

Short Communication

Seed-borne fungi in domestic bird feed in Saudi Arabia

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Seed-borne fungi from seeds of barley, canary, maize, oats, rice, rapeseed, red millet, sunflower, white millet and yellow millet were investigated. These seeds are available in the market as bird feed, supplied by companies in various countries as healthy and nutritious food for domestic birds. A total of 81 fungal species were isolated. The genus *Aspergillus* was predominant with 23 species, followed by *Penicillium* with 18, *Ulocladium* with 7, *Alternaria* with 6 and *Chaetomium*, *Curvularia* and *Drechslera* with 5 species each. Other genera isolated were *Aureobasidium*, *Cladosporium*, *Fusarium*, *Mucor*, *Rhizopus* and *Syncephalastrum*.

Key Words—domestic birds; feed; Saudi Arabia; seed-borne fungi.

Seed-borne fungi are well known to cause mycotoxicoses in poultry and other livestock (Mehan and Chohan, 1973; Mirocha and Christensen, 1974). *Alternaria*, *Aspergillus*, *Fusarium* and *Penicillium* are some of common genera which produce mycotoxins (Bokhary and Naguib, 1983). The reduction in germination of seeds, pre- and post-emergence death, and discoloration of seeds are other undesirable effects of seed-borne fungi (Ashokhan et al., 1979; Handoo and Aulakh, 1979; Shafie and Webster, 1981; Abou-Heilah, 1984; Bokhary, 1986). The seeds used in bird feed are imported under different brand names and are described as nutritious and healthy food for domestic birds. Our aim, therefore, is to report on the contamination of seed-borne fungi in imported feeds for domestic birds.

Different brands of bird feed imported from various countries were purchased from local markets. These feeds contain mixtures of the following seeds: barley, canary, maize, oats, rice, rapeseed, red millet, sunflower, white millet and yellow millet.

The media used for the detection of seed-borne fungi were Czapek-Dox Agar (CZA), potato-dextrose agar (PDA), potato-dextrose agar+0.3% yeast extract (PDA+yeast), malt-extract agar (MEA), and Sabouraud-dextrose agar (SDA). All these media were supplied by Oxoid Ltd., London (U.K.). For surface sterilization, seeds were immersed in 1% sodium hypochlorite for 10 min. A total of 500 mixed seeds from the different feed brands (25 seeds per plate) were plated in 13-cm Petri plates for each medium. These plates were incubated at room temperature (22–25°C) for 10 d, until the seeds germinated, and thereafter fungi were detected and isolated. Isolated pure cultures were maintained on PDA+yeast.

Eighty-one species belonging to 13 genera of fungi were isolated from seeds supplied as bird feed (Table 1). Among the predominant genera were *Aspergillus* with 23 spp. and *Penicillium* with 18 spp. These were followed by *Ulocladium* (7 spp.), *Alternaria* (6 spp.), and *Chaetomium*, *Curvularia* and *Drechslera* with 5 species each.

Aspergillus amylovorus, *Aspergillus candidus*, *Aspergillus fumigatus*, *Aspergillus niger*, *Fusarium equiseti*, *Mucor circinelloides*, *Penicillium chrysogenum* and *Ulocladium atrum* were the predominant species, found in all types of seeds and also generally exhibiting a higher percentage of contamination than other species (Table 1). *Aspergillus ellipticus*, *Chaetomium bostrychoides* and *Cladosporium cladosporioides* were other common species found in 8 out of 10 types of seed studied. The highest percentage of contamination was shown by *A. niger* (46%) of rapeseed grains, followed by *A. ellipticus*, which exhibited 41% infection of rice grains. More than 30% infection of seeds was also shown by *A. amylovorus*, *A. candidus*, *A. flavus*, *A. fumigatus*, *A. oryzae*, *C. cladosporioides*, *F. equiseti*, *P. chrysogenum*, and *U. atrum*, but on different types of seed.

Forty-one species were reported from rice grains, the maximum record in this study, followed by barley seeds, on which 33 spp. are reported.

The percentage germination and level of contamination of seeds are given in Table 2.

The genera of fungi isolated, e.g., *Alternaria*, *Aspergillus*, *Chaetomium*, *Curvularia*, *Drechslera*, *Fusarium*, *Mucor*, *Penicillium*, *Rhizopus*, *Syncephalastrum* and *Ulocladium*, have already been reported from locally grown barley (Kassim, 1987; Hashem, 1990) and from other seeds like maize, rice and rapeseed (Abdel-Hafez, 1984; Sejeny et al., 1984; Bokhary, 1991), but the species diversity and

Table 1. Percentage infection by seed-borne fungi on various types of seed.

Isolates ^{a)}	% Infection ^{b)}										
	B	C	M	O	Ra	Ri	Rm	Sf	Wm	Ym	
<i>Alternaria alternata</i>	6	9	7	2	3	11	4				
<i>A. brassicicola</i>	7		4			13					
<i>A. chlamydospora</i>			6	17	2	3					
<i>A. chrysanthemi</i>							2	16			
<i>A. consortialis</i>	4	2	9		3	5			7		
<i>A. eureka</i>						4			7	11	
<i>Aspergillus amylovorus</i>	19	9	29	23	16	32	2	3	8	11	
<i>A. apica</i>	19	9	29	23	16	32	2	3	8	11	
<i>A. avenaceus</i>		13		3							
<i>A. awamori</i>	7	6		2	7			2			
<i>A. caespitosus</i>							4		1	3	
<i>A. candidus</i>	16	21	14	32	15	15	9	11	2	19	
<i>A. carbonarius</i>	12					6			4	6	
<i>A. carneus</i>	7	3	2		4	17					
<i>A. chevalieri</i> (<i>Eurotium chevalieri</i>)								16			
<i>A. clavatus</i>		12					4			1	
<i>A. ellipticus</i>	32	16	39	22	2	41	21	16		6	
<i>A. flavus</i>	16			16	4	3	9	3	11	4	
<i>A. fumigatus</i>	17	9	23	31	15	16	6	13	23	19	
<i>A. japonicus</i>		3		1				2			
<i>A. leporis</i>		14									
<i>A. nidulans</i> (<i>Emericella nidulans</i>)	12		13			6					
<i>A. niger</i>	21	32	43	34	46	39	21	33	43	20	
<i>A. oryzae</i>	6		12			6					
<i>A. parasiticus</i>			12			6					
<i>A. phoenicis</i>		6							2		
<i>A. restrictus</i>						16					
<i>A. subsessilis</i>										12	
<i>A. terreus</i>	16										
<i>Aureobasidium pullulans</i>	12						3				
<i>Chaetomium atrobrunneum</i>		5									
<i>C. bostrychodes</i>	6	4	9	13	2	19	2	4			
<i>C. cochlioides</i>		3									
<i>C. elatum</i>									6		
<i>C. funicola</i>								17			
<i>Cladosporium cladosporioides</i>	16	29	4	21	36	13	16	41		6	
<i>C. herbarum</i>						21					
<i>C. tenuissimum</i>									7		
<i>Curvularia clavata</i>						11					
<i>C. deightonii</i>										7	
<i>C. ellisi</i>		16	6		6	12	6			5	
<i>C. geniculata</i>		12									
<i>C. lunata</i>	4	3	2								
<i>Drechslera australiensis</i>						11					
<i>D. bicolor</i>					12						
<i>D. cynodontis</i>										3	
<i>D. dematioidea</i>								16			
<i>D. dictyoides</i>		3									
<i>Fusarium equiseti</i>	12	6	17	12	6	19	24	19	29	36	

Isolates ^{a)}	% Infection ^{b)}										
	B	C	M	O	Ra	Ri	Rm	Sf	Wm	Ym	
<i>F. flocciferum</i>					7						
<i>F. graminearum</i>									7		
<i>Mucor circinelloides</i>	12	12	6	3	7	4	21	16	16	3	
<i>M. zonatus</i>						9					
<i>Penicillium aragonense</i>			12								
<i>P. argillaceum</i>	16					11				6	
<i>P. asturianum</i>								6			
<i>P. brevicompactum</i>	2	6									
<i>P. capsulatum</i>								6			
<i>P. castellae</i>								16			
<i>P. chrysogenum</i>	12	16	4	9	5	16	32	21	16	23	
<i>P. cieglerei</i>				7							
<i>P. citrinum</i>					16						
<i>P. concentricum</i>		12					3				
<i>P. coraligerum</i>										6	
<i>P. funiculosum</i>	16			3			9				
<i>P. islandicum</i>	6		4			3	1	2		1	
<i>P. manginii</i>			3			6					
<i>P. rubrum</i>	2		6			4				2	
<i>P. rugulosum</i>				3	1		3				
<i>P. thomii</i>		16			2				3		
<i>P. verrucosum</i>							3	14			
<i>Rhizopus microsporus</i>					3	16		2			
<i>R. oryzae</i>						14	3		6		
<i>Syncephalastrum racemosum</i>	14	3	19	16		5					
<i>Ulocladium atrum</i>	14	21	11	23	16	26	32	29	20	29	
<i>U. chartarum</i>	4	3							3	6	
<i>U. chlamydosporum</i>	13	9	4		3	16		2			
<i>U. consortiale</i>							6		3	9	
<i>U. oudemansii</i>				6	4	9					
<i>U. septosporum</i>	3		2			6		13	4	3	
<i>U. tuberculatum</i>	4	2		7		3	2			6	

a) Total no. of fungal genera=13; total no. of fungal species=81.

b) B=barley; C=canary; M=maize; O=oats; Ra=rapeseed; Ri=rice; Rm=red millet; Sf=sunflower; Wm=white millet; Ym=yellow millet.

numbers were different in this study. The numbers of *Aspergillus* and *Penicillium* spp. were higher in the present study. Several other genera reported earlier like *Acremonium*, *Choanephora*, *Epicoccum*, *Geotrichum*, *Nigrospora*, *Phoma*, *Pleospora* and *Setosphaeria*, were not encountered in the present study.

Percentage germination and contamination of seeds on various media shown clearly that there are different growth for each kind of seeds on the different used media. The highest percentage of germination was shown by barley, canary, maize, oats and rapeseed on SDA and by canary, maize, oats, rapeseed, rice and red millet on PDA, while the highest percentage of contamination was shown by barley, canary, maize, oats, rice and sunflower on PDA + yeast.

From the present study, the main genera of fungi as-

sociated with mycotoxin production include species of *Aspergillus*, *Penicillium*, *Fusarium*, and *Alternaria*. Aflatoxins produced by *Aspergillus flavus*, *Aspergillus nomius*, *Aspergillus parasiticus* are by far the most widely studied mycotoxins and are presumed to be the most wide spread of fungal toxicants (Bokhary and Naguib, 1983; Kassim et al., 1987).

Aflatoxins are now considered to be among the world's most dangerous contaminants in foods and feeds (Wainwright, 1992). Species of *A. flavus* isolated from barley and maize from Saudi Arabia were shown to produce aflatoxins (Al-Julaifi and Al-Khaliel, 1992; Ewaidah, 1992). Mycotoxicoses produced by some *Aspergillus* spp. and *Penicillium* spp. are a major cause of problems with barley grains used for poultry feed in Saudi Arabia (Ewaidah, 1988). In the present investigation, *A.*

Table 2. Percentage germination and contamination of seeds on various media.

Medium ^{a)}	Barley ^{b)}		Canary		Maize		Oats		Rapeseed		Rice		Red millet		Sunflower		White millet		Yellow millet	
	germ	cont	germ	cont	germ	cont	germ	cont	germ	cont	germ	cont	germ	cont	germ	cont	germ	cont	germ	cont
F.P.	93	44	86	35	89	39	93	43	94	32	84	29	93	36	83	29	76	24	76	32
CZA	96	37	97	39	93	39	97	34	96	38	94	43	90	42	89	31	93	22	99	33
MEA	93	36	96	29	94	42	96	39	94	28	93	41	97	36	99	35	99	32	99	38
PDA	95	46	99	42	98	46	99	46	98	38	99	49	97	39	99	43	99	33	95	32
SDA	100	32	100	36	98	43	99	39	95	36	98	46	95	49	95	49	94	43	96	42
PDA+yeast	83	62	86	69	76	70	92	73	86	69	94	54	82	76	93	56	99	39	99	44

a) F.P.=Filters paper; CZA=Czapek-Dox agar; MEA=Malt-extract agar; PDA=Potato-dextrose agar; PDA+yeast=Potato-dextrose agar+0.3% yeast extract; SDA=Sabouraud-dextrose agar.

b) germ=% germination; cont=% contamination.

flavus, *A. fumigatus*, *A. terreus*, *Emericella nidulans*, *Penicillium chrysogenum* and *P. citrinum* were found as toxinogenic fungi, and this accords with the earlier finding of barley grain for poultry feed in Saudi Arabia (Al-Julaifi and Al-Khaliel, 1992).

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