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Preliminary report of Dehulling effect on the occurrence and distribution of *Aspergillus flavus* in maize grains stored in Mubi market

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ABSTRACT

The effect of dehulling on the occurrence and distribution of Aspergillus flavus in some stored maize grains in Mubi grains market has been carried out using direct plating technique. All the undehulled samples yielded the presence of Aspergillus flavus, while none was detected in the dehulled samples that were immediately cultured. The dehulled samples that were stored over one month revealed Aspergillus flavus in five samples out of ten, while none was detected in the remaining five samples. The results indicated that dehulling eliminates the fungal (A. flavus) infestation of maize grains and that poor storage condition might have favoured the growth of Aspergillus flavus that were recovered in some stored dehulled maize samples.

Key words: *Aspergillus flavus, Dehulled , undehulled maize, Mubi, Nigeria.*

INTRODUCTION

Fungi particularly the moulds are well known with their ability of growing on and infesting crops both in the field (pre-harvest) and during storage (post-harvest). One of the most important moulds concerned with the infestation of maize is *Aspergillus flavus*.

Aspergillus flavus is considered extremely important with regards to aflatoxin production. The fungus can germinate at lower moisture level of 15-17% but infection and growth require higher moisture [5]. *Aspergillus flavus* is common on cereal grains [2] and it is frequently isolated from maize [7].

The implication of aflatoxins in a variety of human and animal sicknesses and their effects on agricultural productivity have inspired scientists to investigate further on *Aspergillus flavus* and the toxins it produces (aflatoxins). *Aspergillus flavus* has been studied both in the field and in Cultures by various workers in the world and the production and activity of aflatoxin has been assessed [8].

In addition to the production of dangerous Mycotoxins (aflatoxins), which are a group of highly toxic and carcinogenic metabolites, *Aspergilus flavus* can also behave as a true pathogen [1].

The aim of this research work is to investigate the presence of *Aspergillus flavus* in stored maize samples and to recommend the possible ways of controlling contamination and infestation.

MATERIALS AND METHODS

Thirty samples of white maize were obtained from Mubi grains market in Adamawa State of Nigeria using a slightly modified method of Jone, 1972. The samples were labeled A-C, each sample was divided into 1- 10. That is sample A, was divided A_1-A_{10}, B into B_1-B_{10} and C, into C_1- C_{10}. The sample subscript number is designating a sample number. A is undehulled maize samples, B- is dehulled maize samples and C- is dehulled maize samples but stored for over one month. Dehulling refers to the traditional method of processing maize sample which involves the removal of the pericarp and germ, which could imply removal of fungi and the associated Mycotoxins [4].

Isolation of *Aspergillus flavus*

The isolation was carried out according to the method described by [6]. The fungus was isolated using plating technique. Sabroud dextrose agar (Biotec) was used as the growth medium. The agar medium was amended with twenty five parts per million (25 PPM) each of Chloramphenicol and streptomycin to minimize bacterial contamination. In each plate, one seed was placed in the center of the plate, and one seed in each quadant, which means five seeds per plate. The plates were incubated at room Temperature 22-25°C. Control plates were also prepared with the same growth medium (Sabroud dextrose agar). These were incubated together with the seeded plates.

RESULTS

Mould Isolation

The cultured maize samples yielded a mixed growth of various fungal species including *Aspergillus flavus*. The *Aspergillus flavus* was identified by its bright to dark green velvety colonies on the growth media, and sometimes it display yellow-green and goldish to red brown colony characteristics on surface and reverse pigmentations respectively. All the plates, in which undehulled white maize samples were cultured yielded colonies of *Aspergillus flavus* among others.

In some plates, the growth some of the plates the colonies of Aspergillusflavus were outgrown by colonies of spreader moulds such as mucor.

Furthermore, all the plates in which dehulled maize samples were cultured immediately yielded no *Aspergillus flavus* but some other fungi species like Mucor. Some of the dehulled samples that were stored for over one month yielded *Aspergillus flavus* in culture while others did not. The control plates yielded no growth.

Diagrams A & B (plate A & B).

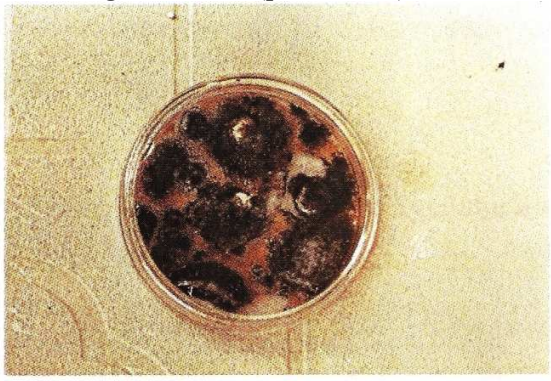

Plate A-Containing mixed culture of *Aspergillus flavus* (Six Days).

Plate B-Containing pure culture of *Aspergillus flavus* (Six Days).

The subculture from *Aspergillus flavus* colonies yielded a heavy growth of the mould with some unwanted species but later the *Aspergillus flavus* was isolated in pure culture by further sub-culturing on sabraud dextrose agar (plate A & B).

Table 1: Recovering of *A. flavus* in Undehulled maize sample

Sample	*A. flavus*	Nature of Growth
A1	I	+++
A2	I	+++
A3	I	+++
A4	I	+++
A5	I	+++
A6	I	+++
A7	I	+++
A8	I	+++
A9	I	+++
A10	I	+++

Table 2: Dehulled maize samples that were immediately Cultured

B1	N
B2	N
B3	N
B4	N
B5	N
B6	N
B7	N
B8	N
B9	N
B10	N

Table 3: Recovering of *A. flavus* in the dehulled maize samples that were stored over one month

Sample	*A. flavus*	Nature of Growth
B1	I	-
B2	I	-
B3	I	+
B4	I	+
B5	I	-
B6	I	++
B7	I	-
B8	I	-
B9	I	++
B10	I	+

A=Undehulled Samples
Subscripts No. = Indicates the sample No.
B=Dehulled Samples
C=Dehulled Samples that has been stored for over one month
N=Not isolated
I= Isolated
+++ =Heavy Growth
++ = Moderate growth
+ = Scanty growth

DISCUSSION

Reports have shown that it is possible to isolate *Aspergillus flavus* in pure culture from samples of maize which have been in storage for over five months [4,8]. This work equally has confirmed such findings.

Table 1 above, showed all ten samples (Samples labeled A_1-A_{10}) yielded *Aspergillus flavus* in culture. This is in agreement with work of [8] in which 100% of the ten samples of maize that has been in storage for over five months yielded *Aspergillus flavus* in culture. This indicates that the mould might have infested the maize either in the field or during storage. This is because the *A. flavus* outbreak can occur in the filed during preharvest or on crops during storage at substrate moisture content of 14% and a temperature of 24°C-40^{o}C.

Aspergillus flavus is considered imperative with regards to aflatoxins production, and these mycotoxins are highly carcinogenic and toxic and therefore, presence of *A. flavus* can endanger

the health of maize consumers particularly in Africa continent where the presence of Mycotoxin' s in food is often overlooked due to public ignorance about their existence, lack of regulatory mechanisms, dumping of food products and the introduction of contaminated commodities into human food chain during the chronic food shortage due to draught, war, political and economic instability. The largest Mycotoxins poisoning epidemic in the last decade occurred in Kenya in the year 2004 (as reported by CDC, 2004) as a result of consumption of maize contaminated with Mycotoxins.

Table 2: above, showed significant effect of dehulling on the occurrence of *A. flavus* in maize. This is because all the ten samples (labeled B_1 B_{10}) have shown no growth of *A. flavus*. This results is in conformity with the work of [8], where the ten dehulled samples cultured yielded no *Aspergillus flavus*. This result is also in agreement with the work of [4] who reported that semi-processed maize grains had lower isolation frequency of *A. flavus*.

Table 3: Showed the result of dehulled maize samples that has been in storage for over one month yielded *A. Flavus.* This might be as a result of poor storage condition because this work was conducted during the raining season and the storage conditions which include moisture and low temperature might have favorably supported the growth of the fungi. Out of the ten samples cultured five (50%) showed presence of *A. Flavus* while the remaining five (50%) have yielded no *A. flavus* in culture.

CONCLUSION

In conclusion, since *Aspergillus flavus* has great impact on human and animal life. It is therefore, imperative to take measures of curtailing or preventing its infestation and contamination (Mycotoxins). There is also need to enlighten the grains marketers to improve their storage facilities. Hence, the following recommendations are suggested:

1. Early detection of grain mould development should be observed (change in colour of the seeds) by monitoring regularly the storage facilities.
2. Grain moisture content should be drastically reduced to less than 15% within 48 hours of harvest by adequately drying the maize after harvesting before storage.

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