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TRAINING ACTIVITIES ON FOOD CONTAMINATION CONTROL
AND MONITORING WITH SPECIAL REFERENCE TO MYCOTOXINS

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PROBLEMS OF MYCOTOXINS IN AFRICA



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GENERAL

Mycotoxin is a term reserved for a group of highly toxic substances produced as secondary metabolites by several fungi. These substances often differ greatly in structure, chemical and physical properties but possess the capability of producing pathological and other undesirable conditions in man and animals. Such conditions are normally referred to as mycotoxicoses. Mycotoxins constitute a major problem for human and animal health because, under normal conditions an exist in those regions of the earth with warm climate and high relative humidity, foods and feeds readily provide ideal conditions for both mycelial growth and toxin elaboration. In such circumstances, even a food with no evident sign of mould contamination may be rich in mycotoxin. Most mycotoxins are of low molecular weight and are therefore of high mobility. Consequently, they often penetrate deep into the food, away from the surface.

In historical terms, outbreaks of such human mycotoxicoses as ergotism and mushroom poisoning were recorded several decades ago. Cases of other human and animal mycotoxicoses were subsequently reported, and prominent among the human out-breaks were Alimentary Toxic Aleuka (ATA), Yellow Rice Intoxication and Stachybotryotoxicosis.

Prior to 1960, mycotoxins and their various effects enjoyed minimal prominence in the scientific literature but came into scientific focus after the discovery of aflatoxin in the early 1960s. Since then, several new mycotoxins have been discovered and there has been a sustained interest in some of these with

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a resultant rapid growth in knowledge of their chemistry, toxicology and epidemiology.

As at 1982, approximately 220 moulds have been established as being toxigenic, and they belong to the genera listed in Table I. Although most research on this subject has been carried out on the aflatoxins, and much of the indirect evidence of human toxicity and carcinogenicity relate to the aflatoxins, it has been shown that many other mycotoxins constitute significant health hazard for man and livestock where the livers, kidneys, circulating systems and blood-forming organs appear to be the main target organs. Table 2 represents a summary of some of the important mycotoxins, the producer organisms, and the corresponding toxic effects.

Table I

Genera of fungi containing toxigenic species

(Mossel, 1982)

Absidia	Helminthosporium	Scopulariopsis
Acremonium	Microsporon	Sporidesmium
Alternaria	Mucor	Stachyboris
Aspergillus	Myrothecium	Talaromyces
Byssochlamys	Nigrospora	Thamnidium
Cephalosporium	Paecilomyces	Thermoascus
Chaetomium	Penicillium	Thermomyces
Cladosporium	Phoma	Trichoderma
Cladotrichum	Pithomyces	Thricothecium
Claviceps	Phizoctonia	Verticillium
Dacylomyces	Rhizopus	Zygosporium
Fusarium	Sclerotinia	

Table 2

Mycotoxins, producer organisms and toxic effects

Mycotoxin	Origin	Toxic effect
1	2	3
Aflatoxin	Asp. flavus and parasiticus; possibly also other species and genera	Carcinogenicity, with special affinity to the liver; immunosuppression
Aflatrem	Asp. flavus	Tremorgenicity
Alternariol	Various species of Alternaria	Haemorrhagic effects
Aspergillic acid	Asp. flavus	Neurotoxicity
Aspertoxin	Asp. flavus	Similar to sterigmatocystin
Byssochlamic acid	Byssochlamys fulva	Inhibition of some essential enzymes; haemorrhagic effects
Chaetoglobosin	Various species of Chaetomium	Similar to cytochalasins
Chrysophanol	Pen. islandicum	Mutagenicity
Citreoviridin	Pen. citreoviride	Neurotoxic effects
Citrinin	Various species of Penicillium; Asp. niveus	Nephrotoxicity
Citromycetin	Pen. frequentans and roseopurpureum	Similar to sterigmatocystin
Cyclochlorotin	Pen. islandicum	Hepatotoxicity, carcinogenicity
Cyclopiazonic acid	Pen. cyclopium and camemberti; Aspergillus spp.	Carcinogenicity; necrotic effects
Cytochalasins	Species of Phoma and Helminthosporium; Asp. clavatus	Disruption of contractile micro-
Diacetoxyscirpenol	Fus roseum	Dermatitic and intestinal haemorrhagic effects; immunosuppression

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1	2	3
Diplodiol	<i>Diplodia macrospora</i>	Anorexia, lethargy
Emodin	<i>Asp. wentii</i> ; <i>Pen. brun-</i> <i>neum</i> and <i>cyclopium</i> ; <i>Cladosporium</i> spp.	Diarrhoeagenicity, mutagenicity
Fusitremergen	<i>Asp. caespitosus</i> and <i>fumigatus</i> ; <i>Pen. piscarium</i>	Tremorgenicity
Fusarenone	<i>Fus. nivale</i>	Inhibition of protein synthesis by cells of haematopoietic tissues
Fusarin C	<i>Fus. moniliforme</i>	Mutagenicity
Fusariogenin	<i>Fus. sporotrichiodes</i>	Alimentary toxic aleukia (ATA)
Griseofulvin	<i>Pen. islandicum</i>	Carcinogenicity
Islandicin	<i>Pen. islandicum</i>	Mutagenicity
Islanditoxin	<i>Pen. islandicum</i>	Hepatotoxicity
Janthitrem	<i>Pen. janthinellum</i>	Tremorgenicity
Kojic acid	Various species of <i>Aspergillus</i> and <i>Penicillium</i>	Convulsant effects
Luteoskyrin	<i>Pen. islandicum</i>	Carcinogenicity with special affinity to the liver
Maltoryzine	<i>Asp. oryzae</i>	Haemorrhagic and neurotoxic effects
Mollicellins	<i>Chaet. mollicellum</i>	Mutagenicity
Moniliformin	Various species of <i>Fusarium</i>	Myocardial degeneration
Mycophenolic acid	Various species of <i>Penicillium</i>	Toxicity of leucocytes, lead- ing to anaemia
Neosolanin	<i>Fus. tricinctum</i>	General and der- mal toxicity.

1	2	3
Bivalenol	<i>Fus. nivale</i>	Similar to fusarenone
Ochratoxins	Various species of <i>Aspergillus</i> and <i>Penicillium</i>	Hepatotoxicity; nephrotoxicity
Oxaline	<i>Pen. oxalicum</i>	Neurotoxicity
Paspalinine	<i>Claviceps paspali</i>	Tremorgenicity
Patulin	Various species of <i>Penicillium</i> and <i>Aspergillus</i> Byss. nivea	General toxicity, and possibly carcinogenicity
Paxilline	<i>Pen. paxilli</i>	Tremorgenicity
Penicillic acid	Various species of <i>Penicillium</i> and <i>Aspergillus</i>	Carcinogenicity; cardiotoxicity
Penitrems: of Tremortin P.R. toxin (Epoxyoptalones)	<i>Pen. roqueforti</i>	Hepatotoxicity; nephrotoxicity
Psoralene	<i>Sol. sclerotiorum</i>	Dermatitic effects
Roquefortine	<i>Pen. roqueforti</i>	Neurotoxicity, leading to convulsive seizures
Roridin E	<i>Stach. atra</i>	Alimentary toxic aleukia (ATA)
Roseotoxin B	<i>Trich. roseum</i>	Inflammatory effects
Rubratoxins	<i>Pen. rubrum</i> and <i>purpurogenum</i>	Haemorrhagic effects; hepatotoxicity
Rugulosin	<i>Pen. rugulosum</i>	Hepatotoxicity; carcinogenicity
Secalonic acid D	Various species of <i>Penicillium</i>	Haemorrhagic effects
Simatoin	<i>Pen. islandicum</i>	Hepatotoxicity
Slaframine	<i>Rhiz. leguminicola</i>	Interaction with parasympathic nerve system

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I	2	3
Solaniol	Fus. solani	Neurotoxicity
Sporidesmin	Pith.chartarum	Hepatotoxicity; facial eczema
Sporofusariogenin	Fus. sporotrichioides	Alimentary toxic aleukia (ATA)
Stachybotryotoxin	Stach. atra.	Circulatory, haemorrhagic and dermatitic effects
Sterigmatocystin	Various species of Aspergillus and Chaetomium	Carcinogenicity, with special affinity to the liver
Tenuazonic acid	Alt. alternata and tenuissima	Haematological disorders
Terreic acid	Asp. terreus	Hepatotoxicity
Territrems	Asp. terreus	Tremorgenicity
Tremortin (Penitrem)	Various species of Penicillium	Tremorgenicity
Trichothecene group ("T-2 toxin" etc.)	Various species of Fusarium; Trich. roseum	Alimentary toxic aleukia, neurotoxicity; teratogenicity; inflammations
Tryptoquivaline	Asp. clavatus	Tremorgenicity
Verrucarins	Stach. atra; Myrothec. roridum	Haemorrhagic effects
Verruculogen	Various species of Penicillium; Asp. caespitosus	Tremorgenicity
Vomitoxin (Deoxynivalenol)	Various species of Fusarium	Emesis
Xanthoscin	Asp. candidus	Myocardial and pulmonary lesions

1	2	3
Ianthomegnin, viomellin and related mycotoxins	Various species of Aspergillus and Trichophyton; Pen. viridicatum; Microspor. cockii	Hepatotoxicity nephrotoxicity
Zearalenol	Fus. roseum	Affecting the uterus
Zearalenone	Various species of Fusarium	Emesis; inter- ference with steroid hormone systems

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INCIDENCE OF MYCOTOXINS AND MYCOTOXICOSES IN AFRICA

Mycotoxicoses as a health and economic problem have engaged the attention of farmers, veterinarians, mycologists and governments the world over. In the African region, for reasons of underdevelopment, and its attendant handicaps, the amount of documentation on the problem of mycotoxins is meagre, though conclusively indicative of the existence of an established problem. Based on available information, it is convenient to treat this subject under the following sub-regional groupings:

- South/East African sub-region
- West African sub-region
- North African sub-region.

South/African sub-region

1. Schneider et al (1979) reported an outbreak of mortality in a flock of mutton marine sheep, in which 109 out of 568 sheep died, in the South Western Cape Province. This outbreak was characterized by haemorrhagic septicaemia, anaemia and leucocytopenia. The cause of this outbreak was traced to the uninterrupted consumption of sheep cubes processed from fungus infested wheat, barley and rye straw. Toxigenic strains of Stachybotrys chartarum were incriminated. This report, represented the first description of an outbreak of stachybotryotoxicosis in sheep associated with the ingestion of S. chartarum infested feed component in South Africa.

2. During the spring and summer of 1979, Ancock et al (1980) reported field outbreaks of porcine hyperoestrogenism in the Natal Midlands. One outbreak was triggered off in the Ipero

District in pigs after the consumption of a mixed ration containing 0.95 mg/kg zearalenone. The carrier feed component was yellow maize (10 mg/kg). It was observed that dilution of the contaminated maize with good quality white maize drastically reduced the incidence and severity of clinical signs within 3 - 4 days. Another outbreak involving pigs weighing 40 kg was observed, following the consumption of a mixed ration containing chemically undetectable level of zearalenone. The carrier maize component this time, contained only 0.06 mg/kg, zearalenone. This report again represented the first recorded field outbreak of porcine hyperoestrogenism associated with the ingestion of F.graminearum-infected maize in South Africa.

3. Pienaar et al. (1981), reported four outbreaks of leucoencephalomalacia in horses, in widely separated areas within South Africa. In each instance, mouldy maize contaminated with Fusarium verticillioides (Moniliforme) was involved. Clinical signs and pathological lesions were identical to those seen in experimentally produced cases of F.verticillioides poisoning in horses.

4. The existence of a link between mycotoxins and cancer in Africa, has been suggested by many workers, (Oettle, 1965; Butler 1974, Linsell et al., 1979, Morasas et al, 1979). The mycotoxin hypothesis for the aetiology of hepatic carcinoma was advanced by Oettle (1965). Particularly with respect to aflatoxins, there is a wealth of indirect evidence which suggest that aflatoxins play a role in human neoplasia induction. This type of carcinoma shows a well defined geographical distribution whereby high incidence areas occur in the sub-saharan areas of Africa and South East Asia.

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Pears and Linsell (1977), reported a high degree of positive correlation between ingestion levels of aflatoxin, and adult incidence rates of hepatocellular carcinoma, based on a 7-year study of cancer registration in the Murang district of Kenya and in Swaziland. The high exposure rate of man to aflatoxin in this sub-region of Africa and perhaps beyond, has been clearly demonstrated by Alpert et al. (1971), who reported that, 40% of food samples tested in Uganda contained aflatoxin, with 15% exceeding the 1 ppm level. Butler (1974), observed that in Kenya, Swaziland and Mozambique where, the incidence of hepatomas is high, aflatoxins are often detected in human foods. In the view of Linsell and his associates (1979), aflatoxin ingestion in Mozambique is the highest found in all Africa, averaging 222 ng/kg a day, compared with virtually zero in the U.S. As a consequence, the cancer incidence rate in Mozambique is 58 times that of the U.S.

5. Indirect evidence of the possible role of mycotoxins other than aflatoxins; has been provided by Morasas et al. (1979) from the Republic of Transkei where the South-Western district is reputed to have the highest rate of oesophageal cancer rate in the entire continent, while the rate in the northwestern region of the country is relatively low. Morasas and his associates isolated F.graminearum, F.verticillioides and F.sacchari var. subglutinans as well as, the mycotoxins deoxynivalenol and zearalenone from corn which forms the dietary staple of the two districts. The level of natural contamination was however considerably higher in the high incidence area of oesophageal cancer than in the low incidence area, suggesting a link between deoxynivalenol and zearalenone with this disease.

6. An evidence of the acute toxicity effect of aflatoxin to man as distinct from the carcinogenic effect, was presented by Serck-Hansen (1970) who reported the death of a boy in Uganda from acute liver damage, following the ingestion of a cassava meal heavily contaminated with aflatoxin.

7. Apart from the above described cases implicating mycotoxins in outbreaks of animal and human diseases, the mycotoxin problem is further underlined by the common occurrence of toxigenic mould strains in the African environment.

Rabie et al. (1982) found four new moniliformin producing species of Fusarium namely, F.acuminatum, F.concolor, F.equisetii and F.semitectum. Isolated of F.acuminatum and F.concolor produced as much as 3.4 and 9.5 g/kg of the toxin respectively. It was further shown that South African isolates of F.oxysporum, F.avenaceum, F.fusarioides and F.moniliforme var. subglutinans produced large amounts of moniliformin in corn ears in the field.

8. Working in South Africa with Diplodia macrospora isolated from Zambian white maize, Kriek and Morasae (1979), demonstrated the toxicity of maize cultures of the Zambian D.macrospora in rats and ducklings. Pulmonary haemorrhage, alveolar, septal and perivascular oedema of the lung, mild cholangitis and a mild renal tubular nephrosis were the most important histological changes.

9. Itakura et al. (1980) reported the isolation of mycotoxigenic fungi from Ugandan foodstuffs. Strains (209 in all) of these fungi, showed strong acute toxicity on mice while A.flavus,

A.oryzae and A.candidus caused liver atrophy. A.flavus in addition caused marked pleomorphism of liver cell nuclei. A few strains of P.funoulosum caused swellings and nuclear pleomorphism of proximal tubules of the kidney.

West African Sub-region

Information of the mycotoxin problem in this sub-region came from Nigeria and Senegal. Here, none of the information contained studies linking mycotoxin ingestion to incidence of cancer. They are rather reported of surveys of local foodstuffs for aflatoxin contamination or potential contamination from which inferences could be made (Table 3).

1. Alozie et al. (1980), screened 16 of the commonest local foodstuffs and indigenous beverages and found that all 8 samples of beverages were contaminated with aflatoxin. The foodstuff samples (except ewedu, dawadawa and shokoyokoto) also contained aflatoxin.

2. Emerole et al. (1982) found aflatoxin in crops and spices procured from markets in Western part of Nigeria.

3. Uraih and Ogbadu (1980) working in Northern Nigeria, found low levels of aflatoxin in sorghum (Borghum vulgare) after harvesting and attributed the low aflatoxin content of the grains to the prevailing high temperatures and low moisture levels at the time of the study.

4. From the Eastern part of Nigeria, Nwukolo and Okonkwo in a survey of common foods in the Savanna and forest regions of Nigeria, found high levels of aflatoxin contamination in foods

(sorghum, millet, groundnut, dried fish, palm oil), stored under sub-optimal conditions. They suggested that such aflatoxins may work synergistically with other carcinogens to produce the high incidence of primary liver cancer common in young people under the age of 40. The report of a high rate of contamination of Nigerian foodstuffs by Nwokolo and Okonkwo, and the indication of a high incidence rate of liver cancer amongst young adults in Nigeria suggested, that, the mycotoxin problem in Nigeria may not differ from that of South/East African sub-region. Population based studies of a link between dietary mycotoxin and cancer incidence in Nigeria are urgently required.

Table 3

Mycotoxin contamination of foods and feeds in
the West African Sub-region

Organism	Source	Mycotoxin	Reference
-	Nigerian foodstuffs and indigenous beverages	Aflatoxin	Alozie et al. (1980)
-	Crops and spices from Nigerian markets	Aflatoxin	Emerole et al. (1982)
-	Sorghum after harvesting	Aflatoxin	Uraih & Ogbadu (1980)
-	Sorghum, millet groundnut, dried fish, palm oil under storage	Aflatoxin	Nwekolo and Okonkwo (1978)
<u>Fusarium</u> <u>Rigidiosu-</u> <u>lum</u>	Dry forages from Senegal	-	Le Bars & Labouche (1979)
<u>A.flavus</u> <u>A.niger</u> <u>A.nidulans</u>			

- - Not Determined

North African Sub-region

Available information on this subject from the North-African sub-region comes from Egypt, Tunisia and the Sudan.

I. In a study on the relationship between aflatoxin and kwashiorkor amongst Sudanese children, Hendricke et al (1982) screened sera and urine samples from 44 kwashiorkor children, 32 with marasmic kwashiorkor and 70 with marasmus for their aflatoxin content using high performance liquid chromatography.

Aflatoxin was detected more often and at higher concentrations in sera from children with kwashiorkor. Aflatoxicol, a metabolite of aflatoxins B₁ and B₂, was detected in children with kwashiorkor and marasmic kwashiorkor. The study showed in clear terms that these children and perhaps the Sudanese population are exposed to aflatoxins in their environment. Survey studies of the mycotoxin contamination of the North African environment clearly show that as in the other sub-regions the problem of mycotoxin is real (Table 4).

Table 4

Mycotoxin contamination of foods and feeds in
North-African sub-region

Organism	Source	Mycotoxin	Reference
<u>Stachybotrys</u> <u>Chartarum</u> <u>S.microspora</u> <u>S.kampalensis</u>	Wheat straw from Egypt	Verrucarol Verrucaric J Roridin E Satratoxin H	El.Kaaly & Moubasher (1982)
<u>A.flavus</u>	Egyptian feeds	Aflatoxins	Abdel-Fattah et al. (1982)
<u>A.egyptiacus</u> <u>A.carneus</u> <u>A.terricola</u>	Egypt	Aflatoxins B ₁ , B ₂ , G ₁ , G ₂	Moubasher et al. (1977)
-	Egyptian food samples under storage	Aflatoxin	Qulet et al. (1983)
-	Tunisian food samples under storage	Aflatoxin	Boutrif et al. (1977)

- = Not Determined

Control Measures

Ideally, the prevention of mycotoxicoses and the attendant human and economic loss calls for either the exclusion of the causative agents from our environment or the prevention of the growth and metabolism of toxigenic fungal strains on foodstuffs and animal feeds. The second option is a more feasible proposition, and indeed, is the approach adopted in most African countries.

1. Control through Prevention of Fungal Growth:

To achieve this, the following measures are necessary:

- (a) Prompt removal of harvested crops to shelter where they should not be allowed to gain moisture
- (b) High moisture foodstuffs and agricultural products should be dried down to a water content \sphericalangle equivalent \sphericalangle level to a water activity of 0.70. The alarm water content equivalent of this is shown for some common foods (Table 5). The technique of solar drying is recommended.
- (c) Damaged fruits, kernels and mouldy foodstuffs should be discarded.
- (d) Where possible, and depending on the food, refrigeration should be applied.
- (e) Agricultural products should be protected with chemicals against insects and vermins.

All the enumerated preventive measures are applied in most African countries. Their ineffectiveness as evidenced by the wide spread contamination of foods with toxigenic moulds and mycotoxins, derives from the unregulated application of the

measures and the absence of any monitoring system in most of our countries. Monitoring of high risk food in Africa will be a very effective control measure as has been demonstrated in the United States of America. In this regard, it should be the responsibility of governments to install low cost mycotoxin contamination testing service for producers, and purchasers alike. This is the practice in some advanced countries where the assault on mycotoxins is executed with a missionary zeal.

2. Control through application of High temperatures

Prolonged cooking with water, and roasting at high temperature are a common feature of African culinary practice. Unfortunately, these measures are only enough to lower the level of mycotoxin in a food without eliminating the chronic toxicity risk.

3. Research Efforts at Mycotoxin Control

Many workers in Nigeria are currently engaged in research on mycotoxin control. Some of the findings though encouraging, are yet to provide the answer to the problem.

- (a) Control using gamma-radiation: Ogbadu (1980), explored the effectiveness of low doses of radiation and reported a decrease in aflatoxin B₁ production by irradiated spores of A. flavus in Nigerian foods. Total inhibition was achieved in soya beans and groundnuts at 500 Krad.

- (b) Benzoic acid and its derivatives: Uraih and Offonry (1981), reported complete inhibition of A.flavus in groundnuts at the following concentrations: Benzoic acid (10 mg/g) sodium benzoate 24 mg/g; salicylic acid, 20 mg/g; ethyl-p-aminobenzoate, 3 mg/g (for aflatoxin inhibition) and (10 mg/g for mycelial growth).
- (c) Woodsmoke: Uraih and Ogburn, (1982), found that wood smoke inhibited aflatoxin synthesis on fish and exerted fungistatic effect at reduced water content.

4. Other Relevant Efforts

- (a) Doyle and Marth (1978) reported that at high temperature, SO_2 at 2000 ppm or more, will reduce aflatoxin to an acceptable level.
- (b) Brekke et al. (1978), described a pilot-scale treatment of 4.86 metric ton of corn at 11% moisture content, using 1.1% NH_3 which reduced the aflatoxin B_1 level from 90 mg/kg to a nondetectable level during 7 months of in-door storage. Swine feeding tests on this ammoniated corn gave good results.
- (c) Hitokoto et al. (1980) demonstrated that cloves, staranise seeds and allspice caused complete inhibition of A.ochraceus, A.flavus and A.versicolor. The active principles-eugenol (cloves) and thymol (Thyme) caused complete inhibition of the moulds at 0.4 mg/ml while anethol (staranise seeds) was effective at 2 mg/ml.

Table 5

Alarm water content of some staple foods
assuming $a_w = 0.70$ at 20°C

Food	Alarm water content
Nuts	4 - 9
Whole milk powder	7
Cocoa	7 - 10
Soybeans	9 - 13
Dried whole egg	10
Skim milk powder	10
Dried lean meat and fish	10
Rolled oats	11
Rice	12 - 15
Pulses	12 - 15
Dried vegetables	12 - 22
Wheat flour	13 - 15
Dried soup mixtures	13 - 21
Dried fruits	18 - 25

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