PREVALENCE OF FUNGI AND TOXIGENICITY OF *A. FLAVUS* AND *A. OCHRACEUS* ISOLATES RECOVERED FROM FEEDS AND THEIR CONTROL

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Abstract

Two hundred samples of single feeds (yellow corn, soyabean, wheat, hay and tibin) and compound feeds (poultry ration, processed animal feeds, broiler concentrates, layers concentrates and meat and bone meal); 20 samples from each, were screened for fungal contamination. The most predominant genus was *Aspergillus* which was isolated from all samples. Other moulds were isolated, but in low frequency as *Penicillium sp., Fusarium* sp., *Cladosporium* sp. and *Alternaria sp.*The isolated *A. flavus* and *A. ochraceus* were tested for production of aflatoxin B1 and ochratoxin A, respectively. The results of aflatoxin B1 production by *A. flavus* isolated from different feeds revealed that the higher incidence of toxigenic *A. flavus* was recorded from layers concentrates (50%), followed by broilers concentrates and hay(40%), whereas, *A. flavus* isolated from soyabean and poultry ration samples represented low incidence (12.5% and 10%), respectively. The results of ochratoxin A production by A. *ochraceus* isolated from different feeds revealed that the higher incidence of toxigenic *A. ochraceus* was reported from poultry ration (100%), followed by processed animal feeds, broilers concentrates and yellow corn (50%), whereas, the incidence of toxigenic *A. ochraceus* isolated from hay and layers concentrate samples showed low incidence (25%) for both. Evaluation of 2 commercial antifungals by plate assay revealed that MIC of Muv-Anti mould and mycostatin was 0.75 and 1.0 μg/ml for all fungi isolated from feeds. Following the addition of Muv- Anti Mould to mouldy poultry rations, the fungal population was decreased after 2 days of treatment, and completely inactivated after 1 week. The antimycotoxin (Sat. F. Dray) eliminated aflatoxin B1 and ochratoxin A, when added to contaminated feeds at a concentration between 2 and 3%.

INTRODUCTION

Up-to-date, the dramatical increase in population in the world requires an efficient modern animal production industry and the manufacture of good quality feeds and food. Hence, great attention has been paid to the increased importance of fungi and their mycotoxins, which are serious fungal metabolites for animal productivity.

Mycotoxins are formed by certain fungal species, whenever environmental factors are conductive during the growth of these frequently occurring mycomycetes on foodstuffs and animal feeds; the process takes place as a secondary metabolism. These natural toxins have broad spectrum effects (Hassan *et al.,* 2002, 2003 and 2004). When foodstuffs for human consumption are preserved or prepared by the conventional heating process, the mycotoxins are capable of surviving undamaged, thus, reaching the eventual consumer. These fungi and mycotoxins have serious effects upon the growth rate and health of human being and animals, as some mycotoxins had been found to have carcinogenic, tremorgenic, haemorrhagic, dermatitides and hormonal effects (Wray, 1981, Hassan, 1998, and Hassan *et al.,* 2005).

The aim of the present work was to screen single and compound feeds for fungal contamination, evaluation of the isolated *Aspergillus flavus* and *Aspergillus ochraceus* for production of respective mycotoxins and testing of some commercial fungal inhibitors and antimycotoxin for prevention and control of fungal growth and degradation of mycotoxins.

MATERIALS AND METHODS

Feed samples

Two hundred samples of feedstuffs (20 of each of yellow corn, wheat, soyabean, hay, tibin, layers concentrates, meat and bone meals, poultry ration, broilers concentrates and processed animal feeds were collected for investigation of fungal contamination and detection of aflatoxin and ochratoxin contamination.

Standards of aflatoxins and ochratoxin A

Standards of aflatoxins B1, B2, G1 and G2 and ochratoxin A were purchased from Sigma Chemical Company (USA).

Mould and toxin inhibitors

Mycostatin (Delta Pharma), Muv-Anti mould (Medical Company for Veterinary Products and Feeds), Sat-F-Dray (HSACS) compound (Sat Farma Vet).

Thin layer chromatographic apparatus: (Col Parmer Instrument Company, 981 series, Chicago 11160648 USA).

Isolation and identification of moulds

Each sample of feeds was subjected to isolation and identification of fungi as described by Refai (1979).

Productions and estimation of mycotoxins

The production and estimation of aflatoxins were carried out according to Gabal *et al.* (1994) and those of ochratoxin A as described by (Davis *et al.,* 1969).

Control of fungal growth and detoxification of feeds

The commercial feed samples were subjected to fungal count colony, then, were treated by addition of 0.2 g of Muv- Anti mould /100 g *of* sample (Chen and Day, 1974). After thorough mixing, the samples were incubated at 25°C and examined after 2 and 7 days for fungal growth. These tests were repeated 3 times.

The detoxification of feeds (experimentally contaminated with aflatoxin B1 and ochratoxin A) by commercial Sat-F-Dray (HSCAS) compound was done as described by Harvey *et al.* (1991), where 25 g of each of feed in 100 ml capacity flasks were mixed with 50 ppb of aflatoxin B1 or ochratoxin A. Sat-F-dray (HSCAS) was added to feed samples at different concentrations (0, 0.25, 0.5, 1, 2, 3, 4, 5, 6, and 8%).The flasks containing the treated feed were left for 7 days, then, the mycotoxins in feed were measured.

Statistical analysis

The obtained data were computerized and analyzed for significance, calculation of standard error and variance according to (Selvin, 1996 and SPSS 11, 2001).

RESULTS

Prevalence of fungi in single feeds

The present study showed that, the genus *Aspergillus* was isolated from all samples of single feeds (100%), while, 90% of yellow corn and hay samples were positive for *Rhizopus* spp. and *Penicillium* spp., respectively. *Fusarium* spp. were most frequent in hay (60%), followed by yellow corn (50%), tibin (40%) and wheat (20%) but, were not found in soyabean (Table 1).

Prevalençe of members of *Aspergillus, Fusarium* and *Penicillium* species in single feeds

Table 2 demonstrates the prevalence of *Aspergillus, Penicillium* and *Fusarium* species isolated from single feeds, where 317 isolates were identified into species. *Aspergillus* isolates (225) were the most common, followed by *Penicillium* (68) and Fusarium (24).

Aspergillus niger was the most common (86 isolates) and was found in all types of feeds with the highest rate of isolation (100%) from hay and tibin, followed by soyabean (90%), yellow corn and wheat (70%).The second most common *Aspergillus* species was *A.flavus* (64 isolates), which was recovered from all hay samples (100%), followed by soyabean and tibin samples (80%), then, yellow corn (60%), it was not found in wheat samples.

The remaining *Aspergillus* species were isolated at lower rates, namely: *A. candidus* (25 isolates), *A.fumigatus* (21), *A.terreus* (18), *A. ochraceus* (13), and

A.parasiticus (4). The (68) *Penicillium* isolates were identified into (8) species, namely: *P. thomii* (30 isolates), followed by *P. chrysogenum* (11), *P. digitatum* (7), as well as *P. viridicatum* and *P. funiculosum* (6 isolates each), *P. oryzoe* (5), *P.sclerotigenum,* while, *P. verrucosum* was isolated only once from a sample of soyabean. Twenty – one isolates of *Fusarium* species were identified as *F. solani* (8), *F. tabacium* (5), *F. violacium* and *F. oxisporium* (each 4) and *F. tricinctum* (3). *Fusarium* species were not found in soyabean.

Prevalence of fungi in compound feeds

It is clear from Table 3 that, fungi isolated from compound feeds were identified in 9 genera. *Aspergillus* and *Penicillium* species were recovered from all types of feeds, where *Aspergillus* species were found in 94 samples. The rate of isolation of *Aspergillus* species was (100%) in poultry ration and broilers concentrates, and it was (90%) in the other feeds. *Penicillium* species were recovered from 68 samples with the highest rate (80%) in layers concentrates, followed by poultry ration, processed animal feeds and broilers concentrates (70%), and lastly bone and meat meals (50%).

Prevalence of members of *Aspergillus, Fusarium* and *Penicillium* species in compound feeds

As demonstrated in Table 4, *Aspergillus* species were the most commonly isolated from poultry ration, followed by broilers concentrates, layers concentrates, bone and meat meals and processed animal feeds. The 203 isolates could be identified into 8 species, with *A. flavus* at the top of the list, followed by *A. niger, A. terreus, A. candidus, A. ochraceus, A. fumigatus, A. parasiticus* and *A. glaucus*. *Penicillium* isolates were recovered from 68 samples and could be identified into 8 species, where, *P. digitatum* was the most common, followed by *P. chrysogenum, P. thomii, P. viridicatum, P. restrictum, P. citreoviride* and *P. purpurogenum*. *Fusarium* isolates were isolated mainly from poultry ration. They were identified as *F. solani, F. oxysporium, F. tricinctum* and *F. moniliform.*

Productions and estimation of aflatoxin B1 and ochratoxin A by *A. flavus* and *A. ochraceus* isolated from feeds

The results of aflatoxin B1 production by *A. flavus* isolated from different feed samples revealed that the incidence of toxigenic *A. flavus* was variable; it was the highest in layers concentrates (50%), followed by broilers concentrates (40%) and hay (40%), while, it was 10% among *A. flavus* isolated from poultry ration (Table 5). The relations between the prevalence of *A. flavus* isolates in feeds, colony count and their toxigenicity were irregular. In some cases as in hay samples, the prevalence of *A. flavus* was (100%), colony count ($1.9 \times 10^1 \pm 0.05$), incidence of toxigenic isolates (40%) and maximum level of aflatoxin produced (2.3 ppm). On the other hand, *A.*

flavus isolated from poultry ration showed similar incidence (100%) and colony count ($1.9 \times 10^1 \pm 0.049$), but, the toxigenicity of isolates was (10%) and the produced level of aflatoxin was comparatively low (0.625).

The result of ochratoxin A production by *A. ochraceus* isolated from different feed samples revealed that the highest incidence of toxigenic *A. ochraceus* was recorded in poultry ration (100%), followed by processed animal feed (50%), broilers concentrates (50%) and yellow corn samples (50%), while, it was the lowest in hay and layers concentrate samples (Table 6).

The relations between the prevalence of *A. ochraceus* isolates in feeds, colony count and their toxigenicity were also irregular, where in some cases as in poultry ration, the results showed low prevalence of *A. ochraceus* (15%), colony count ($0.5 \times 10^1 \pm 0.028$), high incidence of toxigenic isolates (100%) and low level of ochratoxin produced (0.5 ppm). On the other hand, the isolated *A. ochraceus* from layers concentrates showed a low incidence (20%), colony count ($0.75 \times 10^1 \pm 0.029$) and low toxigenicity of isolates (25%), but, had a maximum level of ochratoxin produced (1.2 ppm).

Decontamination of feeds

In the present work, the ability of Muv-Anti mould and mycostatin (commercial compounds) to inhibit the growth of *A. flavus* and *A. ochraceus* and their toxin production was tested. The concentration of 0.75 µg/ml of Muv-Anti mould inhibited the growth of most but not all of *A. flavus*, whereas, the same concentration inhibited all tested isolates of *A. ochraceus*. However, the concentration of 1.0 µg/ml inhibited the growth of all *A. flavus* and *A. ochraceus* (Table 7). The concentration of 1.0 u/ml of mycostatin inhibited the growth of all *A. flavus*, whereas, the same concentration did not inhibit all tested isolates of *A. ochraceus*. However, the concentration of 2.0 µ/ml inhibited the growth of all *A. flavus* and *A. ochraceus* (Table 8).

When the Muv- Anti mould was added to mouldy poultry feeds, the colony count of *Aspergillus* was decreased from (5.2×10^1 to 1.2×10^1) and *Mucor* species from (4.8×10^1 to 2×10^1) after 2 days, however, the feeds became mycologically negative after 1 week of treatment (Table 9).

Detoxification of feeds contaminated with aflatoxin B1 or ochratoxin A

As shown in Table 10, Sat F Dray successfully eliminated aflatoxin B1 and ochratoxin A from feed samples. The levels of 50 ppb of aflatoxin B1 in feeds required 2% and 3% of Sat .F. Dray to be added to animal and poultry feeds, respectively. Ochratoxin A contamination required 2% of Sat.F.Dray in both animal and poultry feeds to degrade 94% and 84% of toxin contents in feeds. However, the concentration of 3% of Sat.F.Dray removed all ochratoxin A in feeds.

DISCUSSION

The present study showed that 12 genera of fungi were represented by various species in feeds, of which, the genus *Aspergillus* was the most predominant in samples of single feeds. Other genera were present in irregular frequency. These findings were in agreement with the results of El-Far *et al.* (1995), Refai *et al.* (1996), Hassan and El Sharnouby (1997), Hassan (1998), El-Hamaky *et al.* (2001) and Hassan *et al.* (2002), who recovered most of these fungi from single feed samples.

The predominance of *Penicillium* and *Fusarium* spp (Table 1) in hay and tibn samples may be due to the exposure of these feeds to different climatic conditions during preparation, particularly wet climate and irrigating water. It is known that, low temperature favours the growth of *Penicillium* and *Fusarium* spp., moreover, they are field fungi and more frequent in recently harvested cereals. This point was already discussed by Hassan and Omran (1996) and Hassan *et al.* (2004).

In single and compound feed samples, species of the genus *Aspergillus* were the predominant isolated fungi. These fungi require high temperature, high humidity and low oxygen, which may have resulted from processing machine favouring the growth of such fungi. On the other hand, they seemed to resist the aggressive heat treatment during processing of compound feeds, which probably destroyed or inhibited the growth of many fungi. This is evident in the present work, where the mean total colony counts of moulds in compound feeds were comparatively lower than those in single feeds. Such findings were observed also by El-Hamaky *et al.* (2000), Mogeda *et al.* (2002) and Hassan and Mogeda (2003).

In the present work, it was realized that, not all isolates of *A. flavus* produced aflatoxins, and there was no regular relation between the prevalence of fungi, colony count and their toxigenicity. This made the judgment of the safety of the tested feeds difficult if we depend only on mould count or on the mere isolation of *A. flavus.* The same applies to other mycotoxin-producing fungi. These findings were in agreement with the results of Adebajo *et al.* (1994) and Hassan *et al.* (2003).

The efficiency of Muv- Anti mould in inhibiting the growth of moulds in feeds indicated in the present study substantiates results obtained by Deyoe and Quardi (1970), Chen and Day (1974), Stewart *et al.* (1977), Chen *et al.* (1979), Sofos and Busta (1981) and Hassan (1994). However, in parallel to the use of mould inhibitors, the feed sample must be also free from mycotoxins. In the present work, Sat. F. Dray, which is composed of hydrated sodium calcium aluminosilicate (HSCAS) is known to bind with toxins, and consequently, inactivate them (Harvey *et al.*, 1991). It successfully eliminated aflatoxin B1 and ochratoxin A from feed samples. The levels of 50 ppb of aflatoxin B1 in feeds, required 2% and 3% of Sat.F.Dray to be added to animal and poultry feeds, respectively. Ochratoxin A contamination required 2% of Sat. F. Dray in both animal and poultry feeds to degrade 94% and 84% of toxin contents in feeds. However, the concentration of 3% of Sat.F.Dray removed all ochratoxin A in feeds. These results substantiate the findings of other authors (Barto, 1985, Abdel-Baset, 1989, Hassan and El Sharnouby, 1996 and Hassan, 2004).

In conclusion, frequent testing programs of feedstuffs during different steps of production must be monitored before use. The fungal inhibitors and antimycotoxins may be added if the level of contamination exceeds the permissible limit (5-25ppb). The treated feed must not be used before at least 1 week after treatment.

Table 1. Prevalence of fungi in single feeds.

Isolates	Yellow corn		Soya bean		Tibin		Hay		Wheat	
	No. +ve	%	No. +ve	%	No. +ve	%	No. +ve	%	No. +ve	%
Aspergillus spp.	20	100	20	100	20	100	20	100	20	100
Penicillium spp.	6	30	14	70	14	70	18	90	16	80
Fusarium spp.	10	50	–	–	8	40	12	60	4	20
Mucor spp.	10	50	4	20	–	–	4	20	4	20
Rhizopus spp.	18	90	4	20	–	–	–	–	8	40
Cladosporium spp.	4	20	4	20	4	20	14	70	4	20
Alternaria spp.	–	–	3	15	14	70	10	50	4	20
Curvularia spp.	–	–	–	–	–	–	4	20	–	–
Scopulariopsis spp.	–	–	4	20	–	–	–	–	–	–
Chaetomium spp.	4	20	–	–	–	–	–	–	–	–

Table 2. Prevalence of members of *Aspergillus, Fusarium* and *Penicillium* species in single feeds.

Isolates	Yellow corn		Soya bean		Wheat		Hay		Tibin		Total
	No	%	No	%	No	%	No	%	No	%	No
Aspergillus sp.	20	100	20	100	20	100	20	100	20	100	225
A. candidus	12	60	6	30	3	15	4	20	–	–	25
A. fumigatus	–	–	8	40	10	50	–	–	3	15	21
A. flavus	12	60	16	80	-	-	20	100	16	80	64
A. niger	14	70	18	90	14	70	20	100	20	100	80
A. ochraceus	3	15	–	–	–	–	4	20	6	30	13
A. parasiticus	–	–	–	–	–	–	4	20	–	–	4
A. terreus	6	30	8	40	–	–	4	20	–	–	18
Penicillium sp.	6	20	14	70	16	80	18	90	14	70	68
P. chrysogenum	2	33.3	2	14.2	5	31.3	–	–	2	14.3	11
P. digitatum	1	16.6	2	14.2	–	–	3	16.6	1	7.14	7
P. funicolosum	–	–	2	14.2	–	–	3	16.6	1	7.14	6
P. oryzae	–	–	1	7.14	3	18.8	–	–	1	7.14	5
P.sclerotigenum	–	–	–	–	2	12.5	–	–	–	–	2
P. thomii	2	33.3	5	35.7	6	37.5	9	50	8	57.1	30
P .viridicatum	1	16.6	1	7.14	-	-	3	16.6	1	7.14	6
P. vercossum	–	–	1	7.14	–	–	–	–	–	–	1
Fusarium sp.	10	50	–	–	4	20	12	60	8	40	24
F. oxysporium	–	–	–	–	–	–	4	33.3	–	–	4
F. solani	-	-	-	-	4	100	2	16.6	2	25	8
F. tabacinum	–	–	–	–	–	–	4	33.3	1	12.5	5
F. tricinctum	–	–	–	–	–	–	–	–	3	37.5	3
F. violacium	–	–	–	–	–	–	2	16.6	2	25	4

Table 3. Prevalence of fungi in compound feeds.

Isolates	meat and bone meals		Poultry ration		Processed animal feed		Layers concentrates		Broilers concentrates	
	No.	%	No.	%	No.	%	No.	%	No.	%
Aspergillus spp.	18	90	20	100	18	90	18	90	20	100
Penicillium spp.	10	50	14	70	14	70	16	80	14	70
Fusarium spp.	–	–	10	50	–	–	3	15	4	20
Mucor spp.	18	80	14	70	12	60	16	80	18	90
Rhizopus spp.	4	20	4	20	6	30	6	30	8	40
Cladosporium spp.	4	20	4	20	4	20	–	–	4	20
Scopulariopsis spp.	–	–	4	20	4	20	4	20	–	–

Table 4. Prevalence of members of *Aspergillus, Fusarium* and *Penicillium* species in compound feeds.

Isolates	Bone and meat meals		Poultry ration		Processed animal feed		Layers concentrates		Broilers concentrates		Total no. of isolates
	No.	%	No.	%	No	%	No.	%	No.	%	
Aspergillus sp.	18	90	20	100	18	90	18	90	20	100	203
A. candidus	-	-	6	30	8	40	4	20	4	20	22
A. flavus	12	60	20	100	8	40	12	60	16	80	68
A. fumigatus	4	20	4	10	-	-	-	-	-	-	8
A. glaucus	-	-	3	15	-	-	-	-	-	-	3
A. niger	12	60	10	50	6	30	14	70	14	70	56
A. parasiticus	–	–	–	–	4	20	–	–	–	–	4
A. ochraceus	-	-	3	15	4	20	4	20	3	15	14
A. terreus	8	40	8	40	4	20	4	20	4	20	28
Penicillium sp.	10	50	14	70	14	70	16	80	13	70	68
P. citreoviride	-	-	-	-	3	21.4	3	18.7	2	14.2	8
P. chrysogenum	1	10	3	21.4	2	3	5	31.3	3	21.4	14
P. digitatum	2	20	2	14.2	4	28.6	5	31.3	4	28.8	17
P. funiculosum	-	-	3	21.4	-	-	-	-	-	-	3
P.purpurogenum	2	20	-	-	-	-	-	-	-	-	2
P. restrictum	–	–	5	35.7	–	–	–	–	1	7.1	6
P. thomii	3	30	-	-	4	6	2	12.5	3	21.4	12
P. viridicatum	2	20	1	7.1	1	7.1	1	6.25	1	7.1	6
Fusarium sp.	–	–	10	50	–	–	3	20	4	20	17
F. moniliforme	-	-	2	20	-	-	-	-	-	-	2
F. oxisporium	–	–	3	30	–	–	1	33.3		–	4
F. solani	–	–	2	20	–	–	1	33.3	4	100	7
F. tabacium	–	–	1	10	–	–	1	33.3	–	–	2
F. tricinctum	–	–	2	20	–	–	–	–	-	-	2
Total											288

Table 5. Aflatoxin production by *A. flavus* isolated from feeds.

Source of A. flavus	Aflatoxin production				Toxigenic isolates		Levels of aflatoxin B1 (ppm)		
	No. of tested isolates	No. of +ve isolates	%	Mean count	No.	%	Max.	Min.	Mean
Yellow corn	20	12	60	3.9×10^1 ±0.0124	2	16.6	0.8	0.4	0.6
Soya bean	20	16	80	1.68×10^1 ±0.037	2	12.5	4.0	2.0	3.0
Hay	20	20	100	1.9×10^1 ±0.04	8	40	4.0	0.4	2.3
Tibin	20	16	80	2.62×10^1 ±0.055	4	25	2.0	0.4	1.2
Broiler concentrates	20	16	80	1.25×10^1 ±0.028	4	25	2.0	0.2	1.1
Poultry ration	20	20	100	1.9×10^1 ±0.042	2	10	1.2	0.05	0.625
Layer concentrates	20	12	60	1.75×10^1 ±0.039	6	50	6.0	0.4	4.1
Meat and bone meal	20	12	60	0.83×10^1 ±0.019	4	33.3	1.2	0.4	0.8

Table 6. Ochratoxin A production by *A. ochraceus* isolated from feeds.

Source of A. ochraceus	Incidence of isolates				Toxig. isolates		Levels of ochratoxin (ppm)		
	No. tested	No. of +ve	%	Mean count	No.	%	Max.	Min.	Mean
Yellow corn	20	3	15	1×10^1 ±0.069	1	33.3	0.40	0.40	0.40
Hay	20	4	20	0.75×10^1 ±0.02	1	25	0.80	0.80	0.80
Tibin	20	6	30	0.83×10^1 ±0.021	2	33.3	1.20	0.40	0.80
Processed animal feeds	20	4	20	0.75×10^1 ±0.019	2	50	0.40	0.20	0.30
Broilers concentrates	20	3	15	0.5×10^1 ±0.049	1	50	0.05	0.05	0.05
Poultry ration	20	3	15	0.5×10^1 ±0.028	2	66.6	0.80	0.20	0.50
Layers concentrates	20	4	20	0.75×10^1 ±0.029	1	25	1.20	1.20	1.20
Total	140	27	19. 2	0.4×10^1 ±0.234	10	37	4.85	2.85	4.05

Table 7. MIC of Muv-Anti mould on *A. flavus* and *A. ochraceus.*

Source of isolates	No. of tested isolates	MIC of tested drug (µg/ml) on A. flavus						No. of tested isolates	MIC range of tested drug (µg/ml) on A. ochraceus					
		0.06	0.125	0.25	0.5	0.75	1.0		0.06	0.125	0.25	0.5	0.75	1.0
Yellow corn	5	2×10^{3}	1×10^{2}	0.2×10^{1}	0.1×10^{1}	0	0	2	2×10^{3}	6×10^{2}	5×10^{2}	3×10^{1}	0	0
Poultry ration	6	2×10^{3}	5×10^{2}	1×10^{1}	0	0	0	2	9×10^{2}	8×10^{2}	7×10^{1}	0	0	0
Layer's conc.	5	3×10^{3}	7×10^{2}	0.5×10^{1}	0	0	0	4	6×10^{2}	7×10^{2}	8×10^{1}	0	0	0
Broiler's conc.	4	8×10^{3}	7×10^{2}	5×10^{2}	2×10^{1}	1×10^{1}	0	2	8×10^{3}	6×10^{3}	3×10^{2}	1×10^{1}	0	0
Hay	9	3×10^{3}	2×10^{2}	1×10^{2}	4×10^{1}	0	0	4	7×10^{3}	5×10^{2}	4×10^{2}	3×10^{1}	0	0
Tibin	10	4×10^{3}	6×10^{2}	2×10^{1}	0	0	0	6	6×10^{2}	7×10^{2}	9×10^{1}	5×10^{1}	0	0
Proc. animal feed	3	8×10^{2}	9×10^{1}	1×10^{1}	2×10^{1}	0	0	3	6×10^{3}	2×10^{3}	4×10^{2}	7×10^{1}	0	0
meat and bone meal	4	6×10^{3}	4×10^{2}	3×10^{1}	0	0	0	2	0.06	0.125	0.25	0.5	0.75	1.0

0: No growth of fungal isolate

Table 8. MIC of Mycostatin on *A. flavus* and *A. ochraceus*.

Source of isolates	No. of tested isolates	MIC range of tested drug (µg/ml) on A. flavus					No. of tested isolates	MIC range of tested drug (µg/ml) on *A. ochraceus*					
		0.125	0.25	0.5	0.75	1.0		0.125	0.25	0.5	0.75	1.0	2.0
Yellow corn	5	2×10^3	5×10^2	9×10^1	8×10^1	0	2	1×10^3	2×10^2	5×10^1	6×10^1	0	0
Poultry ration	6	1×10^3	8×10^2	3×10^2	9×10^1	0	2	3×10^4	7×10^3	4×10^3	5×10^2	8×10^1	0
Layer conc.	5	5×10^3	6×10^2	2×10^2	8×10^1	0	4	7×10^3	6×10^3	6×10^2	4×10^2	7×10^1	0
Broiler conc.	4	1×10^3	3×10^2	3×10^2	7×10^1	0	2	7×10^3	9×10^2	6×10^2	4×10^2	3×10^1	0
Hay	9	7×10^4	5×10^3	3×10^2	1×10^1	0	4	6×10^3	9×10^2	6×10^2	5×10^1	0	0
Tibin	10	9×10^2	2×10^2	6×10^1	8×10^1	0	6	3×10^2	5×10^2	7×10^2	9×10^1	8×10^1	0
Proc. animal feed	3	5×10^3	4×10^3	6×10^2	8×10^1	0	3	9×10^3	4×10^2	8×10^1	7×10^1	0	0
meat and bone meal	4	6×10^3	9×10^2	5×10^1	3×10^1	0	2	0.1253	0.25	0.5	0.75	1.0	2.0

0: No growth of fungal isolate

Table 9. Effect of Muv-Anti mould on mould growth in poultry feeds 2 days after addition.

Isolates	Poultry ration samples										
	1	2	3	4	5	6	7	8	9	10	mean
Aspergillus spp.	$1x10^1$	$2x10^1$	$1x10^1$	$1x10^1$	$1x10^1$	$1x10^1$	$2x10^1$	$1x10^1$	$1x10^1$	$1x10^1$	$1.2x10^1$
Fusarium spp.	0	0	$1x10^1$	0	$1x10^1$	0	$2x10^1$	0	0	0	$0.4x10^1$
Mucor spp.	$2x10^1$	$3x10^1$	$1x10^1$	$2x10^1$	$2x10^1$	$1x10^1$	$3x10^1$	$3x10^1$	$2x10^1$	$1x10^1$	$2.0x10^1$
Penicillium spp.	0	0	0	$1x10^1$	0	0	$1x10^1$	0	0	0	$0.2x10^1$
Total mould count	$3x10^1$	$5x10^1$	$3x10^1$	$4x10^1$	$4x10^1$	$2x10^1$	$8x10^1$	$4x10^1$	$3x10^1$	$2x10^1$	

N.B. After one week the growtrh of all fungi was inhibited

Table 10. Detoxification of Aflatoxim B1 and ochratoxin A in poultry and animal feeds by Sat. F. Dray (HSCAS).

feed	Concentration of chemical /% of detoxification of aflatoxin B1								Concentration of chemical /% of detoxification of ochratoxin A							
	0%		1%		2%		3%		0.5%		1.0%		2.0%		3.0%	
	At	%D	At	%D	At	%D	At	%D	At	%D	At	%D	At	%D	At	%D
Poultry feeds	50	0	26±0.01	48	5±0.01	90	0	100	30±0.5*	40	23±0.2	54	8±0.8	84	0	100
Animal feeds	50	0	7.5±0.2	85	0	100	0	100	34±0.3	32	20±0.01	60	3±0.0	94	0	100

At = original amounts of toxin before treatment = 50ppb. %D = % of Detoxification.
HSCAS = Hydrated Sodium Calcium Aluminosilicate. * = Mean of two testing.

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مدي انتشار الفطريات وسمية فطري الأسبرجلس فلافس والأسبرجلس أوكراشيوس المعزولة من العلائق وكيفية السيطرة عليها

رشا حمزة سيد الأهل[1]، عاطف عبد العزيز حسن[1]، عبد العظيم محمد البراوي[1]، رمضان تاج الدين مصطفي سالم[1]، وائل مصطفي توكل[2]، حسام عبد اللطيف[3]، محمد كمال رفاعي[3]

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أجريت هذه الدراسة علي ٢٠٠ عينة من العلائق المفردة (الذرة الصفراء- فول الصويا- القمح- قش التبن) والعلائق المركبة (علائق الدواجن- علائق الحيوان المصنعة- مركزات البادئ والبياض للدواجن ومسحوق العظم واللحم). وقد خضعت هذه العينات لاستبيان التلوث الفطري، وكانت أكثر السلالات تواجدا سلالة الأسبرجلس وقد عزلت من كل العينات. وقد عزلت الفطريات الأخري ولكن بنسب منخفضة مثل البنسيليوم والفيوزاريوم ووالكلادوسبوريوم والألترناريا. وقد تم اختبار عترات الأسبرجلس فلافس والأسبرجلس أوكراشيوس علي قدرتها لإفراز سموم الأفلاتوكسين والأوكراتوكسين علي التوالي. وقد أفادت النتائج إفراز سموم الأفلاتوكسين ب[1] إلي أن أعلي سمية للأسبرجلس فلافس وجدت في العترات المعزولة من مركزات البادئ وقش التبن (٤٠%)، في حين أن العترات المعزولة من فول الصويا وعلائق الدواجن كانت منخفضة النسبة (١٢،٥%، ١٠% علي التوالي). و أفادت نتائج إفراز سموم الأوكراتوكسين أ لفطر الأسبرجلس أوكراشيوس المعزولة من مختلف العينات درجة سمية عالية للعترة المعزولة من علائق الدواجن (١٠٠%) تليها علائق الحيوان المصنعة ومركزات البادئ والذرة الصفراء (٥٠%) في حين أن العترات المعزولة من قش التبن ومركزات البياض مثلت أقل نسبة سمية (٢٥% لكل منها). وقد تم تقييم اثنين من المضادات الفطرية التجارية وتبين أن أقل جرعة مثبطة من الموف-أنتيمولد وميكوستاتين كانت ٠،٧٥ ، ١ نانوجرام لكل مليليتر لجميع الفطريات المعزولة من العلائق. وبعد إضافة المضاد الفطري موف-أنتيمولد لعلائق الدواجن الملوثة انخفضت كثافة التلوث الفطري بعد يومين من الإضافة وتوقف تماما بعد اسبوع. وقد تم اختبار مضاد السموم الفطرية (سات-إف-دراي) الذي استطاع القضاء علي الأفلاتوكسين والأوكراتوكسين أ عندما أضيف للعلائق بنسبة ٣،٢%.