ACR Original Article

Occurrence of mycoflora in staple grains on retail outlets in Kaduna and Nasarawa States of Nigeria.

Mokogwu ATH[1], Onohwakpor EA[2], Mokogwu EE [3]

[1]Department of Chemical Pathology, Faculty of Clinical Medicine, Delta State University, Abraka, Nigeria
[2]Department of Obs-Gyn, Faculty of Clinical Medicine, Delta State University, Abraka, Nigeria
[3]Department of Agricultural Extension and Management, FCAHPT, NVRI, Vom, Nigeria

Abstract

Aim: Mycoflora and toxins from mould when found in grains can be harmful and even deadly when consumed by man. This work was designed to ascertain the presence and extent of growth of mycoflora in staple grains in Kaduna and Nasarawa States of Nigeria.
Methods: The staple grains (local variety) consisting of maize (Zee mays), beans (Cowpeas-Vigna simensis), groundnuts (Arachis hypogaea) and yam (Diascorea spp) were obtained from selected six Local Government Areas of the two States. Sampling was done twice; in dry and rainy seasons. Mycoflora isolation and identification were done by standard techniques.
Results: In rainy season, there was significant difference ($p<0.001$) in the mean of Aspergillus flavus and the rest fungi isolates in Kaduna and Nasarawa States. Also in dry season, there was significantly different ($p<0.001$) in the mean of Aspergillus flavus and the rest fungi isolates in both Kaduna and Nasarawa States. Equally in both Kaduna and Nasarawa States, there was significant difference ($p<0.05$) in the mean of fungi isolates in the four staple grains, with the mycoflora isolates in maize being the highest in the two States compared to the rest of the grains during rainy and dry seasons. Rainy and dry seasons comparison of the mean fungi isolates from the four grains' samples shows no significant difference ($p>0.05$) in fungi isolates amongst the grains in both Kaduna and Nasarawa States. Conclusion: The occurrence of Aspergillus flavus, Fusarium spp., Phoma spp., Fusarium moniliforme, A fumigates, A niger, Penicillium funiculosum and other toxigenic mycoflora in grains clearly shows the urgency for increased surveillance particularly as it constitutes great danger when consumed by humans.

Keywords: Mycoflora, occurrence, maize, beans, groundnuts, yam, Kaduna and Nasarawa States.

Introduction

Mycoflora most commonly known as fungi are actually moulds which infect crops in the field or during storage. Mould and toxins from mould when found in grains can be harmful and even deadly when consumed by man. Moulds usually grow under specific conditions of temperature and humidity or in disease / saturated soil[1,2,3]. Mouldy feeds may cause a variety of health problems in animal and humans especially respiratory diseases from breathing in mouldy spores[3,4,5]. Mouldy feeds are also less palatable and may cause a reduction in feed intake, resulting in weight loss. When moulds are shocked by sudden fluctuations in temperature (freezes or hot spells), they exude poisons called mycotoxins [6,7]. They suggested that individual moulds / mycoflora or more mycotoxins together may have a greater toxic effect than any one alone. Penn[8] in 2004 stated that mycoflora could be considered nature's garbage disposal. Without them, the term "biodegradable" would not be so significant to our planet and we would have mountains of leaves, dead trees and other organic materials sitting around us, all deposited since the beginning of time. Mould growth and mycotoxin production and contamination may also occur during mixing and delivery of grains and animal feeds. When produced on human foods or animal feedstuffs, mycotoxins constitute a potential toxic hazard which cause disease and reduced production [5,9,10,11,12,13,10,14,15,16]. Mycotoxicoses is the toxic disease

Corresponding Author
Mokogwu ATH
Department of Chemical Pathology,
Faculty of Clinical Medicine, Delta State University,
Abraka, Nigeria
Mobile no: +2348038606571
Email: tmokogwu@yahoo.com

caused by exposure to mycotoxins. The toxigenic mycoflora are not directly involved with the disease in the host and may no longer be present in the contaminated grains or foodstuffs[17]. Each mycotoxin is produced by one or more very specific fungal species. In some cases, one species can form more than one mycotoxin for example the aflatoxins can be formed by *Aspergillus flavus, Aspergillus parasiticus* and limited other *Aspergilli* while Ochratoxin A is considered to be the main product of *Aspergillus ochraceus* in tropical regions and *Penicillium verrucosum* in temperate areas[18]. WHO[19] in 2002 stated that any crop or foodstuff that is stored for more than a few days is a target for mould growth and mycotoxin formation. EMAN[18] in 2004, documented that the major commodities affected are cereals, nuts, dried fruits, coffee, cocoa, spicies, oil seeds, dried peas and beans and fruits particularly apples. EMAN[18] also in 2004 stated that mycoflora metabolic products may be found in bears, and wine resulting from the use of contaminated barley, other cereals and grapes in their production. These metabolic products enter the human food chain through meat or other animal products such as egg, milk and cheese as the result of livestock eating contaminated feed hence they cause a diverse range of toxic effects[20]. Chrietensen[2] in 1982 opined that chronic effects are of concern for the long-term health of the human population as some of the most common metabolites are carcinogenic, genotoxic or may target the kidney, liver or immune system. WHO[21] in 2000 stated that National and International Organizations are constantly evaluating the risk that such metabolites pose to man. Gbodi *et al*;[22] in 1986a isolated *Fussarium spp* and *Neurospora spp* and noted high levels of aflatoxin in maize in Plateau State of Nigeria. However, despite an initial interest generated, such research work has not been extended to other areas to ascertain the ever-increasing economic importance of mycoflora and their toxin producton in Nigeria. Therefore, this work was designed to ascertain the presence and extent of growth of mycoflora in maize, beans, ground nuts and yam on retail outlets in parts of Kaduna and Nasarawa States of Nigeria.

Materials and Method

Except where otherwise stated, all chemicals and solvent used in this work were of analytical grade as supplied by BDH Chemicals Ltd, Poole, England.

Media/ReagentsSterile Normal Saline: Nine grammes (9g) of NaCl were dissolved in a litre of distilled water and autoclaved at 121ºC for 15 minutes. This was allowed to cool on the bench and properly labeled. **Sabouraud's dextrose agar (SDA):** Sixty-five grammes (65g) of SDA were suspended in a litre of distilled water and brought to boil to dissolve completely. This was then sterilized by autoclaving at 121ºC for 15 minutes and 0.5g of chloramphenicol was aseptically added to the litre of the autoclaved cooled medium. About 20cm^3 were dispensed into Petri dishes. Also, 5cm^3 amount was dispensed into long test tubes plugged with cotton wool as slants. **Semi-synthetic medium:** Forty grammes (40g) of SDA were suspended in a litre of distilled water and prepared as stated above.

Area of Study

Kaduna and Nasarawa States, out of which six local government areas were selected. Kaduna: Jaba, Jamaa, Kaura, Lere, Sanga and Zango-Kataf. Nasarawa: Akwanga, Keffi, Lafia, Nasarawa-Eggon, Obi and Okona.

Sample Grains

Staple grains (local varieties) consisting of maize (*Zea mays*), beans (*Cowpeas-Vigna simensis*), groundnuts (*Arachis hypogaea*) and yam (*Diascorea spp*) were obtained from the selected six local government areas' retailed outlets in each of the States. About 2kg of each foodstuff were purchased from the selected six local government market outlets. Samples were done twice: Dry season, between the Months of September and February and at rainy season between March and August, from 1997 to 2005. All foodstuffs sampled were labeled, placed separately in a paper bag and kept under cool, dry conditions in order to prevent fungal growth or possible production of mycotoxin(s) (Jacobsen BJ *et al*;[23]).

Fungi Isolation

Method of Gabal MA *et al*;[24] in 1994 was adopted. About 10g of visibly mouldy grains or ground/ pulverized yam samples were placed in 250cm^3 conical glass flasks with screw caps each containing 100cm^3 sterile normal saline solution. The cap was screwed tight and later allowed to stand on the bench for 2mins. 1.0cm^3 of the supernatant was further diluted 1:10 in clean sterile long glass test-tubes in sterile saline. The tubes were similarly shaken. Then 0.5cm^3 each of the

diluents were aseptically inoculated onto the surface of freshly prepared SDA plates containing 0.5g chloramphenicol/l to suppress the growth of any contaminating bacteria. The inoculum was evenly spread on the agar surface in a Petri dish with sterile loop. Plates were then incubated at 20^{o}C incubator for 2-3 weeks.

Identification of Fungi (Mycoflora)
Fungi growth on plates were identified based on gross morphology and microscopy (25,26,27,28,) at the Federal College of Veterinary and Medical Laboratory Technology, National Veterinary Research Institute, Vom-Jos and at the Biotechnology Centre, Institute of Bioscience & Technology, Cranfield University, Silsoe Bedford, UK.

StatisticalAnalysis.

Statistical analysis was computed using SPSS (Statistical Package for Social Science) and Microsoft Excel Statistical Tool-Pack.

Results

The results of the mycoflora (fungal) isolates from the two States and in their individual grains are shown. Table 1 shows the mean of fungi isolated from the four foodstuffs during rainy season in the two States, while table 2 represents that of dry season. In rainy season, there was significant difference ($p<0.001$) in the mean of fungi labeled "a" (*Aspergillus flavus*) and those labeled "b" as well as those not labeled (the rest fungi isolates) in Kaduna and Nasarawa States (table 1). Also in dry season, there was significantly different ($p<0.001$) in the group of fungi labeled 'a" and those labeled "b" as well as those not labeled in both Kaduna and Nasarawa States (table 2). Equally in both Kaduna and Nasarawa States, there was significant difference ($p<0.05$) in the mean of fungi isolates in the four foodstuffs (table 3), with the mycoflora isolates in maize being the highest in the two States (Kaduna- 12.75 ±10.01; Nasarawa- 12.67 ± 6.14) compared to the rest of the grains during rainy season (table3) and Kaduna-13.29 ±9.75; Nasarawa-15.86 ± 8.02 at dry season (table 4). Rainy and dry seasons comparison of the mean fungi isolates from the four grains' samples shows that there was no significant difference ($p>0.05$) in fungi isolates amongst the grains in both Kaduna and Nasarawa States (table 5). During the rainy season in Kaduna State, *A. flavus* was the highest mycoflora isolate followed by *Fusarium spp., A fumigates, Fusarium moniliferme and A niger* in that order (table 1).

Table 1: Mean of mycoflora isolates from the grains during rainy season in Kaduna and Nasarawa states
Fungi isolates in numbers

State	KADUNA		NASARAWA	
1	**20.25^{a}**	**±9.95**	**19.50^{a}**	**±7.69**
2	**13.50^{b}**	**±5.32**	**14.30^{b}**	**±3.11**
3	**9.00^{b}**	**±4.54**	**9.00**	**±4.54**
5	**6.00^{b}**	**±0.00**	**7.50**	**±2.78**
6	**6.00^{b}**	**±0.00**	**6.75**	**±2.12**
7	**6.75^{b}**	**±2.12**	**6.75**	**±2.12**
8	**6.86^{b}**	**±2.27**	**6.75**	**±2.12**
10	**9.00^{b}**	**±1.13**	**6.75^{b}**	**±2.12**
11	**12.75**	**±3.85**	**12.75^{b}**	**±5.01**
12	**14.25**	**±5.50**	**11.25^{b}**	**±2.12**
13	**6.00^{b}**	**±0.00**	**6.00**	**±0.00**
14	**6.00^{b}**	**±0.00**	**6.00**	**±0.00**
15	**6.00^{b}**	**±0.00**	**6.00^{b}**	**±0.00**
16	**6.75**	**±2.12**	**14.25^{b}**	**±5.50**
20	**6.00**	**±0.00**	**6.00**	**±0.00**
21			**7.50**	**±3.00**
22			**6.00**	**±0.00**
23	**6.00^{b}**	**±0.00**	**8.00**	**±3.46**
F-Value		**8.53**		**9.10**
P-Value		**p < 0.001**		**p < 0.001**

Prevalence expressed as ẋ ± STD per State while each number represents the Fungal isolates from all four foodstuffs: Maize, Rice, Yam and Cassava. P – values are assessed horizontally.

1=*A. flavus*, 2=*A. fumigates*, 3=*A. niger*, 4=*A. nidulans*, 5= *Cephalosporium spp*., 6=*Chaetonium spp*., 7=*Cunninghamella spp*., 8=*Curvularia spp*., 9=*Drechslera rostrate*, 10=*Drechslera sativa*, 11=*Fusarium monilliforme*, 12=*Fusarium spp*., 13=*Helminthosporium spp.*, 14=*Neurospora spp*., 15=*Nigrospora spp.*, 16=*Penicillum fumiculosum*, 17=*Pnicillum spp*., 18=*Phoma sorghina*, 19=*Phoma spp*, 20=*Rhizoctonnia solani*, 21=*Rhizopus spp*., 22=*Syncephalastrum racemosum*, 23=*Trichoderma spp.*

Note: Similar superimposed letters indicate means that are not significantly different from each other.

P < 0.001= Very high significant difference.

While in dry season (table 2),also *A flavus* has the highest frequency followed by *Fusarium spp.*/ *Phoma spp*., Fusarium *moniliforme*, *A fumigates* and *A niger* in that order. Equally in Nasarawa State, *A. flavus* was the highest mycoflora isolate in wet season followed by *Penicillium funiculosum, A fumigates, Fusarium monilifoerme, Fusarium spp.*, and *A niger* in that oreder (table 1), while at dry season, also *A. flavus*, was the highest mycoflora isolate, followed by *Phoma sorghina, Fusarium spp., Phoma spp., A fumigates, Cephalosporium spp., and Fusarium moniliforme* in that order (table 2).

Table 2: Mean of mycoflora isolates from the grains during dry season in Kaduna and Nasarawa States
Fungi isolates in numbers

State	Kaduna		Nasarawa	
1	**19.50[a]**	**±10.01**	**19.50[a]**	**±10.01**
4	**10.50**	**±4.24**	**12.00[b]**	**±4.54**
5	**9.00**	**±4.54**	**10.50**	**±4.24**
9	**7.50**	**±2.78**	**7.50**	**±2.78**
11	**12.75**	**±6.76**	**9.75**	**±4.46**
12	**13.50[B]**	**±4.24**	**14.25[b]**	**±5.50**
13	**6.00**	**±0.00**	**6.75**	**±2.12**
14	**6.00[b]**	**±0.00**	**6.75**	**±2.12**
17	**6.75**	**±2.12**	**7.50**	**±2.78**
18	**8.25**	**±3.11**	**15.00a**	**±6.41**
19	**13.50[B]**	**±4.24**	**12.75[b]**	**±3.85**
20	**8.40**	**±3.29**	**6.75**	**±2.12**
21	**6.00**	**±0.00**	**6.00**	**±0.00**
22	**6.00**	**±0.00**	**6.00**	**±0.00**
F-Value		**5.93**		**5.74**
P-Value		**p < 0.001**		**p < 0.001**

Prevalence expressed as ẋ ± STD per State while each number represents the Fungal isolates from all four foodstuffs: Maize, Rice, Yam and Cassava. P – values are assessed horizontally.

1=*A. flavus*, 2=*A. fumigates*, 3=*A. niger*, 4=*A. nidulans*, 5= *Cephalosporium spp*., 6=*Chaetonium spp*., 7=*Cunninghamella spp*., 8=*Curvularia spp*., 9=*Drechslera rostrate*, 10=*Drechslera sativa*, 11=*Fusarium monilliforme*, 12=*Fusarium spp*., 13=*Helminthosporium spp.*, 14=*Neurospora spp*., 15=*Nigrospora spp.*, 16=*Penicillum fumiculosum*, 17=*Pnicillum spp*., 18=*Phoma sorghina*, 19=*Phoma spp*, 20=*Rhizoctonnia solani*, 21=*Rhizopus spp*., 22=*Syncephalastrum racemosum*, 23=*Trichoderma spp.*

Note: Similar superimposed letters indicate means that are not significantly different from each other.

P < 0.001= Very high significant difference.

In Kaduna State the grain that is most highly vulnerable to fungal infestation, during the rainy season was maize followed by beans, groundnut and yam while in dry season maize was also the highest followed by groundnut, beans and yam (tables 3&4).

Table 3: Mean of mycoflora isolates from individual grains during rainy season in the two states

States	Individual foodstuffs					
	Maize	**Beans**	**Groundnuts**	**Yam**	**F-value**	**P-value**
Kaduna	**12.75 ± 10.01[a]**	**10.00 ± 4.67[a]**	**9.00 ± 4.56[a]**	**6.46 ± 1.66[b]**	**0.90**	**p > 0.005**
Nasarawa	**12.67 ± 6.14[a]**	**8.63 ± 4.36[b]**	**10.33 ± 7.07[a]**	**7.85 ± 2.88[b]**	**1.99**	**p > 0.005**

Note:

Prevalence expressed as ẋ ± STD per foodstuff in Adamawa and Benue States

P – values are assessed horizontally.
Similar letters indicate means that are not significantly different.

P > 0.005 = Not significantly different.

Table 4: Mean of mycoflora isolates from individual grains during dry season in the two states

States	Individual foodstuffs					
	Maize	**Beans**	**Groundnuts**	**Yam**	**F-value**	**P-value**
Kaduna	**3.29 ± 9.75[a]**	**10.15 ± 4.51[a]**	**10.71 ± 5.85[a]**	**7.64 ± 2.80[b]**	**2.86**	**p < 0.005**
Nasarawa	**15.86 ± 8.02[a]**	**9.69 ± 3.90[b]**	**9.86 ± 6.49[b]**	**9.00 ± 4.05[b]**	**2.50**	**p < 0.005**

Note:Prevalence expressed as ẋ ± STD per foodstuff in Adamawa and Benue States
P – values are assessed horizontally.
Similar letters indicate means that are not significantly different.
P < 0.005 = Significantly different.
P > 0.005 = Not significantly different.

Table 5: Rainy and dry season comparison of the mean mycoflora isolates from the four grains in the two states.

Grains	Rainy	Dry	D, t – values	Df	P-value
Kaduna					
Maize	**12.75 ± 10.01**	**13.29 ± 9.75**	**-0.84862**	**20**	**p < 0.05**
Beans	**10.00 ± 4.67**	**10.15 ± 4.51**	**0.51389**	**19**	**p < 0.05**
Groundnuts	**9.00 ± 4.56**	**10.71 ± 5.85**	**0.18590**	**14**	**p < 0.05**
Yam	**6.46 ± 1.66**	**7.64 ± 2.80**	**0.32935**	**08**	**p < 0.05**
Nasarawa					
Maize	**12.67 ± 6.14**	**15.86 ± 8.02**	**0.44740**	**27**	**p < 0.05**
Beans	**8.63 ± 4.36**	**9.69 ± 3.90**	**0.11058**	**22**	**p < 0.05**
Groundnuts	**10.33 ± 7.07**	**9.86 ± 6.49**	**0.57132**	**19**	**p < 0.05**
Yam	**7.85 ± 2.88**	**9.00 ± 4.05**	**0.14050**	**13**	**p < 0.05**

Notes: P < 0.005 = Significantly different.
P > 0.005 = Not significantly different

In Nasarawa State, maize was also the most vulnerable grain that is sensitive to mycoflora growth closely followed by groundnut, beans and yam at wet season and equally maize, groundnut, beans and yam during dry season (tables 3 & 4). There were also seasonal variations in the growth of some mycoflora. While *A fumigatus* and *A niger* were note isolated during dry season, common mycoflora contaminants like *Rhizopus spp.* and *Syncephalastrum raecemosum* were not isolated in Kaduna State at wet season.

Discussion

Data on the occurrence of mycoflora in staple grains on retail outlets in Nigerian markets are scarce.Fromourfindings, the high growth of *A. flavus, Fusarium moniforme, Fusarium spp, and Penicellium fumiculosum as well as Phoma spp.* in rainy and dry seasons in Kaduna and Nasarawa States are in agreement with the report of Gbodi TA *et al;*[22] who in 1986a and 1986b[29], noted a very high growth of mycoflora in field and stored grains of maize and acha in Plateau State. Our findings also correlate with the work of Da-Silva JB *et al*;[15] who in 2000 in Brazil, observed high incidence of *A. flavus, Fusarium moniliforme and Fusarium spp.* in some agricultural grains. Also, our findings of high *A. flavus* is in line with the work of Gabal MA *et al*;[24] who in 1994 isolated a total of 103 *A. flavus* out of 150 agricultural commodities and animal feeds in Egypt. Blaney and Williams[30] who in 199 in Australia observed that high summer temperatures, irregular rainfalls and insect damage are probably important factors in allowing *Aspergillus flavus* and *A. parasiticus* to invade summer crops in the field. Hell, K *et al;*[31] in 2000b equally noted that insect is important in the spread of *A. flavus* in pre-harvest maize. From our findings, the same may be true particularly with a climate that varies from equatorial in the South, tropical in the Central region to arid in the far North of Nigeria. There were also seasonal variations in the growth of some mycoflora particularly in Kaduna State which also has the highest isolates of *Aspergillus flavus*. Equally our findings of high growth of *Fusarium monilifoerme* and *Fusarium spp.* are also in agreement with the assertion of Salifu[32] who in 1981 and Agboola[33] who in 1984 stated that *Fusarium spp.* are among the food spoilage moulds that commonly infect grains in Nigeria. Our findings that maize is perhaps more vulnerable to mycoflora growth is in line with the observation of Turner PC *et*

al;[34] who in 2000 stated that maize is frequently infected with *Aspergillus spp*. in West African and also in tune with the work of Mashinini and Dutton,[35] who in 2006 isolated *Fusarium spp.* in wheat and wheat based products in South Africa.

Conclusion

The presence of these toxigenic mycoflora in staple grains portends great danger when consumed by man and animals.

References

1. Christensen C.M. Molds, mushrooms and mycotoxins. University of Minnesota Press, Minneapolis. 1975: 264 pp.
2. Christensen C.M. Storage of cereal grains and their products. American Association of Cereal Chemists, Inc. St. Paul, Minnesota. 1982.
3. Meronuck, R.A. Mold in grain storage. St. Paul, Minnesota: University of Minnesota Extension Service Folder. 1987: AG-FO-0564.
4. Hasseltine, C.W. and Mehlman, M.E. Mycotoxins in human and animal health. Park Forest, South, Illinois. Pathotox. Publishers. 1977.
5. Hudler G. Magical and mischievous mold. Princeton University Press .1998: pp 248.
6. Lawlor, P.G. and Lynch, P.B. Mycotoxins in pig feeds. 1: Source of toxins, prevention and management of mycotoxicosis, *Irish Vet. Journal.* 2001: 54(3):117-120.
7. Lawlor, P.G. and Lynch, P.B. Mycotoxin management. African farming. Jan/Feb. ed. Reprinted with permission from *Irish Vet. Journal*. 2005.
8. Penn R. Mold: Volatile organic compounds and mycotoxins: a primer for home-owners. www.envirochex.com 2004 p.1-7 visited 27/11/04.
9. Hayes A.W. Mycotoxins: A review of biological effects and their role in human diseases. *Clinical Toxicology* 1980: 17:45-83.
10. Acharya P.B., Mohanti C.R. and Gangopadhyay S. Production of aflatoxin B_1 in rice and rice products. Indian Phythopathol. 1994: 37(4): 611-616.
11. Hunter B.T. Mycotoxins: A significant public health problem, 2nd edn. Pp 316-331. Mycoloque Publications. Water-Lo Ontario. 1989.
12. Matassian M.K. Poisons of the past: Mould, Epidemic and History. Yale University Press, New Haven. 1989.
13. Kendrick B. Mycotoxins in food and feed in the fifth kingdom, 2nd ed 1992: 316-331. Mycologue Publications. Waterloo Ontario.
14. Gonzalo J.D. and Heaman J.B. Fumonisin toxicoses in domestic animal. A review. Vet Human Toxicol. 1994: 36(6):548-555.
15. Da-Silva J.B., Pozi C.R., Mallozzi M.A., Ortega E.M. and Correa B. Mycoflora and the occurrence of AFB_1 and fumonisin B_1 during storage of Brazillian sorghum. J Agric Food Chem. 2000: 48(9):4352-4356.
16. Enrique B. Mycotoxin contamination of animal and human foods.Ann Pharm-Fr. 2000: 56(6suppl):464-469.
17. Tucker, J.B. Mycotoxins and Gulf war illness: A possible link. 2001. Available at http://wwwNgwrc Site visited 24/07/05 and 09/12/2016.
18. EMAN. European Mycotoxin Awareness Network. European Commission(EC) Co-ordinated by Leatherhead Food International UK. 2004: Website http:www.my-cotoxins.org.com Visited 11/27/2004, 04/07/2006 and 09/12/2016.
19. WHO. Evaluation of certain mycotoxins in food. World Health Organization Technical Report Series 906. 2002.
20. Christensen, C.N. and Kaufmann, H.H. Grain storage: The role of fungi in quality loss. Mnneapolis, Minnesota: The University of Minnesota Press. 1969.
21. WHO. Human exposure assessment: WHO Envronmental Heajth Criteria No. 214. 2000.
22. Ggbodi T.A., Nwude N., Aliu T.O. and Ikedi C.O. The mycoflora and some mycotoxins found in maize (Zee mays) in the Plateau State of Nigeria. *Vet Human Toxicol.* 1986a: 28(1):1-5.
23. Jacobsen B.J., Bowen K.L., Shelby R.A., Diener U.K., Kemppainen B.W. and Floyd J. Mycotoxins and mycotoxicoses. Alabama A & B and Auburn University. c/o William S. Gazaway, Extention Plant Pathologist, Auburn University, Alabama. 2004. Printed from the drop down menu "file" in IT 27/11/2004 and 09/12/2016.
24. Gabal M.A., Hagazi S.A. and Hassanin Nagwa. Aflatoxin production by *Aspergillus flavus* field isolates . Vet Human Toxicol. 1994: 36(6):519-521.
25. Thom C.and Raper K.B. Manual of Aspergilli. Willians and Wilkins Co., Baltimore MD.1945:263.
26. Sigurd F. Practical mycology for the identification of fungi. Halfner Publ. Co. Inc., New York. 1968 pp 31.
27. Abey S.D. Foundation in Medical mycology. Jeann D' Arc Library, Jeanne D' Arc Street, Hamra, Beirut. Lebanon. 1995.
28. Larone D.H. Medically important fungi: A guid to identification 3rd ed. 1995. American Society for Microbiology Press, 1325 Massachusetts Ave., N.W., Washington, DC.
29. Ggbodi T.A., Nwude N., Aliu T.O. and Ikedi C.O. The mycoflora and some mycotoxins found in acha (Digitaria exils stapl). Food and Chemical Toxicol. 1986b: 24:339-342.
30. Blaney B.J. and Williams K.C. Effective use in livestock feeds of mouldy and weather-damaged grain containing mycotoxins: Case histories and economic assessments pertaining to pig and poultry industries of Queensland. Australian Journal of Agricultural Research. 1991: 42: 993-1012.
31. Hell k., Cardwell K.F., Satamou M. and Schulthess F. Influence of insect infestation on aflatoxin contamination of stored maize in four agroecological regions in Benin, West Africa.Afr. Entomol. 2000b: 6:169-177.
32. Salifu A. Micotoxins in short season sorghum in Northern Nigeria. Samaru Journal of Agricultural Research. 1981: 1:83-87.
33. Agboola S.D. Research for effective storage in Nigeria. Edtted by CO Ikediobi (Zaria: Biochemical Society of Nigeria) 1994: pp 38-62.
34. Turner P.C., Mendy M., White H., Fortun M., Hall A.J. and Wild C.P. Hepatitis B infection and aflatoxin biomarker levels in Gambian children. *Trop. Med. Internal Health.* 2000: 5:837-841. (Pubmed).
35. Mashinini K. and Dutton M.F. The incidence of fungi and mycotoxins in South Africa wheat and wheat-based products. *J. Environ Sci. Health B.*2006: 41(3):285-296.