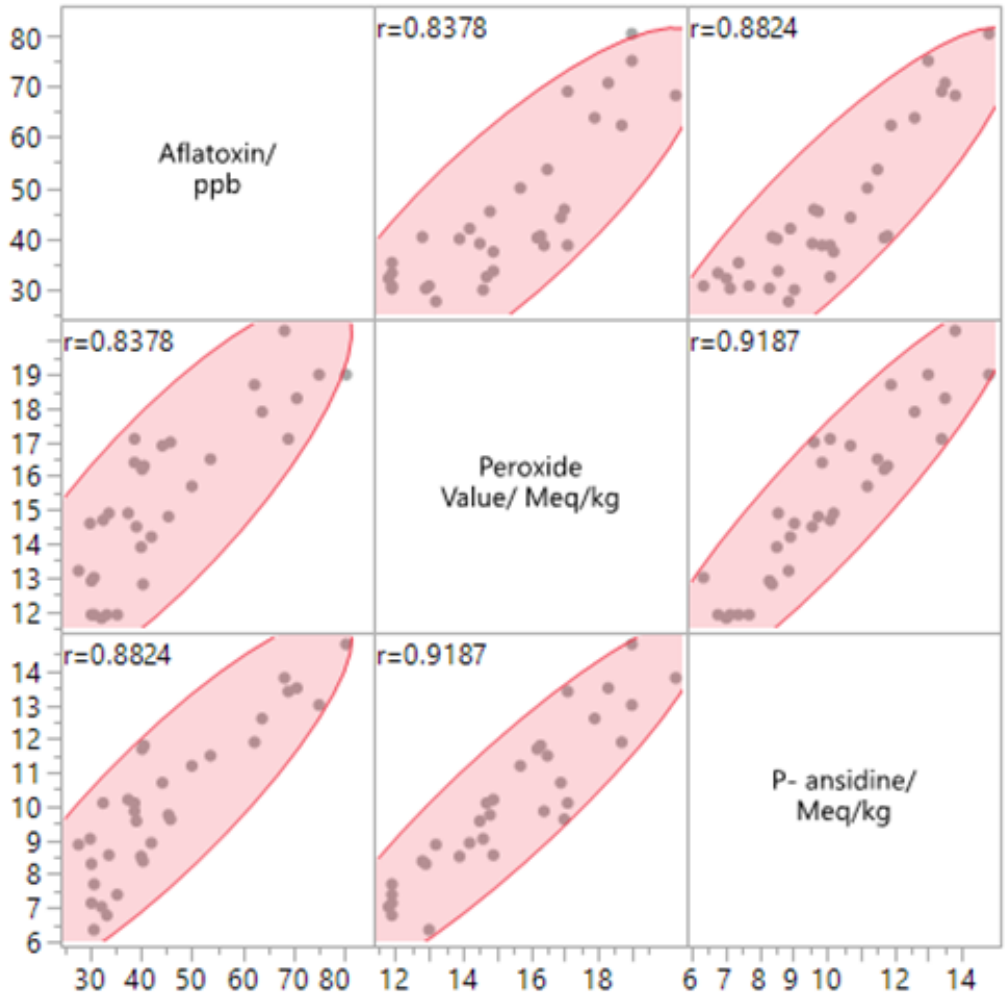

Fig. 3: Correlation matrix of aflatoxin production and quality (Peroxide and p-Anisidine value) for shelled raw infested peanuts.

P-Anisidine Value

P-Anisidine trends for the four pre-storage treatments (raw-clean, raw-inoculated, partially-roasted-not blanch, and partially roasted blanch) in the two packing systems (Polypropylene woven sacks and hermetic bags with oxygen absorbers) are presented in fig 4 (a, b, c, and d). The amount of p-Anisidine value also increased with storage time, and the values were higher in the samples stored in the polypropylene woven sacks (PS) as compared to the hermetic bags with oxygen absorber (HPO). It is known that this hermetic system suppresses oxidation because of lack of oxygen in the sack. The slight increase of p- Anisidine values noted during the storage

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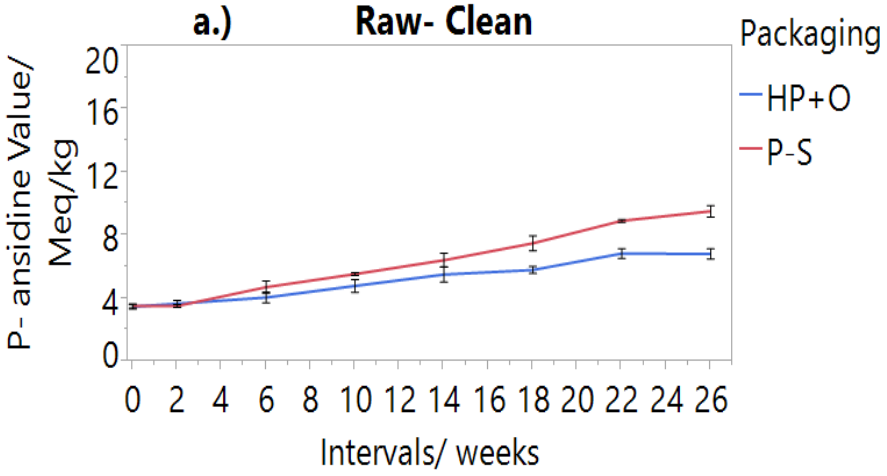
period may be due to increased lipid oxidation caused by factors such as light and elevated temperatures (Kamal-Eldin, 2006; Merrill et al, 2008).

The mean of the p-Anisidine values of peanut samples indicated that there were significant differences in the values between the two packaging systems at p- value < 0.0001 (Table 1). Thus the p-Anisidine value in peanuts could have been affected by the packing system. For the four pre-treatments regardless of the packaging system, the partially-roasted-blanched peanut samples were statistically not different from the partially roasted not blanched and raw inoculated samples at p values = 0.27 & 0.14, respectively. The partially roasted not blanched peanuts in the polyethylene sacks recorded the worst quality degradation of 11.68 meq/kg, as compared to raw clean samples in hermetic bags recording 5.12 meq/kg. Hence, hermitic bags were the best in maintaining quality. P-Anisidine values for the partially roasted blanch samples in polypropylene woven sacks were expected to be higher than those for the partially roasted non-blanched samples because of the testa (seed coat) as an anti-oxidant (Constanza et al, 2012) which is supposed to protect peanuts from oxidation. However, these not blanched samples had higher aflatoxin levels, and these in turn can be a factor for higher oxidation (p-Anisidine) levels.



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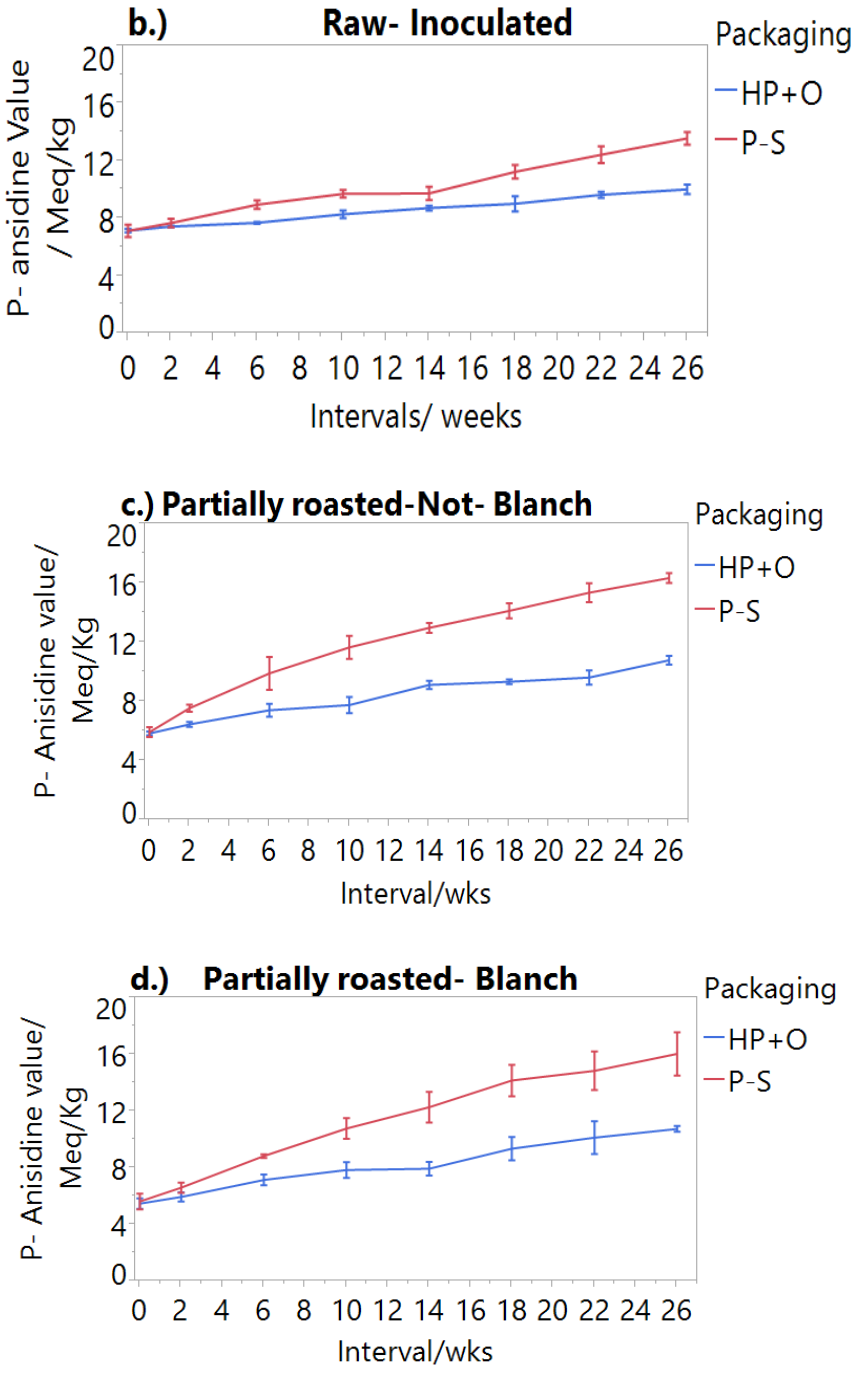

Fig. 4: (a, b, c,& d)- P-Anisidine Value results of the raw clean(a), raw inoculated(b), partially roasted not blanched (c) and partially-roasted-blanched (d) peanut treatments in the two packaging systems (PS, and HPO).

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Aflatoxin levels also had a strong correlation with peroxide and p-Anisidine values (Darko et al 2018). There was no significant difference in quality (p-Anisidine value) between peanut samples stored in Ashanti and Northern regions of Ghana at P>0.05. Although we expected that, samples from the Northern region be higher than from Ashanti region because of higher storage temperatures, ranging from 17.6°C–42.5 °C in Northern region and 24.2-37.9°C in Ashanti region (Table 2). On the other hand there was higher aflatoxin levels in the samples from Ashanti region due to relatively high humidity, hence the reason of no difference in values. The p-Anisidine values of raw clean samples in polypropylene woven sacks (6.19 meq/kg) and hermetic bags (5.12 meq/kg) was best in quality maintainance. This was followed by partially roasted blanched peanut samples in hermetic bags (7.04 meq/kg) and then partially roasted not blanched samples in hermetic packs followed (8.27 meq/kg), with raw-inoculated samples recording 8.48 meq/kg recording the least (Table 1).

There was a strong correlation (r= 0.88**,** r^2=0.78, RMSE-= 1.08, p-value <0.0001) between the aflatoxin production and the quality index (p-Anisidine) (Fig. 3). The trend line indicates that the higher the aflatoxin produced, the higher the quality deterioration. Comparing p-Anisidine values of the raw-inoculated samples in polypropylene woven sacks to those of the rest of the treatment combinations, raw clean treatment samples in both polypropylene woven sacks and hermetic bags were best in reducing oxidation by 38% and 49 %, respectively; followed by partially-roasted-blanched samples in hermetic bags recording 29.88% and 17.6% for partially roasted not blanched samples in hermetic bags (Table 3)

4.0 CONCLUSION AND RECOMMENDATION

There was no significant increase of aflatoxin levels for partially roasting, and blanching peanuts over the 26 weeks period of storage. This can be attributed to the fact that, roasting can kill the aflatoxigenic fungi and halt aflatoxin production during storage, thus aiding in reducing or eliminating aflatoxin levels along the peanut value chain. The raw infested peanuts in polypropylene sacks increased slightly from about 30ppb to about 60 ppb for the storage period because of low peanut moisture levels and low humidity

From the study, it was concluded that hermetic storage of peanuts can suppress aflatoxigenic fungi growth and aflatoxin production under ambient conditions. Therefore, to reduce and also prevent aflatoxin production in storage under favorable environmental conditions, it would be best to store peanuts hermetically, since sorted peanuts can still produce aflatoxin if there is even the least trace of fungi on them.

Raw clean peanuts can best maintain quality during storage but might still have high aflatoxin levels. Therefore to have low levels of aflatoxin before, during, and after storage, as well as to

maintain peanut quality, it would be best to partially roast peanuts, blanch them and sort out the infested and discolored ones, and hermetically store them.

The perfect window was missed due to timely access to money, and so time for the study (December to June) did not favor aflatoxin production very much, therefore, I recommend a research work to be done for a whole year under Ghana environment starting from July-August because that is when actual peanuts storage time starts and also when the temperatures and humidity is conducive for aflatoxin production.

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