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Evaluation of Fungal Contamination on Livestock Feed Ingredients of Plants origin Stored for Five Years

Guluwa, L.Y.[1]*, Agada, G. O. A[2], Wumnokol, D. P.[1], Dalokom, D. Y.[4], Agbu, C. S.[5], Dastu, A. J.[3]

[1]Department of Animal Production Technology, College of Agriculture, Garkawa, Plateau State, Nigeria

[2]National Veterinary Research Institute Vom, Plateau State, Nigeria

[3]Department of Animal Health Technology, College of Agriculture, Garkawa, Plateau State, Nigeria

[4]Department of Crop Production Technology, College of Agriculture, Garkawa, Plateau State, Nigeria

[5]Ministry of Agriculture, Nasarawa State P.M.B. 056, Lafia, Nigeria

ABSTRACT: The purpose of this experiment was to evaluate the extent of Fungal contamination of livestock feed stuffs of plants origin stored for five years at Plateau State College of Agriculture, Garkawa. The feed ingredients were: wheat offals, cowpea husks, cotton seeds, kidney beans seeds, baobab seeds, bambara nut husks, palm kernel cake, baobab seed meal, rice offals, yam peels, ground nut cake and sweet orange peels. Microbiological quality of these feedstuffs were conducted using standard microbiological technique for food and animal feeding stuffs at Microbiology Unit of Central Diagnostic Laboratory at National Veterinary Research Institute Vom, Plateau State, Nigeria. The study revealed a total of eight fungi spp: *Aspergillus niger*, *Aspergillus fumigatus*, *Penicillum* spp, *Rhizopus* spp, *Mucor* spp, *Trichophyton rubrum*, *Aspergillus* spp and *Aspergillus flavus*. From the findings, cowpea husks, cotton seed, kidney beans seed, baobab seed and bambara nut husks were contaminated by *Aspergillus niger* (28.57%) as the dominant species of fungi followed by: *Aspergillus fumigatus* (19.05%) (baobab seed, bambara nut husks, blood meal, and baobab seed meal) and baobab seed (*Aspergillus niger*, *Aspergillus fumigatus* and *Penicillium* spp). Wheat offals and palm kernel cake had the highest total fungi counts of 4.0 x 10^2 cfu/g followed by yam peels, sweet orange peel and bone meal with 3.0 x 10^2 cfu/g and baobab seed meal had 2.2 x 10^2 cfu/g. To reduce the growth of mould on livestock feed ingredients stored over long periods of time, regular monitoring of moisture levels of these ingredients should be checked before embarking on storage.

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KEYWORDS: Feed ingredients, livestock, fungi spp, and colony forming units

Corresponding Author: Guluwa, L.Y.

INTRODUCTION

The major sources of rural livelihoods in Nigeria is crop and livestock production which contributes significantly to poverty alleviation by providing employment opportunities, food and other economic resources to the less privileged. Livestock's feed ingredients derived from plant and animal origins are threatened by feed contamination by fungi and their toxic metabolites. They are prone to mold when improperly stored and may accumulate mycotoxins (Ezekiel *et al*., 2012).It has been reported in literature that feed ingredient contamination with toxins may occur within feed ingredientssupply chain, starting from the field, during harvest, storage, processing and feeding channels in the farm. The organisms thrive well when the environmental temperature, humidity and pH favore their survival (Chukwuka, *et al.,* 2010; Zain, 2011).

Fungi genera of toxigenic of most concern worldwide to poultry feeds, general in agriculture and health in the tropics are *Aspergillus*, *Fusarium* and *Penicillium* (Kpado and Bankole, 2005; Ghaemmaghami*et al.,* 2018) also *Aspergillus* has been reported as predominant genus in dairy and feeds production (Dhand, *et al.,* 1998). Hossain *et al*. (2011) and Njobeh *et al.* (2012) in their studies reported that mycotoxins are known for intimidating the safety and security of livestock and human beings. Mycotoxicoses, have been noted to reduce animal productivity like body weight gain, litter sizes, deformed offspring, egg production and significant immune suppression (Streit *et al*. 2013) and economics losses incurred by the feedstuff producer (Chukwuka, *et al.,* 2010).

The objective of the study is to determine fungal species percentage occurrence, total colony forming units of fungi, and to recommend strategies for the control of mycotoxins.

MATERIALS AND METHODS

Sample Collections and Location

One kg of livestock feed ingredients stored for five years in the feed mill unit of Plateau State College of Agriculture, Garkawa, for students' practical were collected. The following samples: wheat offals, cowpea husks, cotton seeds, kidney beans seeds, baobab seeds, bambara nut husks, palm kernel cake, baobab seed meal, rice offals, yam peels, ground nut cake and sweet orange peels were thoroughly mixed to give a representative samples before collection in a sterile polyethylene bag for fungi metabolite analysis. As at the time of the experiment the feed ingredients were stored in a covered plastics containers in a cool, dry, well ventilated store to prevent fungal and bacterial growth and to preserve quality and nutritional value. However, the storage condition was not varied.

Fungi Enumeration and Isolation

The feed ingredients were soaked in distilled water for 24 hours. Thereafter, fungi were isolated by serial dilution and spread plate method. Samples of different feed ingredients were serially diluted and 0.1 ml (dilution 10-3) of samples was inoculated on potato dextrose agar (PDA) (Oxoid, UK). The inoculated plates were incubated at 28 ±2 ^{o}C for 3-5 days. Pure cultures were obtained by sub-culturing onto PDA and incubated at 28 ±2 ^{o}C. Microscopic characterizations were used for fungi identification. A drop of lactophenol cotton blue stain was dropped on a clean glass slide. A small tuft of the fungus colony was transferred into the drop, using a sterilized cooled needle. The fungus was teased out and a cover-glass was placed over the preparation. The slide was examined under x 10 and x 40 objective of the microscope.

Percentage occurrence and fungal colony

Percentage occurrence or frequency = count of similar fungi specie isolated in all the feedstuff divided total number of different count of fungi species multiply 100. Fungal colonies were counted and recorded as colony forming units per gram (CFU/g).

RESULTS AND DISCUSSION

Table 1: Percentage occurrences of fungal isolates from stored feed ingredients of plants origin

Fungi isolates	Frequency	Percentage
Aspergillus niger	6	33.33
Aspergillus fumigates	3	16.67
Penicillium spp	2	11.11
Rhizopus spp	3	16.67
Mucor spp	2	11.11
Trichophyton rubrum	1	5.56
Aspergillus flavus	1	5.56
Total number of isolates	21	100.00

Presented in Table 1 is percentage occurrence of fungal isolates from feed ingredients stored over five years at Plateau State College of Agriculture, Garkawa for students' practical. Appearance of fungal isolate on each ingredient is as presented in Table 2. Percentage fungi isolates varies from 33.33% for *Aspergillus niger*, 16.67% for *Aspergillus fumigates* and *Rhizopus* spp each, 11.11% for *Penicillium* spp and *Mucor* spp each, while 5.56% for *Trichophyton rubrum*, and *Aspergillus flavus*, respectively. *Aspergillus niger* dominated followed by *Aspergillus fumigates* and *Rhizopus* spp, Penicillium *spp and Mucor* spp as well as *Trichophyton rubrum* and *aspergillus flavus* with the least percentage occurence. Contaminations by *Aspergillus species* could be responsible for the production of aflatoxins. This means that livestock feed formulated with *aflatoxin* contaminated feed ingredients might likely cause liver and brain damage even though aflatoxin susceptibility varies between species because they are made of different toxic compounds (Baldissera *et al*., 2018). Therefore, contamination of feedstuffs by moulds may lead to reduction of nutritional value, feed quality, and leaving behind poisonous mycotoxin.

Table 2: Fungal total count and number isolates obtained from feed ingredients of plants origin

Feed Ingredients	TFC (cfu/g)	Fungal Isolates
Cowpea husk	1.2 x 10^2	*Trichophyton rubrum, Aspergillus niger*
Yam peel	3 x 10^2	*Rhizopus* species
Cotton seed	2 x 10^2	*Aspergillus niger*
Wheat offal	4 x 10^2	*Rhizopus* species
Kidney beans seed	1.4 x 10^2	*Aspergillus niger*
Rice offal	2.1 x 10^2	*Mucor species, Aspergillus flavus,*

Baobab seed	2 x 10^2	*A. niger, A. fumigatus, Penicillium* species
Bambara nut husk	1.8 x 10^2	*Aspergillus niger, Aspergillus fumigates*
Sweet orange peel	3 x 10^2	*Mucor* species
Baobab seed meal	2.2 x 10^2	*Aspergillus fumigates*
Palm kernel cake	4 x 10^2	*Aspergillus niger, Penicillium* species
Ground nut cake	1.1 x 10^2	*Rhizopus* species

TFC = total fungi count, cfu/g = colony forming units per gram

Total fungal count is presented in Table 2. Total fungi count is very useful in detecting and stating the quantity of fungal growth on the test materials and allows for identification of fungi species. Highest means of total fungal counts of 4.0 x 10^2 cfu/g were observed in wheat offals and palm kernel cake, followed by 3.0 x 10^2 cfu/g in yam peels and sweet orange peel, 2.2 x 10^2 cfu/g was seen in baobab seed meal and less than 2.1 x 10^2 cfu/g is as presented in Table 2.

CONCLUSION AND RECOMMENDATIONS

Fungi genera observed in this study were *Aspergillus* spp, *Penicillium* spp, *Rhizopus* spp, *Mucor* spp and *Trichophyton rubrum.* The presence of microscopic fungi on livestock feedstuffs may affects their organoleptic attributes, feed quality and leaving behind poisonous mycotoxin which will later contaminate finished feeds. However, continuous growth of mould on these feedstuffs may result to spoilage leading to loss of nutrients and if not control could synthesize different mycotoxins. If not control, human being as consumers of animals' products like meats and eggs may threaten human welfare. But, *Penicilium* isolated in this present study may appear to be non-toxigenic. Promoting feed quality will helps in increasing farmers' income through better animal's trade. Proper drying of feedstuff and deliberate efforts to avoid ingestion of aflatoxins contamination feedstuffs % is very critical in reducing the growth of mould and prevention of harmful effect on the end users.

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