Toxic effects of mycotoxins in humans

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Mycotoxicoses are diseases caused by mycotoxins, i.e. secondary metabolites of moulds. Although they occur more frequently in areas with a hot and humid climate, favourable for the growth of moulds, they can also be found in temperate zones. Exposure to mycotoxins is mostly by ingestion, but also occurs by the dermal and inhalation routes. Mycotoxicoses often remain unrecognized by medical professionals, except when large numbers of people are involved. The present article reviews outbreaks of mycotoxicoses where the mycotoxic etiology of the disease is supported by mycotoxin analysis or identification of mycotoxin-producing fungi. Epidemiological, clinical and histological findings (when available) in outbreaks of mycotoxicoses resulting from exposure to aflatoxins, ergot, trichothecenes, ochratoxins, 3-nitropropionic acid, zearalenone and fumonisins are discussed.

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Introduction

Mycotoxins are secondary metabolites of moulds that exert toxic effects on animals and humans. The toxic effect of mycotoxins on animal and human health is referred to as mycotoxicosis, the severity of which depends on the toxicity of the mycotoxin, the extent of exposure, age and nutritional status of the individual and possible synergistic effects of other chemicals to which the individual is exposed. The chemical structures of mycotoxins vary considerably, but they are all relatively low molecular mass organic compounds.

The untoward effect of moulds and fungi was known already in ancient times (*1*). In the seventh and eighth centuries BC the festival "Robigalia" was established to honour the god Robigus, who had to be propitiated in order to protect grain and trees. It was celebrated on 25 April because that was the most likely time for crops to be attacked by rust or mildew (*2*).

In the Middle Ages, outbreaks of ergotism caused by ergot alkaloids from *Claviceps purpurea* reached epidemic proportions, mutilating and killing thousands of people in Europe. Ergotism was also known as *ignis sacer* (sacred fire) or St Anthony's fire, because at the time it was thought that a pilgrimage to the shrine of St Anthony would bring relief from the intense burning sensation experienced. The victims of ergotism were exposed to lysergic acid diethylamide (LSD), a hallucinogen, produced during the baking of bread made with ergot-contaminated wheat, as well as to other ergot toxins and hallucinogens, as well as belladonna alkaloids from mandragora apple, which was used to treat ergotism (*3*). While ergotism no longer has such important implications for public health, recent reports indicate that outbreaks of human mycotoxicoses are still possible (*4*).

Some mycotoxicoses have disappeared owing to more rigorous hygiene measures. For example, citreoviridin-related malignant acute cardiac beriberi ("yellow rice disease" or *shoshin-kakke* disease in Japanese) has not been reported for several decades, following the exclusion of mouldy rice from the markets. Citreoviridin is a metabolic product of *Penicillium citreonigrum*, which grows readily on rice during storage after harvest (*5*), especially in the colder regions of Japan (*6*). Another mycotoxicosis not seen for decades is alimentary toxic aleukia, common in the 1930s and 1940s in the USSR. This disease was caused by trichothecenes produced by *Fusarium* strains on unharvested grain.

General interest in mycotoxins rose in 1960 when a feed-related mycotoxicosis called turkey X disease, which was later proved to be caused by aflatoxins, appeared in farm animals in England. Subsequently it was found that aflatoxins are hepatocarcinogens in animals and humans, and this stimulated research on mycotoxins.

There is a long history of the use of certain moulds in the production of cheese and salami and in the fermentation of beer and wine. Moulds are also used in the production of drugs (antibiotics). The classification of mould metabolites as antibiotics or mycotoxins is based on their toxicity or beneficial effect in treating diseases. Some mould metabolites that were initially considered to be antibiotics (e.g. citrinin) were subsequently found to be highly toxic (*7*), and are currently classified as toxins. Ergot alkaloids are still used, *inter alia*, in the treatment of parkinsonism, as prolactin inhibitors, in cerebrovascular insufficiency, migraine treatment, venous

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Table 1. **Outbreaks of aflatoxicosis**

Country	No. of subjects	Symptoms and signs	Exposure			Material analysed	Toxin	Liver histopathology	Reference
			Source	Duration	Toxin				
Uganda	1; **1**[a]	Abdominal pain, oedema of legs, palpable liver, on ECG prolongation of P-R interval, right bundle branch block	Cassava	5–30 days	Aflatoxin 1.7 ppm	–	–	Centrilobular necrosis, polymorphonuclear infiltration and fibrin in sinusoids, fatty changes in midzonal region	*24*
India	397; **106**	Brief febrile episode, vomiting, anorexia, jaundice, ascites, oedema of legs, massive gastrointestinal bleeding	Maize	Several weeks	Aflatoxin B_1 (5/5)[b] 6.25–15.6 ppm	Serum Urine	Aflatoxin B_1 (2/7) Aflatoxin B_1 (0/7)	Bile duct proliferation with periductal fibrosis, multinucleated giant cells, foamy cytoplasm, bile stasis in bile ducts, dilated biliary canaliculi	*25*
			Maize	–	Aflatoxin B_1 (7/70)[c] < 0.1 ppm	–	–		*26*
India	994; **97**	Fever, vomiting, oedema of feet, jaundice, hepatomegaly, ascites, splenomegaly	Maize	–	Aflatoxin B_1 (13/14) 0.01–1.1 ppm	–	–	Cholangiolar proliferation, perivenous collagenosis, luminal obliteration, extensive fibrosis, giant cell transformation of hepatocytes, moderate to severe cholestasis and proliferation of cholangioles	*27*
Kenya	20; **12**	Brief febrile episode, vomiting, abdominal discomfort, anorexia, jaundice, oedema of legs, ascites,tachycardia, tenderness of liver (rarely enlarged), melaena, gastrointestinal bleeding	Maize	Several weeks	Aflatoxin B_1 (2/2) 3.2–12 ppm, aflatoxin B_2 (2/2) 1.6–2.7 ppm	Liver (autopsy)	Aflatoxin B_1 (2/2)	Marked centrilobular necrosis, slight fatty infiltration, and no proliferation of bile ducts	*28*
USA	1; **0**	Non-pruritic macular rash, nausea, headache	Purified aflatoxin B_1	2 days	Aflatoxin B_1 5.5 mg[d]	–	–	Normal	*29*
		Nausea	Purified aflatoxin B_1	2 weeks	Aflatoxin B_1 35 mg[d]	Urine	Aflatoxin M_1 (0/1)[e]	Normal	

[a] Figures in bold are the number of deaths.

[b] Figures in parentheses are number positive/number analysed.

[c] Maize samples taken from the affected families one year after the outbreak.

[d] Total dose.

[e] Three days after ingestion of purified aflatoxin B_1.

insufficiency, thrombosis and embolisms, for the stimulation of cerebral and peripheral metabolism, in uterine stimulation, as a dopaminergic agonist (*8*).

The toxic effects of mycotoxins (e.g. ochratoxins, fumonisins, zearalenone, etc.) are mostly known from veterinary practice. Mycotoxicoses, which can occur in both industrialized and developing countries, arise when environmental, social and economic conditions combine with meteorological conditions (humidity, temperature) which favour the growth of moulds.

Involvement of mycotoxins in disease causation should be considered in instances when a disease appears in several persons, with no obvious connection to a known etiological agent, such as microorganisms. Given current trade patterns, mycotoxicoses resulting from contaminated food, locally grown or imported, could occur in developing and developed countries alike. Strict control of food and feed and appropriate public health measures are therefore of considerable importance in reducing the risks to human and animal health.

This review covers only the human aspects of the untoward effects of mycotoxins. However, owing to the frequent nonspecific effects of mycotoxin involvement, the results of animal experiments are useful for understanding possible effects on humans. Since review articles and books are available dealing with specific topics such as the chemistry, analytical procedures, metabolism, and economic aspects of mycotoxins (*9–18*), these aspects of mycotoxin toxicology are not presented here. Mycotoxicoses are usually insufficiently treated in medical textbooks and are not covered in curricula of many medical schools. The aim of this article is to summarize current understanding of the clinical aspects mainly of mycotoxicoses in humans, and to

stress the importance of this class of naturally occurring toxins.

Ergot

Ergot is the common name of the sclerotia of fungal species within the genus *Claviceps*, which produce ergot alkaloids. The sclerotium is the dark-coloured, hard fungal mass that replaces the seed or kernel of a plant following infestation. Ergot alkaloids are also secondary metabolites of some strains of *Penicillium, Aspergillus* and *Rhizopus* spp. (*8*).

The ca. 40 ergot alkaloids isolated from *Claviceps* sclerotia can be divided into three groups:
– derivatives of lysergic acid (e.g. ergotamine and ergocristine);
– derivatives of isolysergic acid (e.g. ergotaminine);
– derivatives of dimethylergoline (clavines, e.g. agroclavine) (*12*).

The source of the ergot strongly influences the type of alkaloids present, as well as the clinical picture of ergotism (*19*).

Claviceps purpurea produces ergotamine-ergocristine alkaloids, which cause the gangrenous form of ergotism because of their vasoconstrictive activity. The initial symptoms are oedema of the legs, with severe pains. Paraesthesias are followed by gangrene at the tendons, with painless demarcation. The last-recorded outbreak of gangrenous ergotism occurred in Ethiopia in 1977–78; 140 persons were affected and the mortality was high (34%) (*20*).

The other type of ergotism, a convulsive form related to intoxication with clavine alkaloids from *Claviceps fusiformis,* was last seen during 1975 in India when 78 persons were affected (*21, 22*). It was characterized by gastrointestinal symptoms (nausea, vomiting and giddiness) followed by effects on the central nervous system (drowsiness, prolonged sleepiness, twitching, convulsions, blindness and paralysis). The onset of symptoms occurred 1–48 hours following exposure; there were no fatalities.

Ergotism is extremely rare today, primarily because the normal grain cleaning and milling processes remove most of the ergot so that only very low levels of alkaloids remain in the resultant flours. In addition, the alkaloids that are the causative agents of ergotism are relatively labile and are usually destroyed during baking and cooking.

Aflatoxins

Aflatoxins occur in nuts, cereals and rice under conditions of high humidity and temperature and present a risk to human health that is insufficiently recognized. The two major *Aspergillus* species that produce aflatoxins are *A. flavus*, which produces only B aflatoxins, and *A. parasiticus,* which produces both B and G aflatoxins. Aflatoxins M_1 and M_2 are oxidative metabolic products of aflatoxins B_1 and B_2 produced by animals following ingestion, and so appear in milk (both animal and human), urine and faeces. Aflatoxicol is a reductive metabolite of aflatoxin B_1 .

Aflatoxins are acutely toxic, immunosuppressive, mutagenic, teratogenic and carcinogenic compounds. The main target organ for toxicity and carcinogenicity is the liver. The evaluation of epidemiological and laboratory results carried out in 1987 by the International Agency for Research on Cancer (IARC) found that there is sufficient evidence in humans for the carcinogenicity of naturally occurring mixtures of aflatoxins, which are therefore classified as Group 1 carcinogens, except for aflatoxin M_1, which is possibly carcinogenic to humans (Group 2B) (*23*).

Several outbreaks of aflatoxicosis have occurred in tropical countries, mostly among adults in rural populations with a poor level of nutrition for whom maize is the staple food (Table 1). The clinical picture presented by cases indicated acute toxic liver injury, which was confirmed by morphological changes in liver autopsy specimens that were indicative of toxic hepatitis (*27*). Mortality rates in the acute phase were 10–60 %. At the end of one year, surviving patients had no jaundice, and most of them had recovered clinically (*26*).

A case of attempted suicide with purified aflatoxin B_1 is reported to have occurred in 1966 in the USA (*29*). A young woman ingested a total of 5.5 mg of aflatoxin B_1 over 2 days and, 6 months later, a total of 35 mg over 2 weeks. Following the first exposure, she was admitted to hospital with a transient, nonpruritic, macular rash, nausea and headache; the second time she reported nausea only. On both occasions, physical, radiological and laboratory examinations were normal and liver biopsies appeared normal by light microscopy. A follow-up examination 14 years later did not reveal any signs or symptoms of disease or lesions. These findings suggest that the hepatotoxicity of aflatoxin B_1 may be lower in well nourished persons than in experimental animals or that the latent period for tumour formation may exceed 14 years.

Aflatoxins have been detected in the blood of pregnant women, in neonatal umbilical cord blood, and in breast milk in African countries, with significant seasonal variations (*30–32*). Levels of aflatoxins detected in some umbilical cord bloods at birth are among the highest levels ever recorded in human tissue and fluids.

Aflatoxins have been suggested as an etiological factor in encephalopathy and fatty degeneration of viscera, similar to Reye syndrome, which is common in countries with a hot and humid climate (*33*). The clinical picture includes enlarged, pale, fatty liver and kidneys and severe cerebral oedema. Aflatoxins have been found in blood during the acute phase of the disease, and in the liver of affected children (Table 2). However, use of aspirin or phenothiazines is also suspected to be involved in the etiology (*41*).

Table 2. **Presence of aflatoxins in children with Reye syndrome**

Country	No of subjects	Syndrome	Material analysed	Toxin No. of positive samples/no. analysed			Reference
				Aflatoxin B_1	Aflatoxin B_2	Aflatoxin M_1	
Czechoslovakia	27; **27** [a]	Reye syndrome	Liver	26/26 (100)[b]		4/26 (15)	*34*
	25; **25**	Control children[c]	Liver	0/25 (0)		0/25 (0)	*35*
New Zealand	2; **2**	Reye syndrome	Liver	2/2 (100)			*36*
Thailand	23; **23**	Reye syndrome	Brain	13/18 (72)	1/18 (6)	0/18 (0)	*37*
			Liver	17/19 (89)	2/19 (11)	0/19 (0)	
			Kidney	11/14 (79)	0/14 (0)	0/14 (0)	
			Stool	7/17 (41)	4/17 (24)	0/17 (0)	
	15; **15**	Control children[c]	Brain	7/13 (54)	1/13 (8)	0/13 (0)	
			Liver	8/13 (62)	0/13 (0)	0/13 (0)	
			Kidney	6/11 (55)	1/11 (9)	0/11 (0)	
			Stool	3/5 (60)	0/5 (0)	0/5 (0)	
USA	2; **2**	Reye syndrome	Blood	2/2 (100)			*38*
USA	12; **12**	Reye syndrome	Liver	12/12 (100)			*39*
USA	8; **8**	Reye syndrome	Blood	2/5 (40)			*40*
			Liver	6/7 (86)			
	10; **10**	Control children[c]	Liver	1/12 (8)			

[a] Figures in bold are the number of deaths.

[b] Figures in parentheses are percentages.

[c] Control children died with various diseases other than Reye syndrome.

In tropical countries, clinically recognizable jaundice is frequent during the neonatal period. In a large investigation undertaken on 327 babies with jaundice and 80 matching controls in Nigeria, it was found that the occurrence of glucose-6-phosphate dehydrogenase (G6PD) deficiency together with the presence of aflatoxins in the serum are significant risk factors for the development of neonatal jaundice (*42*).

The geographical and seasonal prevalences of aflatoxins in food and of kwashiorkor show a remarkable similarity (*43*). In several tropical countries, aflatoxins have been found more frequently and in higher concentration in liver specimens from children with kwashiorkor than in controls (Table 3). Clinical investigation of aflatoxin elimination in children with kwashiorkor and marasmic kwashiorkor, who were fed an aflatoxin-free diet, proved that aflatoxins in these children are slowly eliminated (*46*). In several studies, aflatoxicol was found in the serum, liver, urine and stools of children with kwashiorkor and marasmic kwashiorkor, in contrast to marasmic and control children where this metabolite was not found. It is not clear whether this difference is causally related to kwashiorkor or is a consequence of the disease.

In recent studies, aflatoxins were found in the brain and lungs of children who had died from kwashiorkor and in control children who had died from various other diseases (*47, 48*). It was suggested that the presence of aflatoxins in the brains of control children might be due to metabolic imbalance or to a failure in the excretory mechanisms of children with conditions such as measles (which in 25% of cases precedes kwashiorkor), renal failure, pyloric stenosis, gastroenteritis. Aflatoxins in the lungs were found in all children diagnosed to have pneumonia, irrespective of the presence of kwashiorkor. This could be due to a reduced clearing ability of the lungs in pulmonary diseases or to exposure via the respiratory route. In the Philippines, a study of the relationship between the presence of aflatoxin in the serum and urine of children and the outcome of acute lower respiratory infection failed to prove a correlation (*50*). However, aflatoxin B_1 was found in the lungs of one textile and two agricultural workers who died from pulmonary interstitial fibrosis (*51*). These individuals were probably occupationally exposed to aflatoxin B_1 via the respiratory route. Aflatoxin B_1 was also detected in the lung tissue of a chemical engineer who had worked for 3 months on a method for sterilizing Brazilian peanut meal contaminated with *Aspergillus flavus,* and who died of alveolar cell carcinoma (*52*).

In the United Kingdom, it was found that intravenous heroin users can be exposed to aflatoxin B_1 from samples of heroin on sale (*53*). Through intravenous administration, aflatoxin B_1 bypasses the detoxifying mechanisms of the liver, which results in direct systemic exposure. In the United Kingdom and the Netherlands, analysis of 121 urine samples obtained from heroin addicts revealed a higher proportion of samples contaminated with aflatoxins B_1, B_2, M_1 and M_2 and aflatoxicol (20%) than those from normal adult volunteers (2%) (*54*). In addition, aflatoxin B_1 was found at much lower concentrations in the latter group.

Table 3. **Presence of aflatoxins in tissues of children with kwashiorkor**

Country	No of subjects	Syndrome	Material analysed[a]	Aflatoxin No. of positive samples/no. analysed								Reference
				Aflatoxins	B_1	B_2	G_1	G_2	M_1	M_2	Aflatoxicol	
Ghana	22; **22**[b]	Kwashiorkor	Liver (a)		20/22 (91)[c]						2/22 (9)	*44*
Ghana, Kenya, Liberia, Sudan, Transkei, Zimbabwe[d]		Kwashiorkor	Serum	136/340 (40)							29/340 (9)	*45*
			Urine	79/239 (33)								
			Liver (a)			46/47 (98)			1/47 (2)		46/47 (98)	
			Liver (b)		5/16 (31)						5/16 (31)	
		Marasmic kwashiorkor	Serum	42/141 (30)							8/141 (6)	
			Urine	47/116 (41)								
			Liver (a)						1/7 (14)		5/7 (71)	
			Liver (b)		1/1						1/1	
		Marasmus	Serum	46/175 (26)							0/175 (0)	
			Urine	52/168 (31)								
			Liver (a)						0/4 (0)		0/4 (0)	
			Liver (b)		0/10 (0)						0/10 (0)	
		Controls[e]	Serum	51/202 (25)							0/202 (0)	
			Urine	98/336 (29)								
			Liver (a)						6/18 (33)			
			Liver (b)		0/27 (0)						0/27 (0)	
Kenya[f]	5; **2**	Kwashiorkor	Urine		2/25 (8)	0/25 (0)			2/25 (8)	1/25 (4)		*46*
			Stool		6/26 (23)	0/26 (0)			2/26 (8)	5/26 (19)	3/26 (12)	
			Liver (a)						0/2 (0)			
	7; **3**	Marasmic kwashiorkor	Urine		4/26 (15)	1/26 (4)			2/26 (8)	4/26 (15)		
			Stool		2/30 (7)	0/30 (0)			3/30(10)	7/30 (23)	0/30 (0)	
			Liver (a)						1/2			
Nigeria	18; **18**	Kwashiorkor	Brain (a)		4/18 (22)	1/18 (5)	4/18 (22)	2/18 (11)	1/18 (11)	3/18 (17)	7/18 (39)	*47*
	19; **19**	Controls[g]	Brain (a)		1/19 (5)	2/19 (10)	5/19 (26)	2/19 (10)	1/19 (5)	6/19 (32)	6/19 (32)	
Nigeria	20; **20**	Kwashiorkor	Lungs (a)		0/20 (0)	0/20 (0)	3/20 (15)	3/20 (15)	0/20 (0)	11/20 (55)	4/20 (20)	*48*
	20; **20**	Controls[g]	Lungs (a)		0/20 (0)	0/20 (0)	6/20 (30)	3/20 (15)	0/20 (0)	10/20 (50)	3/20 (15)	
Sudan	44	Kwashiorkor	Serum	16/44 (36)							6/44 (13)	*49*
			Urine	14/42 (33)								
	32	Marasmic kwashiorkor	Serum	7/32 (22)							4/32 (12)	
			Urine	8/32 (25)								
	70	Marasmus	Serum	11/57 (19)							1/57 (2)	
			Urine	8/70 (26)								
	106	Controls[e]	Serum	7/44 (16)							0/44 (0)	
			Urine	21/106 (20)								

[a] Liver (a), autopsy specimen; liver (b), biopsy specimen.

[b] Figures in bold are the number of deaths.

[c] Figures in parentheses are percentages.

[d] The number of affected children (and fatalities) is not given.

[e] Well nourished children admitted for minor intercurrent illness or for surgery.

[f] Children were fed with an aflatoxin-free diet.

[g] Children died with various diseases other than kwashiorkor.

3-Nitropropionic acid

3-Nitropropionic acid (3-NPA) is a secondary metabolite of *Arthrinium* sp., considered to cause a form of acute food-poisoning called "mouldy sugar-cane poisoning" (*55*). The problem occurred during winter (February and March) in 13 provinces of northern China as a consequence of ingesting sugar-cane that had been stored for at least two months and which was infested with *Arthrinium* sp. In the period 1972–88, a total of 884 persons were involved in outbreaks, with 88 (10%) fatalities (*56*). The main epidemiological feature is the small number of persons in one outbreak (one to five persons), with the victims being mostly children and young people (*56*). The incubation period is generally 2–3 hours following the ingestion of mouldy sugar-cane, and the main clinical symptoms are vomiting, dystonia, staring to one side, convulsions, carpopedal spasm and coma. Delayed dystonia develops in 10–50 % of patients as a consequence of bilateral symmetric necrosis of the basal ganglia. The development of delayed symptoms can be predicted by abnormality in the basal ganglia on cranial computed tomography (CT) scans (*57*). In adults, 3-NPA causes gastro-intestinal symptoms; signs of severe encephalopathy are not common (*58*).

Ochratoxins

Ochratoxins are secondary metabolites of *Aspergillus* and *Penicillium* strains, found on cereals, coffee and bread, as well as on all kinds of food commodities of animal origin in many countries (*59*). The most frequent is ochratoxin A, which is also the most toxic. It has been shown to be nephrotoxic, immunosup-

pressive, carcinogenic and teratogenic in all experimental animals tested so far (*12*).

Acute renal failure in one person, possibly caused by inhalation of ochratoxin A in a granary which had been closed for 2 years, was reported in Italy (*60*). The symptoms developed after 24 hours of transitory epigastric tension, respiratory distress, and retrosternal burning. Acute tubular necrosis was found on biopsy, but the blood was not analysed for ochratoxin A. The presence of the mycotoxin in wheat from the granary was proved qualitatively by thin-layer chromatography.

Owing to the similarity of morphological and functional kidney lesions in ochratoxin A-induced porcine nephropathy and endemic nephropathy, this mycotoxin has been proposed as the causative agent of endemic nephropathy (*61*), although the evidence for this is not substantial. This fatal renal disease occurs among rural populations in Croatia, Bosnia and Herzegovina, Yugoslavia, Bulgaria, and Romania, where it has been estimated that about 20 000 people are either suffering from or are suspected to have the disease (*62*). There is no acute phase of the illness; the first signs and symptoms of the disease are not specific and include fatigue, headache, loss of body weight and pale skin. A mild low-molecular-mass proteinuria without hypertension but with either aplastic or normochromic anaemia gradually develops over several years. The main features of endemic nephropathy are bilateral, primarily chronic lesions of the renal cortex (tubular degeneration, interstitial fibrosis and hyalinization of the glomeruli). In the advanced stage of the disease, the size and weight of kidneys are remarkably reduced, with diffuse cortical fibrosis, usually without signs of inflammation (*63–65*).

Ochratoxin A is found more frequently and in higher concentrations in the blood of inhabitants from endemic regions than control regions (*66, 67*). Many samples of locally produced food and feed collected in the endemic area contained ochratoxin A (*68*). It should be emphasized that the grain analysed had been kept for many months in the inadequate food stores of individual families.

In Tunisia, ochratoxin A has been detected in high concentrations in the blood and food of patients with kidney impairment of unknown etiology (*69, 70*). It has also been found in several countries, both in food and feed (*59*) and in humans (Table 4). So far no cases of endemic nephropathy have been recorded in these countries.

In endemic regions of Croatia, Bulgaria and Yugoslavia, the incidence of otherwise rare urothelial tumours of the pelvis and ureter is 50, 90 and 100 times greater, respectively, than in nonendemic regions (*87–89*). It has been suggested that ochratoxin A may be the causal agent for both endemic nephropathy and urothelial tumours (*90*). IARC classified ochratoxin A as a compound possibly carcinogenic to humans (Group 2B) (*23*).

Trichothecenes

Trichothecenes are mycotoxins produced mostly by members of the *Fusarium* genus, although other genera (e.g. *Trichoderma, Trichothecium, Myrothecium* and *Stachybotrys*) are also known to produce these compounds. To date, 148 trichothecenes have been isolated, but only a few have been found to contaminate food and feed. The most frequent contaminants are deoxynivalenol (DON), also known as vomitoxin, nivalenol (NIV), diacetoxyscirpenol (DAS), while T-2 toxin is rarer (*12*).

Common manifestations of trichothecene toxicity are depression of immune responses and nausea, sometimes vomiting (Table 5). The first recognized trichothecene mycotoxicosis was alimentary toxic aleukia in the USSR in 1932; the mortality rate was 60% (*91*). In regions where the disease occurred, 5–40% of grain samples cultured showed the presence of *Fusarium sporotrichoides,* while in those regions where the disease was absent this fungus was found in only 2–8% of samples. The severity of mycotoxicosis was related to the duration of consumption of toxic grain. Such severe trichothecene mycotoxicoses, the consequence of continuous ingestion of toxins, have not been recorded since this outbreak.

In several cases, trichothecene mycotoxicosis was caused by a single ingestion of bread containing toxic flour (*95*) or rice (*92, 97*).

In experimental animals, trichothecenes are 40 times more toxic when inhaled than when given orally (*98*). Trichothecenes were found in air samples collected during the drying and milling process on farms (*99*), in the ventilation systems of private houses (*100*) and office buildings (*98*), and on the walls of houses with high humidity (*100, 101*) (Table 6). There are some reports showing trichothecene involvement in the development of "sick building syndrome" (*98, 100*). The symptoms of airborne toxicosis disappeared when the buildings and ventilation systems were thoroughly cleaned (*100*).

There are some reports that indicate that trichothecenes may have been used as chemical warfare agents in South-East Asia (Lao People's Democratic Republic and Cambodia) (*102, 103*).

Zearalenone

Zearalenone (previously known as F-2) is produced mainly by *Fusarium graminearum* and related species, principally in wheat and maize but also in sorghum, barley and compounded feeds. Zearalenone and its derivatives produce estrogenic effects in various animal species (infertility, vulval oedema, vaginal prolapse and mammary hypertrophy in females and feminization of males — atrophy of testes and enlargement of mammary glands).

In Puerto Rico, zearalenone was found in the blood of children with precocious sexual development (*104*) exposed to contaminated food. Zearalenone was also found together with other *Fusarium* mycotoxins in

Table 4. **Occurrence of ochratoxin A in human blood samples**[a]

Country	Year	Incidence of positive samples	Mean concentration (ng/ml)	Concentration range of positive samples (ng/ml)	Reference
Bulgaria	1984–90	9/125 (7)[b]		1.0–10.0	*67*
Canada	1994	144/144 (100)	0.88	0.29–2.37	*71*
Croatia	1997				*72*
Zagreb		29/50 (58)	0.26	0.20–1.28	
Rijeka		18/50 (36)	0.17	0.20–0.82	
Osijek		50/50 (100)	0.68	0.20–1.65	
Split		27/49 (55)	0.25	0.20–1.39	
Varazdin		24/50 (48)	0.59	0.20–15.9	
Czechoslovakia	1990	35/143 (24)	0.14	0.10–12.6	*73*
Czech Republic	1994	734/809 (91)	0.23	0.10–13.7	*74*
	1995	404/413 (98)	0.24	0.10–1.9	
Denmark	1986–88	78/144 (54)	1.8	0.10–13.2	*75*
France	1991–92				*76*
Alsace		97/500 (19)		0.10–12	
Aquitaine		385/2055 (19)		0.10–160	
Rhône-Alpes		75/515 (15)		0.10–4	
Federal Republic of Germany	1977	84/164 (51)	0.4	0.1–4	*77*
	1985	89/141 (63)	0.3	0.1–2	*77*
	1988	142/208 (68)	0.75	0.1–8	*78*
Hungary	1995	291/355 (82)		0.2–10	*79*
	1997	213/277 (77)		0.1–1.4	*80*
Italy	1992	65/65 (100)	0.5	0.1–2	*81*
Japan, Tokyo	1992–96	156/184 (85)	0.068[c]	0.004–0.278	*82*
Poland	1983–84	25/397 (6)	0.2	1–13	*83*
	1984–85	52/668 (8)	0.3	1–40	
Sierra Leone	1996	12/36 (33)[d]		1.5–18	*84*
Sweden	1989				*85*
Visby		29/99 (29)	0.26	0.3–7	
Uppsala		3/99 (3)	0.02	0.3–0.8	
Ostersund		6/99 (6)	0.03	0.3–0.8	
Switzerland	1992–93				*86*
North of the Alps		251/252 (100)		0.06–2.14	
South of the Alps		116/116 (100)		0.11–0.75	
Tunisia	1993–95	73/140 (52)		0.1–8.8	*69, 70*

[a] Mean values were calculated for all samples analysed, including those with no detectable concentrations (considered as zero).

[b] Figures in parentheses are percentages.

[c] Mean of positives.

[d] Non-breastfed infants (up to 5 years of age).

"scabby grain toxicosis" in China (Table 5), but the significance of this finding is not clear.

Fumonisins

Fumonisins are mycotoxins produced throughout the world by *Fusarium moniliforme* and related species when they grow in maize. Fumonisins B_1 and B_2 are of toxicological significance, while the others (B_3, B_4, A_1 and A_2) occur in very low concentrations and are less toxic.

In India a single outbreak of acute foodborne disease possibly caused by fumonisin B_1 has been reported (*105*). In the 27 villages involved, the

Table 5. **Outbreaks of trichothecene mycotoxicoses with oral exposure**

Country	Year	No. of subjects affected/ exposed	Clinical symptoms and signs	Onset of symptoms/ recovery	Source of exposure	Toxic fungal species	Toxin	Reference
USSR	1932–47	—[a]	''Alimentary toxic aleukia''		Grain	*Fusarium sporotrichoides*		*91*
			1. Hyperaemia of mucous membranes of oral cavity and pharynx, gastritis, gastroenteritis, excessive salivation, abdominal and oesophageal pain and diarrhoea	A few hours/ 2–3 days		*Fusarium poae*		*12*
			2. Generalized indisposition, vertigo, unpleasant taste in mouth, progressive leukopenia, granulocytopenia and lymphocytosis	3–4 weeks				
			3. Haemorrhagic diathesis and angina, petechial rash, catarrhal diphtheritic, gangrenous pharyngitis, ulcerative and gangrenous laryngitis, aphonia, asphyxia	few days/ 2 weeks				
Japan	1956	25; **0**[b]	''Scabby grain toxicosis'' – nausea, vomiting, drowsiness		Rice	*Fusarium roseum* *Fusarium nivele*		*92*
China	1961–1985	7818/[c]; **0**	''Scabby grain toxicosis'' – nausea, vomiting, abdominal pain diarrhoea, dizziness, headache		Corn Wheat	*Fusarium sp.*	Deoxynivalenol Zearalenone	*93*
China	1984–1985	463/600; **0**	Nausea, vomiting, abdominal pain, diarrhoea, dizziness, headache	5–30 min	Corn Wheat	*Fusarium sp.*	Deoxynivalenol Zearalenone	*94*
India	1987	97/224; **0**	Mild to moderate abdominal pain, feeling of fullness, irritation of throat, diarrhoea, blood in stools	15 min–1hour/ 1–2 days	Wheat	*Fusarium sp.* *Aspergillus flavus*	Nivalenol Deoxynivalenol T-2 Acetyldeoxynivalenol	*95* *96*
China[d]		97/165; **0**	Nausea, vomiting, chills, abdominal pain, thoracic stuffiness, diarrhoea	0–30 min	Rice	*Fusarium heterosporum* *Fusarium graminearum*	T-2	*97*

[a] Tens of thousands of persons were involved, with a mortality rate of 60%.

[b] Figures in bold are the number of deaths.

[c] The number of exposed persons is not given.

[d] The year of the outbreak is not given.

individuals affected were from the poorest social strata, who had consumed maize and sorghum harvested and left in the fields during unseasonable rains. The main features of the disease were transient abdominal pain, borborygmus and diarrhoea, which began half an hour to one hour following consumption of unleavened bread prepared from mouldy sorghum or mouldy maize. Patients recovered fully when the exposure ceased and there were no fatalities. Fumonisin B_1 was found in much higher concentrations in the maize and sorghum from the affected households than from controls.

Fumonisin B_1 was found more frequently and in much higher concentrations in maize in regions of Transkei (*106, 107*), China (*108*) and north-east Italy (*109*) with a higher incidence of oesophageal cancer than other regions. It was postulated that the high incidence of oesophageal cancer was related to the presence of this mycotoxin in maize, which is a staple food in these regions. The incidence and concentration of aflatoxin B_1, deoxynivalenol and fumonisins B_1, B_2 and B_3 were recently determined in maize samples from an area of China (Haimen) with a high incidence of primary liver cancer and from an area with a low incidence (Penlai) (*110*). Aflatoxin B_1 was found in low concentrations in almost all maize samples from both these areas, but the incidence and concentration of deoxynivalenol and fumonisins were much higher in the samples from the area where the incidence of primary liver cancer was high. The authors put forward the hypothesis that fumonisins, which have known cancer-promoting activity in rat liver (*111*), and deoxynivalenol promote the initial lesion caused by aflatoxin B_1.

An IARC working group classified the toxins from *F. moniliforme* as possibly carcinogenic to humans (Group 2B) (*23*).

Other unidentified mycotoxins

The impact of other mycotoxins on human health was reported in persons occupationally exposed to large amounts of different mycotoxin-producing fungi (farmers, workers in silos, etc.). In such cases, exposure to spores via the respiratory tract seems to be of considerable importance.

In Norway an extensive epidemiological study was undertaken between 1967 and 1991 on 192 417 births (*112*) to test the hypotheses that perinatal death was associated with parental exposure

Table 6. **Trichothecene mycotoxicoses after inhalation or/and dermal exposure**

Country	Year	No. of subjects affected/ exposed	Clinical symptoms and signs	Source of exposure	Toxic fungal species	Toxin	Reference
USA	1981–86	4; **0**[a]	''Sick-building syndrome'' recurring maladies, cold and flu-like symptoms, sore throats, diarrhoea, headache, fatigue, dermatitis, intermittent focal alopecia, generalized malaise	Walls and ceiling fibre boards	*Stachybotrys atra*	Verrucarol Verrucarins B and J Satratoxin H Trichoverrins A and B	*100*
Canada[b]			''Sick-building syndrome'' headache, chronic fatigue, cold and flu-like symptoms, dermal irritation	Dust from ventilation system		T-2 Diacetoxyscirpenol Roridin T-2 tetraol	*98*
Lao People's Democratic Republic	1975–81[b]	**>6500**[c]	''Yellow rain''				
Cambodia	1978–81[b]	**>1000**	Vomiting, diarrhoea, haemorrhage, breathing difficulty, itching, chest pain, irritation, of the skin, blisters on exposed skin, nausea, blurred vision, headache, fatigue, dizziness, vertigo	Aircraft spray			*102*
				Leaf	—	T-2 Deoxynivalenol Nivalenol	*103*
				Water	—	Deoxynivalenol	
				Yellow-green powder		T-2 Zearalenone Diacetoxyscirpenol	

[a] Figures in bold are the number of deaths.

[b] The number of affected and exposed persons is not given.

[c] Autopsy findings: necrosis of the lining of the stomach and upper small intestine, congestion of lungs, liver, spleen and sometimes of the kidneys.

to pesticides, *Toxoplasma gondii* infection from sheep or pigs, or mycotoxins found in grain. The proportion of late-term abortions (gestational age 16–27 weeks) was higher among farmers. The risk associated with grain farming was higher after the harvest, in seasons with a poor quality harvest and in pregnancies with multiple fetuses, which suggests that mycotoxins in grain induce labour at an early stage of pregnancy.

Pulmonary mycotoxicosis has been reported in ten persons exposed to large quantities of fungal hyphae and spores during the cleaning of silos (*113*). The clinical picture developed several hours afterwards, with burning eyes, throat and chest, irritating cough and fever. There was no wheezing, cyanosis or other sign of bronchospasm. In five patients, chest X-rays revealed reticular and fine nodular features compatible with interstitial pneumonitis. Histological study of a lung biopsy from one patient showed a multifocal acute process, with primary involvement of terminal bronchioles containing numbers of various spores. Cultures from lung biopsy material revealed at least five fungal species, including one *Fusarium* and one *Penicillium*. However, blood samples were not checked for the presence of mycotoxins. In contrast with the findings in patients with farmer's lung disease, these patients did not develop positive serological reactions to thermophilic actinomycetes or to extracts of fungi obtained from hay or silage. The patients were followed for periods of 1–10 years; they continued their work, avoiding massive re-exposure to fungal dust, and during the observation period there were no further incidents.

Conclusion

Acute mycotoxicoses can cause serious and sometimes fatal diseases. The possibility of mycotoxin intoxication should be considered when an acute disease occurs in several persons when there is no evidence of infection with a known etiological agent, and no improvement in the clinical picture following treatment. Most of the outbreaks of mycotoxicoses described are a consequence of the ingestion of food that is contaminated with mycotoxins. The strict control of food quality, in both industrialized and developing countries, is therefore necessary to avoid such outbreaks. ■

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Résumé

Les effets toxiques des mycotoxines chez l'homme

Les mycotoxicoses sont des maladies provoquées par des mycotoxines, c'est-à-dire des métabolites secondaires des moisissures. Les moisissures se développent plus volontiers dans des zones de climat chaud et humide, mais on en trouve aussi dans les régions tempérées. L'exposition aux mycotoxines a essentiellement lieu par ingestion, mais elle peut se produire également par la voie percutanée ou respiratoire. Elles échappent souvent à l'attention des médecins, sauf lorsque le nombre de personnes touchées est élevé. Le présent article passe en revue les flambées de mycotoxicoses dans lesquelles l'étiologie mycotoxinique est corroborée par une analyse mycotoxicologique ou par l'identification du champignon en cause. Les auteurs discutent les résultats des observations épidémiologiques, cliniques et histologiques effectuées à l'occasion de flambées d'intoxications dues à des aflatoxines, à l'ergot de seigle, aux trichotécènes, aux ochratoxines, à l'acide 3-nitropropionique, à la zéaralénone et aux fumonisines.

Une mycotoxicose aiguë peut revêtir une forme grave, voire mortelle. Il faut évoquer la possibilité d'une mycotoxicose devant un tableau d'intoxication aiguë impliquant plusieurs sujets ne présentant pas de signes d'infection par un agent étiologique connu et chez qui le traitement n'apporte aucune amélioration clinique. La plupart des flambées de mycotoxicoses qui ont été décrites résultent de l'ingestion de denrées alimentaires contaminées par des mycotoxines. Dans les pays développés comme dans les pays en développement, la prévention de ces intoxications repose donc sur un contrôle rigoureux de la qualité des denrées alimentaires.

Resumen

Efectos tóxicos de las micotoxinas en el ser humano

Las micotoxicosis son enfermedades causadas por micotoxinas, metabolitos secundarios de los mohos. Aunque se producen con más frecuencia en las regiones con clima cálido y húmedo, propicio para el crecimiento de los mohos, también se dan en zonas templadas. La exposición a las micotoxinas se produce sobre todo por ingestión, pero también por contacto cutáneo y por inhalación. A menudo los profesionales de la medicina no reconocen las micotoxicosis, salvo cuando afectan a gran número de personas. En el presente artículo se examinan diversos brotes de micotoxicosis en los que la etiología de la enfermedad se ha visto corroborada por el análisis de la micotoxina o la identificación de los hongos que la producen. Se analizan los hallazgos epidemiológicos, clínicos e histológicos disponibles en relación con los brotes de micotoxicosis causados por la exposición a las aflatoxinas, el cornezuelo del centeno, las tricotecenas, las ocratoxinas, el ácido 3-nitropropiónico, la zearalenona y las fumonisinas.

Las micotoxicosis agudas pueden provocar manifestaciones graves, a veces mortales. Se debe sospechar una posible intoxicación por micotoxinas cuando una enfermedad aguda afecta a varias personas y no existen signos ni de infección por un agente etiológico conocido ni de mejora del cuadro clínico tras el tratamiento. La mayoría de los brotes de micotoxicosis descritos se deben a la ingestión de alimentos contaminados por micotoxinas. Así pues, para evitar dichos brotes es necesario un control estricto de la calidad de los alimentos, tanto en los países desarrollados como en los países en desarrollo.

References

1. **Vergilius PM.** *Georgike.* Velika Gorica, Papir, 1994.
2. **Ovidius PN.** *Opera omnia. III Fasti, Tristia, Epistolae ex ponto.* Leipzig, Nova editio stereotypa, 1845.
3. **Van Dongen PWJ, De Groot ANJA.** History of ergot alkaloids from ergotism to ergometrine. *European journal of obstetrics, gynecology and reproductive biology*, 1995, **60**: 109–116.
4. **Schneider DJ et al.** First report of field outbreaks of ergot-alkaloid toxicity in South Africa. *Onderstepoort journal of veterinary research,* 1996, **63**: 97–108.
5. **Uraguchi K**. Mycotoxic origin of cardiac beriberi. *Journal of stored products research,* 1969, **5**: 227–236.
6. **Ueno Y.** The toxicology of mycotoxins. *Critical reviews in toxicology*, 1985, **14**: 99–132.
7. **Reiss J.** Effects of mycotoxins on higher plants, algae, fungi and bacteria. In: Wyllie T, Morehouse L, eds. *Mycotoxic fungi, mycotoxins, mycotoxicoses*, vol. 3. New York, Marcel Dekker, 1978: 118–144.
8. **Flieger M, Wurst M, Shelby R.** Ergot alkaloids – sources, structures and analytical methods. *Folia microbiologica,* 1997, **42**: 3–30.
9. **Purchase IFH**. *Mycotoxins in human health.* Edinburgh, MacMillan, 1971.
10. **Cole RJ.** *Modern methods in the analysis and structural elucidation of mycotoxins.* Orlando, FL, Academic Press, 1986.
11. **Kiessling KH.** Biochemical mechanism of action of mycotoxins. *Pure and applied chemistry*, 1986, **58**: 327–338.
12. *Selected mycotoxins: ochratoxins, trichothecenes, ergot. Report of an Expert Committee.* Geneva, World Health Organization, 1990 (Environmental Health Criteria No. 105).
13. **Scott PM**. Mycotoxin methodology. *Food additives and contaminants*, 1995, **12**: 395–403.
14. **Van Egmond HP**, Mycotoxins: regulations, quality assurance and reference materials. *Food additives and contaminants*, 1995, **12**: 321–330.
15. **Dutton MF**. Fumonisins, mycotoxins of increasing importance: their nature and their effects. *Pharmacology and therapy*, 1996, **70**: 137–161.
16. **Galtier P.** Biological fate of mycotoxins in animals. *Revue de médecine vétérinaire*, 1998, **49**: 49–54.
17. **Pittet A**. Natural occurrence of mycotoxins in foods and feeds — an updated review. *Revue de médecine vétérinaire*, 1998, **49**: 79–92.
18. **Steyn PS.** The biosynthesis of mycotoxins. *Revue de médecine vétérinaire*, 1998, **49:** 469–478.

19. **Burfening PJ.** Ergotism. *Journal of the American Veterinary Medical Association,* 1973, **163**: 1288–1290.
20. **King B.** Outbreak of ergotism in Wollo, Ethiopia. *Lancet,* 1979, 1411.
21. **Krishnamachari KAVR, Bhat RV.** Poisoning of ergoty bajra (pearl millet) in man. *Indian journal of medical research*, 1976, **64**: 1624–1628.
22. **Tulpule PG, Bhat RV.** Food toxins and their implication in human health. *Indian journal of medical research*, 1978, **68** (suppl.): 99–108.
23. *Overall evaluations of carcinogenicity: an updating of IARC Monographs volumes 1 to 42. Report of an IARC Expert Committee.* Lyon, International Agency for Research on Cancer, 1987 (IARC Monographs on the Evaluation of Carcinogenic Risks to Humans, Supplement 7).
24. **Serck-Hanssen A.** Aflatoxin-induced fatal hepatitis? A case report from Uganda. *Archives of environmental health,* 1970, **20**: 729–731.
25. **Krishnamachari KAVR et al.** Hepatitis due to aflatoxicosis. *Lancet*, 1975, 1061–1063.
26. **Bhat RV, Krishnamachari KAVR**. Follow-up study of aflatoxic hepatitis in parts of western India. *Indian journal of medical research,* 1977, **66**: 55–58.
27. **Tandon BN et al.** Study of an epidemic of jaundice, presumably due to toxic hepatitis, in Northwest India. *Gastroenterology,* 1977, **72**: 488–494.
28. **Ngindu A et al.** Outbreak of acute hepatitis caused by aflatoxin poisoning in Kenya. *Lancet*, 1982, 1346–1348.
29. **Willis RM, Mulvihill JJ, Hoofnagle JH.** Attempted suicide with purified aflatoxin. *Lancet,* 1980, 1198–1199.
30. **Coulter JBS et al.** Aflatoxins in human breast milk. *Annals of tropical pediatrics*, 1984, **4**: 61–66.
31. **Lamplugh et al.** Aflatoxins in breast milk, neonatal cord blood, and serum of pregnant women. *British medical journal*, 1988, **296**: 968.
32. **Maxwell SM et al.** Aflatoxin in breast milk, neonatal cord blood and sera of pregnant women. *Toxicology,* 1989, **8**: 19–29.
33. **Olson LC et al.** Encephalopathy and fatty degeneration of viscera in Northeastern Thailand. Clinical syndrome and epidemiology. *Pediatrics,* 1971, **47**: 707–716.
34. **Dvorackova I et al**. Aflatoxin and encephalopathy with fatty degeneration of viscera (Reye). *Annales de la nutrition et de l'alimentation*, 1977, **31**: 977–990.
35. **Dvorackova I, Vesely D, Kusak V.** [Aflatoxins as pathological agents in young children]. *Československa pediatrie*, 1979, **34:** 80–83 (in Czech).
36. **Becroft DMO, Webster DR.** Aflatoxins and Reye's disease. *Lancet,* 1972, 117.
37. **Shank RC et al.** Aflatoxins in autopsy specimens from Thai children with an acute disease of unknown aetiology. *Food and cosmetics toxicology*, 1971, **9**: 501–507.
38. **Hogan GR, Ryan NJ, Hayes AW.** Aflatoxin B_1 and Reye's syndrome. *Lancet*, 1978, 561.
39. **Hayes AW.** Aflatoxin B_1 — its role in the etiology of Reye's syndrome. *Chemische Rundschau,* 1979, **32**: G 711.
40. **Ryan NJ et al.** Aflatoxin B1: its role in the etiology of Reye's syndrome. *Pediatrics*, 1979, **64**: 71–75.
41. **Casteels-Van Daele M, Eggermont E.** Reye's syndrome. *British medical journal*, 1994, **308**: 919–920.
42. **Sodeinde O et al.** Neonatal jaundice, aflatoxins and naphthols: report of a study in Ibadan, Nigeria. *Annals of tropical paediatrics,* 1995, **15**: 107–113.
43. **Hendrickse RG.** Kwashiorkor: the hypothesis that incriminates aflatoxins. *Pediatrics,* 1991, 88: 376–379.
44. **Apeagyei F et al.** Aflatoxins in the livers of children with kwashiorkor in Ghana. *Tropical and geographical medicine,* 1986, **38**: 273–276.
45. **Hendrickse RG, Maxwell SM.** Aflatoxins and child health in tropics. *Journal of toxicology — toxin reviews,* 1989, **8**: 31–41.
46. **De Vries HR et al.** Aflatoxin excretion in children with kwashiorkor or marasmic kwashiorkor — a clinical investigation *Mycopathologia*, 1990, **110:** 1–9.
47. **Oyelami OA et al.** Aflatoxins in the autopsy brain tissue of children in Nigeria. *Mycopathologia*, 1995, **132**: 35–38.
48. **Oyelami OA et al.** Aflatoxins in the lungs of children with kwashiorkor and children with miscellaneous diseases in Nigeria. *Journal of toxicology and environmental health*, 1997, **51**: 623–628.
49. **Hendrickse RG et al.** Aflatoxins and kwashiorkor: a study in Sudanese children. *British medical journal*, 1982, **285**: 843–846.
50. **Denning DW et al.** Aflatoxin and outcome from acute lower respiratory infection in children in The Philippines. *Annals of tropical paediatrics,* 1995, **15**: 209–216.
51. **Dvorackova I, Pichova V.** Pulmonary interstitial fibrosis with the evidence of aflatoxin B_1 in lung tissue. *Journal of toxicology and environmental health,* 1986, **18**: 153–157.
52. **Dvorackova I.** Aflatoxin inhalation and alveolar cell carcinoma. *British medical journal,* 1976, 691.
53. **Hendrickse RG, Maxwell SM.** Heroin addicts, AIDS, and aflatoxins. *British medical journal*, 1989, **296**: 1257.
54. **Hendrickse RG, Maxwell SM, Young R**. Aflatoxins and heroin. *Toxicology,* 1989, **8**: 89–94.
55. **Liu X et al.** Arthrinium sp. and the deteriorated sugarcane poisoning.
In: Aibara et al., eds. *Mycotoxins and phycotoxins. Abstracts of the Seventh International IUPAC Symposium, Tokyo, 16–19 August 1988.* Tokyo, Japanese Association of Mycotoxicology, 1988: 26.
56. **Liu X, Luo X, Hu W.** Studies on epidemiology and etiology of moldy sugarcane poisoning in China. *Biomedical and environmental sciences*, 1992, **5**: 161–177.
57. **Ming L.** Moldy sugarcane poisoning — a case report with a brief review. *Clinical toxicology*, 1995, **33**: 363–367.
58. **Ludolph AC et al.** 3-nitropropionic acid — exogenous animal neurotoxin and possible human striatal toxin. *Canadian journal of neurological sciences*, 1991, **18**: 492–498.
59. **Speijers GJA, Van Egmond HP.** Worldwide ochratoxin A levels in food and feeds. In: Creppy EE et al., eds. *Human ochratoxicosis and its pathologies*. Montrouge, Colloque INSERM/John Libbey Eurotext Ltd., 1993, **231**: 85–100**.**
60. **Di Paolo N et al.** Inhaled mycotoxins lead to acute renal failure. *Nephrology, dialysis and transplantation*, 1994, **9** (suppl. 4): 116–120.
61. **Krogh P.** Mycotoxic porcine nephropathy: a possible model for Balkan endemic nephropathy.
In: Puchlev E, ed. *Endemic nephropathy. Proceedings of the Second International Symposium on Endemic Nephropathy, Sofia, 9–11 November 1972*. Sofia, Publishing House of Bulgarian Academy of Sciences, 1974: 266–270.
62. **Pleština R.** Some features of Balkan endemic nephropathy. *Food and chemical toxicology*, 1992, **30**: 177–181.
63. **Radonić M, Radošević Ž, Zupanić V.** Endemic nephropathy in Yugoslavia. In: Mostovi FK, Smith DE, eds. *The kidney.* Baltimore, Williams & Wilkins, 1966: 503–522.
64. **Heptinstall RH.** *Pathology of the kidney*, vol. 11. Boston, MA, Little Brown & Co, 1974: 828–836.
65. **Vukelić M, Šoštarić B, Belicza M.** Pathomorphology of Balkan endemic nephropathy. *Food and chemical toxicology*, 1992, **30**: 193–200.
66. **Radić B et al.** Ochratoxin A in human sera in the area with endemic nephropathy in Croatia. *Toxicology letters*, 1997, **91**: 105–109.
67. **Petkova-Bocharova T, Castegnaro M.** Ochratoxin A in human blood in relation to Balkan endemic nephropathy and urinary tract tumours in Bulgaria. In: Castegnaro M et al., eds. *Mycotoxins, endemic nephropathy and urinary tract tumours.* Lyon, International Agency for Research on Cancer, 1991: 135–137 (IARC Scientific Publications No. 115).

68. **Pavlović M, Pleština R, Krogh P.** Ochratoxin A contamination of foodstuffs in an area with Balkan (Endemic) nephropathy. *Acta pathologica microbiologica scandinavica, section B*, 1979, **87**: 243–246.
69. **Maaroufi K et al.** Ochratoxin A in human blood in relation to nephropathy in Tunisia. *Human and experimental toxicology*, 1995, **14**: 609–615.
70. **Maaroufi K et al.** Foodstuffs and human blood contamination by the mycotoxin ochratoxin A: correlation with chronic interstitial nephropathy in Tunisia. *Archives of toxicology,* 1995, **69**: 552–558.
71. **Scott PM et al.** Survey of Canadian human blood plasma for ochratoxin A. *Food additives and contaminants*, 1998, **15**: 555–562.
72. **Domijan AM et al.** Plasma ochratoxin A levels in population of Croatia. In: *Abstracts of the Third Croatian Congress of Food Technologists, Biotechnologists and Nutritionists, Zagreb, 10–12 June 1998*. Zagreb, University of Zagreb, 1998: 208**.**
73. **Fukal L, Reisnerova H.** Monitoring of aflatoxins and ochratoxin A in Czechoslovak human sera by immunoassay. *Bulletin of environmental contamination and toxicology,* 1990, **44**: 345–349.
74. **Malir F et al.** The level of ochratoxin A in blood serum of adults in the Czech Republic. *Revue de médecine vétérinaire*, 1998, **149**: 710.
75. **Hald B.** Ochratoxin A in human blood in European countries. In: Castegnaro M. et al., eds. *Mycotoxins, endemic nephropathy and urinary tract tumours.* Lyon*,* International Agency for Research on Cancer, 1991: 159–164 (IARC Scientific Publications No. 115).
76. **Creppy EE et al.** Etude de l'ochratoxicose humaine dans trois regions de France: Alsace, Aquitaine et region Rhone-Alpes. In: Creppy EE et al., eds. *Human ochratoxicosis and its pathologies.* Montrouge, Colloque INSERM/John Libbey Eurotext Ltd., 1993, **231**: 147–158.
77. **Bauer J, Gareis M.** [Ochratoxin A in the food chain]. *Journal of veterinary medicine, B*, 1987, **34**: 613–627 (in German).
78. **Hadlok RM.** Human ochratoxicosis in Germany updating 1993. In: Creppy EE et al., eds. *Human ochratoxicosis and its pathologies*. Montrouge, Colloque INSERM/John Libbey Eurotext Ltd. 1993: **231**, 141–145**.**
79. **Solti L et al.** Ochratoxin A content of human sera determined by a sensitive ELISA. *Journal of analytical toxicology*, 1997, **21**: 44–48.
80. **Tapai K, Teren J, Mesterhazy, A.** Ochratoxin A in the sera of blood donors and ill persons. *Cereal research communications*, 1997, **25**: 307–308.
81. **Breitholtz-Emanuelsson A et al.** Ochratoxin A in human serum samples collected in southern Italy from healthy individuals and individuals suffering from different kidney disorders. *Natural toxins,* 1994, **2**: 366–370.
82. **Ueno Y et al.** A 4-year study of plasma ochratoxin A in a selected population in Tokyo by immunoassay and immunoaffinity column-linked HPLC. *Food and chemical toxicology*, 1998, **36**: 445–449.
83. **Golinski P.** Ochratoxin A in human organism as a result of food and feed contamination. *Roczniki akademii rolniczej w Poznaniu,* 1987, **168** 1–61.
84. **Jonsyn FE.** The intake of aflatoxins and ochratoxin A by infants in Sierra Leone. In: Miraglia M et al., eds. *Abstracts of the Ninth International IUPAC Symposium on Mycotoxins and Phycotoxins, Rome, Italy, 27–31 May 1996.* Rome, Istituto Superiore di Sanita, 1996: 209.
85. **Breitholtz A et al.** Plasma ochratoxin A levels in three Swedish populations surveyed using an ion-pair HPLC technique. *Food additives and contaminants,* 1991, **8**: 183–192.
86. **Zimmerli B, Dick R.** Determination of ochratoxin A at the ppt level in human blood, serum, milk and some foodstuffs by high-performance liquid chromatography with enhanced fluorescence detection and immunoaffinity column cleanup: methodology and Swiss data. *Journal of chromatography B: biomedical applications,* 1995, **666**: 85–99.
87. **Čević S et al.** Epidemiological aspect of Balkan endemic nephropathy in a typical focus in Yugoslavia. In: Castegnaro M et al., eds. *Mycotoxins, endemic nephropathy and urinary tract tumours,* Lyon, International Agency for Research on Cancer, 1991: 5–10 (IARC Scientific Publications No. 115).
88. **Chernozemsky IN.** Balkan endemic nephropathy and the associated tumours of the urinary system: a summary of epidemiological features in Bulgaria. In: Castegnaro M et al., eds. *Mycotoxins, endemic nephropathy and urinary tract tumours*, Lyon, International Agency for Research on Cancer, 1991: 3–4 (IARC Scientific Publications No. 115).
89. **Radovanović Z.** Epidemiological characteristics of Balkan endemic nephropathy in eastern regions of Yugoslavia. In: Castegnaro M et al., eds. *Mycotoxins*, *endemic nephropathy and urinary tract tumours.* Lyon, International Agency for Research on Cancer, 1991: 11–20 (IARC Scientific Publications No. 115).
90. **Castegnaro M et al.** Are mycotoxins risk factors for endemic nephropathy and associated urothelial cancers? *Archiv für Geschwulstforschung,* 1990, **60:** 295–303.
91. **Gajdušek DC.** Acute infectious hemorrhagic fevers and mycotoxicoses in the Union of Soviet Socialist Republics. *Medical science publication No. 2*, Walter Reed Army Medical Center, Washington, 1953**.**
92. **Ueno Y.** Toxicological and biological properties of fusarenon-x**,** a cytotoxic mycotoxin of *Fusarium nivale* Fn-2B. In: Purchase IFH, ed. *Mycotoxins in human health. Proceedings of a Symposium, Pretoria, 2–4 September 1970*. Edinburgh, MacMillan, 1971: 163–178.
93. **Luo XY.** *Fusarium* toxins contamination of cereals in China. In: Aibara et al., eds. *Proceedings of the Seventh International IUPAC Symposium on Mycotoxins and Phycotoxins, Tokyo, 16–19 August 1988*. Tokyo, Japanese Association of Mycotoxicology, 1988: 97–98.
94. **Luo X.** Food poisoning associated with *Fusarium* toxins. In: Aibara et al., eds. *Proceedings of the Seventh International IUPAC Symposium on Mycotoxins and Phycotoxins, Tokyo, 16–19 August 1988*. Tokyo, Japanese Association of Mycotoxicology, 1988: 93.
95. **Bhat RV et al.** Outbreak of trichothecene mycotoxicosis associated with consumption of mould-damaged wheat products in Kashmir Valley, India. *Lancet*, 1989, 35–37.
96. **Ramakrishna Y, Bhat RV, Ravindranath V.** Production of deoxynivalenol by *Fusarium* isolates from samples of wheat associated with a human mycotoxicosis outbreak and from sorghum cultivars. *Applied and environmental microbiology,* 1989, **55**: 2619–2620.
97. **Wang ZG, Feng JN, Tong Z.** Human toxicosis caused by mouldy rice contaminated with *Fusarium* and T-2 toxin. *Biomedical and environmental sciences*, 1993, **6**: 65–70.
98. **Smoragiewicz W et al.** Trichothecene mycotoxins in the dust of ventilation systems in office buildings. *International archives of occupational and environmental health*, 1993, **65:** 113–117.
99. **Lappalainen S et al.** *Fusarium* toxins and fungi associated with handling of grain on eight Finnish farms. *Atmospheric environment*, 1996, **30**: 3059–3065.
100. **Croft WA et al.** Airborne outbreak of trichothecene toxicosis. *Atmospheric environment,* 1986, **20**: 549–552.
101. **Nikulin M et al.** *Stachybotrys atra* growth and toxin production in some building materials and fodder under different relative humidities. *Applied and environmental microbiology,* 1994, **60**: 3421–3424.

102. **Mirocha CJ et al.** Analysis for *Fusarium* toxins in various samples implicated in biological warfare in Southeast Asia. *Journal of the Association of Official Analytical Chemists,* 1983, **66**: 1485–1499.
103. **Watson SA, Mirocha CJ, Hayes AW.** Analysis for trichothecenes in samples from Southeast Asia associated with ''yellow rain''. *Fundamental and applied toxicology,* 1984, **4**: 700–717.
104. **Saenz de Rodriguez CA.** Environmental hormone contamination in Puerto Rico. *New England journal of medicine*, 1984, **310**: 1741–1742.
105. **Bhat RV et al.** A foodborne disease outbreak due to the consumption of moldy sorghum and maize containing fumonisin mycotoxins. *Clinical toxicology,* 1997, **35**: 249–255.
106. **Marasas WFO et al.** Mycoflora of corn produced in human oesophageal cancer areas in Transkei, Southern Africa. *Phytopathology,* 1981, **71**: 792–796.
107. **Jaskiewicz K, Marasas WFO, Van der Walt FE.** Oesophageal and other main cancer patterns in four districts of Transkei, 1981–84. *South African medical journal,* 1987, **72**: 27–30.
108. **Chu FS, Li GY.** Simultaneous occurrence of fumonisin B1 and other mycotoxins in moldy corn collected from People's Republic of China in regions with high incidence of oesophageal cancer. *Applied and environmental microbiology,* 1994, **60**: 847–852.
109. **Pascale M, Doko MB, Visconti A.** Determination of fumonisins in polenta by high performance liquid chromatography. In: Dugo G et al., eds. *Atti Secondo Congresso Nazionale di Chimica degli Alimenti. Messina, La Grafica Editoriale,* 1995: 1067–1071.
110. **Ueno Y et al.** Fumonisins as a possible contributory risk factor for primary liver cancer: a 3-year study of corn harvested in Haimen, China, by HPLC and ELISA. *Food and chemical toxicology,* 1997, **35**: 1143–1150.
111. **Gelderblom WCA et al.** Fumonisins: isolation, chemical characterisation and biological effects. *Mycopathologia*, 1992, **117**: 11–16.
112. **Kristensen P et al.** Gestational age, birth weight, and perinatal death among births to Norwegian farmers, 1967-1991. *American journal of epidemiology,* 1997, **146**: 329–338.
113. **Emanuel DA, Wenzel FJ, Lawton BR.** Pulmonary mycotoxicosis. *Chest,* 1975, **67**: 293–297.