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International Training Course

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

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**MYCOTOXINS
AS NATURAL CONTAMINANTS
OF FOOD PRODUCTS
AND FEEDS**



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MYCOTOXINS AS NATURAL CONTAMINANTS OF FOOD
PRODUCTS AND FEEDS

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Introduction

Mycotoxins are natural contaminants of food products and feeds. It has become absolutely clear, since the discovery of aflatoxins, that under natural conditions it is not only peanuts but many other vegetable substrates are a favourable medium for the growth of microscopic fungi and the development of aflatoxins. There is convincing proof about the possibility of transmitting aflatoxins to farm animals tissues provided there is a high aflatoxin content in feeds.

The proof of the real hazard of aflatoxins for man's health and of their broad, almost ubiquitous occurrence, has greatly facilitated the current progress in mycotoxicology. This has led to the discovery of a large number of mycotoxins, some of which, like aflatoxins, are natural contaminants of food products and feeds. These include ochratoxins, patulin, zearalenone, trichothecenes and some other mycotoxins.

Contamination of food products and feeds presents not only direct hazard for man's health but it is the cause of a considerable economic damage which is determined, primarily by direct loss of food products and feeds; secondly, by the death, a drop in weight gain and in the reproduction capacity of farm animals, an increase in their susceptibility to infection diseases; thirdly, the expenses involved in the development of a system of surveillance of the contamination of food products

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and feeds with mycotoxins; fourthly, the expenses involved in undertaking decontamination of affected raw material for safe production of food and feeds.

This lecture summarizes the information about the frequency of occurrence and the levels of contamination of food products and feeds by mycotoxins under natural conditions.

Aflatoxins are produced by various strains of only two species of Aspergilli, Aspergillus flavus and A. parasiticus, which are wide spread in the world. According to various information from 20 to 98% of A. flavus strains isolated from different sources are capable of producing aflatoxins. It is likewise noteworthy that toxigenous fungi can affect vegetable substrates not only during their storage, growth, harvesting, transportation and processing.

Under natural conditions aflatoxins occur most frequently and abundantly in peanuts, maize and in cotton seed.

P e a n u t s. Approximately 70% of the world output of peanuts is produced in African and Asian countries whose population regard peanuts as the main source of food protein and oil.

India accounts for one third of the world output of peanuts and from 10 to 40% of peanuts samples and 82% of peanut cake analyzed in 1965-1967 contained aflatoxins in considerable quantities (in some peanut cake samples it was as high as 25,000 µg/kg).

Reports are available about the high frequency and high level of contamination of peanuts with aflatoxins in Thailand (49% of samples, average level of aflatoxins exceeded 1000 µg/kg, the highest was 12,300 µg/kg), Taiwan and Indonesia.

All samples of peanut butter in the Philippines which were studied from 1967 to 1969 contained aflatoxins at a concentration of 155 µg/kg.

Aflatoxin has been found in considerable amounts in peanuts African countries -- in Nigeria (up to 1,700 µg/kg) Sudan, Senegal, Mozambique and Swaziland. Thus, in Sudan (where peanut production yields 20% of the national income) aflatoxins have been found in 41% of peanut samples, and in 8% of the samples at an amount exceeding 10,000 µg/kg.

In the United States where close to 2 million tons of peanuts is produced, primarily for food, the frequency of finding aflatoxins in peanuts in different zones ranges from 0.9 to 6.2% (concentration is higher than 25 µg/kg).

In Brazil, in some years aflatoxin was found in peanuts at a level of 10-15 thousand µg/kg.

In Great Britain in 1968-1974 aflatoxins were found at an amount of more than 5 µg/kg in 22% of peanut samples imported from Indonesia, in 45% of samples imported from India and in 100% of samples imported from Nigeria. In 1966-78 an analysis of 740 samples of feeds showed the presence of aflatoxins in 13.6% of samples. In this, 96.7% of samples of peanut meal contained aflatoxin B₁, out of these one half -- at an amount ranging from 100 to 500 µg/kg, and 5% -- contained more than 1000 µg/kg.

86.5% of peanut products imported in Denmark (1979) were contaminated with aflatoxins. In Norway, all samples of imported peanut studied in 1968-73 also contained aflatoxins.

In Yugoslavia (1982) aflatoxins were found in 20% of peanut samples, and in the GDR (1981) in 39 out of 40 samples (the maximum level being 1600 µg/kg) of peanut and in 13 out of 20 samples

of products of peanut processing.

In Australia (1982 data) aflatoxins were found in peanuts at concentrations of up to 3,300 $\mu\text{g}/\text{kg}$. In the state of Queensland (principal peanut producer) in 1976 15% of peanut harvest was contaminated with aflatoxins and in 1977 -- 50% of the harvest.

The high level of peanut contamination with aflatoxins is one of the main causes of reducing its export by peanut producing countries (India, Gambia, Nigeria, Senegal, Sudan). A major part of peanut imports (70%) falls to the share of European countries. In recent years, however, several countries, including the Netherlands, Denmark, France the FRG and Italy have considerably reduced their import of peanut cake. Beginning with 1970 Denmark completely discontinued this import.

M a i z e. On the world level maize is one of the main cereal crops. By volume of maize production the USA ranks first among maize producers -- more than 40%. Studies in the USA beginning with 1964, found aflatoxins in maize at different levels, usually in samples from southern parts of the country. An analysis of more than 1,500 samples of the 1964-1967 harvests showed the presence of aflatoxins in 2.0-3.0% of the samples at levels of 3-37 $\mu\text{g}/\text{kg}$. In South East states up to 30% of maize samples from 1967 to 1970 contained aflatoxins at an average concentration of 66.0 $\mu\text{g}/\text{kg}$. The stronger drought in that region in 1977 was the cause of a high level of aflatoxin contamination in maize. Thus, according to findings published in 1981-1982, 62.5% of maize samples of the 1977 harvest in Alabama contained aflatoxins at a level exceeding 20 $\mu\text{g}/\text{kg}$. The economic losses associated with maize contamination with aflatoxins in the 1977 harvest ran into 2 million

dollars. In North Carolina 78% of maize samples of the same harvest contained aflatoxins (maximum concentration was 3,600 $\mu\text{g}/\text{kg}$). The resulting economic losses were 31.8 million US dollars, including 16 million as a result of the impossibility of using a part of the maize harvest with an extremely high level of aflatoxin contamination. The high contamination of maize with aflatoxins in 1977 has been noted in a number of other states: in Virginia 43% of the samples; in Georgia - approximately 90%. On the whole, in 7 south-east states of the USA (Alabama, Florida, Georgia, Mississippi, North and South Carolina and Virginia) in 1977 56% of the maize harvest (111.4 million bushels) contained aflatoxins at a concentration exceeding the maximum allowable concentrations established in the USA (20 $\mu\text{g}/\text{kg}$). Aflatoxin concentration in 26% of maize samples exceeded 100 $\mu\text{g}/\text{kg}$. It is not without interest to compare the data on maize contamination in the same period in different regions of the country. Unlike South-East states, the analysis of 30-90% of the maize harvest in 1977 in 8 states of the Mid-West in the USA revealed the presence of aflatoxins but in 6 samples.

It should be emphasized that in several cases high frequency of maize contamination in the South-East states has been established directly in the field. Maize contamination with aflatoxins in the field is mainly attributed to drought and early pest damage to grain.

Aflatoxins are found in considerable concentrations in maize in South-East Asian countries. In India, where contaminated maize was the cause of acute hepatitis with a high rate of mortality the concentration of aflatoxins in some of the samples was 12,500 $\mu\text{g}/\text{kg}$. In Thailand (1967-1969) 35% of

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maize samples contained aflatoxin B₁ 400 µg/kg on average.

In the Philippines, where the average daily maize consumption is 65.9 g per capita, according to the Institute of Food and Nutrition up to 94% of maize samples contained aflatoxins (average level being 77 µg/kg, maximum -- 1,330 µg/kg).

Maize holds an important share in the nutrition of the people in several African countries -- the daily per capita rate ranges from 40 to 300 g. Aflatoxins have been found in maize in Kenya, Mozambique, Uganda, Swaziland, Ghana and Nigeria.

There are sporadic reports about identification of aflatoxins in maize in European countries -- France (maximum level being 187 µg/kg), Yugoslavia (in 1976, in 10% of the examined samples). In the USSR, in maize samples of 1980-1981 harvests, in 3.2% of cases aflatoxin B₁ was found at a concentration exceeding the MAC (5.0 µg/kg).

Aflatoxins have been detected in Australia in maize, and they were the cause of toxicoses in farm animals, at concentrations reaching 340 µg/kg.

O t h e r c e r e a l g r a i n s: aflatoxins are found rather rarely and in comparatively low concentrations. In wheat, aflatoxins have been detected in the USA, at a concentration up to 9 µg/kg), in some Central American countries (up to 10 µg/kg) in Pakistan (from 5.0 to 240 µg/kg), in some European countries (from 5.0 to 48.0 µg/kg), and also in Australia (up to 700 µg/kg). The analysis of wheat and some other cereal grains in the USSR, revealed in one sample out of 169 samples of the 1972 harvest aflatoxin B₁ at a concentration of 100 µg/kg and in 24 out of 138 samples of the 1973

harvest -- 20-444 $\mu\text{g}/\text{kg}$. In the southern areas of the Soviet Union (Kazakhstan) aflatoxin B₁ was found in 5 out of 100 samples of wheat in a concentration ranging from 5.0 to 10 $\mu\text{g}/\text{kg}$.

Rice, is a very valuable food product and in some Asian countries -- the main source of food protein and its per capita daily consumption ranges from 170 to 440 g. It should be likewise noted that 90% of the world rice production comes from countries situated in the monsoon zone of Asia. In natural conditions, however, rice is relatively rarely contaminated with aflatoxins. Thus, less than 2% of rice samples out of 400 selected from the trading network in African countries, the Philippines and Thailand contained aflatoxins. There is information about the detection of aflatoxins in rice in India, Pakistan and Indonesia, Vietnam and also in Mozambique, Uganda and Nigeria, the highest established level of rice contamination with aflatoxins under natural conditions is 600 $\mu\text{g}/\text{kg}$.

Sorghum is widely used as a food product in India and in some Central American and African countries. Aflatoxins have been found in sorghum in India, in the USA (up to 50 $\mu\text{g}/\text{kg}$), Guatemala, Uganda and Nigeria (100% of the examined samples contained from 30 to 211 $\mu\text{g}/\text{kg}$).

Information on aflatoxin contamination of other cereal grains is scanty. Aflatoxins have been found in barley used for fodder, in millet and oats.

A favourable substrate for aflatoxin formation are several species of oil crops -- cottonseed, sun-

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flower seed and soybeans. The high level of contamination of cottonseed contamination should be especially highlighted. This is where aflatoxins have been detected repeatedly in the USA, in Central American countries, India, Iran and such European countries as Greece, the FRG, Denmark, and Sweden. Thus, the examination of the 1964-1967 harvests in the USA revealed aflatoxin B₁ in 6.5-8.8% out of 3,000 cottonseed samples and in 12.7-21.5% of samples out of 3,000 samples of meal prepared of these seed. Examination of samples from different areas of the cotton zone in the USA in 1969 and 1970 revealed an extremely high level of contamination of seed from some of the lots — up to 200-300, µg/kg. High occurrence of aflatoxin contamination of cottonseed has been also observed in 1977 in Arizona and California — aflatoxins (B₁ and B₂) were found in all samples selected in the field and the average level was 387 µg/kg.

Aflatoxins were found in 88% of samples of copra and copra meal imported from the USA, at a level of up to 30 µg/kg, and also in 63% samples of copra imported to Finland.

It is noteworthy that the Philippines produce more than 50% of world copra and 53% of the world export of copra and 23% of the export of coconut oil. According to the Institute of Food and Nutrition, in some studies up to 71% of the samples of copra contain aflatoxins (maximum concentration was 513 µg/kg).

T r e e n u t s are also comparatively often subject to aflatoxin contamination. Toxins have been found in Brazilian nuts, almonds, walnuts, pistachio, filberts, cashew and in peean nuts. For instance, in 1972 more than 80% of pistachio

imported in the USA from Iran and Turkey contained aflatoxins in volumes exceeding 20 µg/kg. Aflatoxins were found in 8 samples of filberts imported from Turkey at a concentration up to 100 µg/kg. 14% of the samples of Californian almonds contained aflatoxin B₁ in low concentrations.

In Yugoslavia, aflatoxins have been found in 33% of the samples of walnuts, and in Tunisia more than 50% of Aleppo pine nuts contained aflatoxins at a level ranging from 100 to 2,000 µg/kg.

Aflatoxins occur but rarely under natural conditions in fresh vegetables and fruit, though it has been shown that a large part of A. flavus strains isolated from them are toxicogenic. Detection of aflatoxins was reported for molded oranges and 29 out of 91 samples of fruit processing products (juices, jam, marmelade) at a level of 30 to 50 µg/kg.

There were sporadic cases of aflatoxins presence in coffee and cocobean and also in some spices.

Aflatoxins are found frequently and in considerable amounts in feeds and their ingredients in many countries. Thus, in Great Britain from 1966 to 1978 aflatoxin B₁ was found in 13.6% of the examined samples of feeds. In Poland, 12.7% of the samples of combined feeds contained aflatoxins (4.2% in a concentration exceeding 100 µg/kg); in the FRG aflatoxins have been found in 46 out of 165 samples of combined feeds at concentrations ranging from 7 to 300 µg/kg. In Brazil, 25% of samples examined over the period from 1971 to 1979 contained aflatoxins in quantities exceeding 30 µg/kg (maximum being 7,870 µg/kg).

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In Pakistan, 18.5% of the examined samples of feeds were contaminated with aflatoxins. In Australia (Queensland) from 1971 to 1980 355 samples of feeds were examined and aflatoxins were found in 68 at a concentration in excess of 50 µg/kg.

In the USA (Florida) in 1977 in connection with toxicoses of pigs 2,800 samples of feeds and fodder maize were examined. More than 50% of them contained aflatoxins in the amount of 400 µg/kg. In 1978 11% of 1,131 samples contained aflatoxins at a concentration more than 400 µg/kg, while 2.3% -- higher than 2,000 µg/kg.

Special mention should be made of the possibility of aflatoxins occurrence in products of animal origin -- in milk, in tissues and organs of animals feeding on fodder contaminated with aflatoxins at high concentrations.

In cows, sheep and goats which were given contaminated feeds, aflatoxin M₁ -- a highly toxic metabolite of aflatoxin B₁ was found in their milk. According to published findings, cows feeding on fodder contaminated with aflatoxin B₁, excrete it with milk at a rate of 0.35 to 2-3%. Aflatoxin M₁ has been found both in liquid and powdered milk and in dairy products. Results of some studies have been summed up in Table 1.

An extremely high level of aflatoxin (up to 250 µg/l) has been found in 50% of samples of cow milk taken in Iranian villages in 1973 and 1974. The same study found aflatoxin M₁ in milk samples taken from large farms but in 10 samples at an amount of 8-10 µg/l.

Aflatoxins have been found in cheeses from the FRG, France and Switzerland (0.1-0.6 µg/kg) and Greece (up to

30 µg/kg).

It is important to stress that pasteurization and dehydration of milk do not materially affect the content of aflatoxin M₁. During the production of cheese from contaminated milk 50% of aflatoxin M₁ is being found in the curds. When butter is made, 10% of aflatoxin M₁ passes to sweet cream, 75% remains in skimmed milk.

It has been experimentally shown that aflatoxins may develop in different organs and tissues of pigs and cattle, in poultry meat and eggs, in fish given a sufficiently high content of aflatoxins in the feeds. A case has been described when aflatoxin B₁ was found in muscle tissue, kidneys and liver of raindeer (at a concentration from 0.1 to 0.4 µg/kg) and in the liver and the muscle tissue of pigeons (64.2 and 0.03 µg/kg), which found their feed in the fields where contaminated maize was found.

It is not without interest that under laboratory conditions the development of aflatoxins at a high concentration is possible in many products infected with toxigenic strains -- in grapes, peaches, apples, tomato and pineapple juices, in butter, lard and meat.

Aflatoxin-associated occupational health hazard

Considerable attention has been given in recent years to effect of aflatoxin on man at work. This concerns the personnel engaged in the handling of grain, feeds, peanuts, peanut meal contaminated with aflatoxins, i.e. in those types of work where there is the possibility of inhaling or ingesting dust contaminated with aflatoxins.

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Table 1
Incidence and level of contamination of liquid and dry milk with aflatoxin M₁ in some countries (different literature sources)

Milk samples	Countries	Number of examined samples	Number of samples with aflatoxin M ₁	Contamination level (µg/l or µg/kg)
Liquid milk	Belgium	68	42	0.02 - 0.2
	The GDR	36	4	1.7 - 6.5
	The FRG	61	28	0.01 - 0.25
		419	79	Traces - 0.54
		260	118	0.05 - 0.33
	India	21	3	Up to 13.3
	The Netherlands	95	74	0.09 - 0.5
	Great Britain	278	85	0.03 - 0.52
	Yugoslavia	105	5	-
	Powder milk	The GDR	18	0
The FRG		166	8	0.67 - 2.0
		52	35	Traces - 4.0
		120	7	0.05 - 0.13
Austria		1047	594	Up to 0.2
South Africa		56	0	-
USA		320	24	0.01 - 0.4
		302	192	Traces - 3.9

Studies in the USA have shown that aflatoxin B₁ concentration in air dust may achieve 130 µg/kg at places of storing and processing such grain products as oats, wheat, barley and maize. It has been also shown that the toxin concentration is higher in particles with a smaller diameter. Thus, in particles of less than 7 µm size aflatoxin concentration exceeded 1,000 µg/kg. It was shown that under laboratory conditions when contaminated maize was grounded (the aflatoxin content being 2.25 µg/kg), aflatoxin concentration in the air dust in the premises exceeded its content in grain and reached 2.56-4.56 µg/kg.

The average content of aflatoxin in a samples of dust formed during the conveying of grain from hoppers to a carriage or vice versa was 138 µg/kg.

A report from Czechoslovakia stated that 65% of toxigenous strains of Aspergilli was isolated from samples of air and settled dust in premises where imported products -- peanuts, coconuts filberts, etc. were packed.

Ochratoxins; sources and spread

Ochratoxins, primarily ochratoxin A, are mycotoxins with nephrotoxin action similar to aflatoxins by their toxicity. Ochratoxin A are produced by many species of Aspergilli, primarily A. ochraceus, as well as some penicilli, more often -- P. viridicatum. The best toxin formation temperature for aspergilli is within 20-30°C range, whereas penicilli are capable to produce ochratoxin at 7.5-10°C.

Detection of ochratoxin A in food and feeds was also reported from a number of European countries - Great Britain,

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Denmark, Sweden, France, Hungary, Poland, the GDR, Czechoslovakia, Yugoslavia and also in Canada and the USA.

Occurrence of ochratoxin A in food products varies from 1 to 18.8% whereas the average contamination level - from 5 to 360 $\mu\text{g}/\text{kg}$. Ochratoxin A concentration in feeds reached 27,500 $\mu\text{g}/\text{kg}$. In Denmark, in areas with a high incidence of nephropathy in pigs up to 58% of barley contained ochratoxin, 9% at an amount reaching 2,000 $\mu\text{g}/\text{kg}$ (Table 2).

In addition to the data in Table 2 one should note that in the GDR the analysis of 99 samples of food products with obvious signs of spoil, ochratoxin A was found in 2 samples of maize and in 2 samples of coffee (up to 90 $\mu\text{g}/\text{kg}$). In Great Britain, the flour going into retail sale was proved to contain ochratoxin A at a concentration ranging from 6 to 250 $\mu\text{g}/\text{kg}$ in samples with altered organoleptic properties and at a level of 10 $\mu\text{g}/\text{kg}$ - in unaltered flour.

Ochratoxins may accumulate in animal tissue - in kidneys, liver and in muscle tissue of pigs and poultry and it may also be excreted with milk. In Denmark, the study of kidneys in nephropathic pigs selected at slaughter houses has shown that 35% of them contained ochratoxin A at a level of 2 to 68 $\mu\text{g}/\text{kg}$. In Sweden 25% of nephropathic pigs have shown the presence of ochratoxin in their kidneys at a concentration from 2 to 104 $\mu\text{g}/\text{kg}$.

Patulin: sources and spread

Patulin is produced by different species of Penicillium, Aspergillus and also Byssochlamys nivea. By the nature of toxic action it belongs to neurotropic toxins and possesses carcino-

Table 2
Incidence and level of contamination of vegetable food and feeds with ochratoxin A
in some countries

Type of food or feed	Country	Number of studied samples	Number of samples with ochratoxin A (%)	Contamination level ($\mu\text{g}/\text{kg}$)
<u>Food products</u>				
Maize	USA	293	1.0	83 - 166
	France	463 461	2.6 1.3	15 - 200 20 - 200
Winter wheat	USA	291	1.0	5 - 115
	USA	286	2.8	5 - 115
Spring wheat	Denmark	50	6.0	9 - 189
	USA	127	14.2	10 - 40
Coffee beans	USA	267	7.1	20 - 360
	Yugoslavia	542	8.3	6 - 140
Maize	USA	130	8.5	14 - 135
	Barley	64	12.5	14 - 27
Wheat	Poland	203	4.9	10 - 50
	Poland	150	5.3	50 - 200
Barley	Canada	32	56.3	30 - 27000
	Denmark	33	57.6	28 - 27500
Mixed feeds	Sweden	84	8.3	16 - 409
	Canada	95	7.4	30 - 6000
Wheat, barley, maize, rye, oats	Yugoslavia	191	25.7	45 - 5125
	Barley, oats			
Feeds	Wheat, hay			
	Maize			

genic and teratogenic properties.

Patulin producents affect primarily fruit and some vegetables. Patulin has been found in apples, pears, apricots, peaches, mazzard-cherry, grapes, bananas, raspberry, tomatoes and some fruit juices. In a report by Reiss, patulin was found in some bakery products at a concentration amounting to 200 $\mu\text{g}/\text{kg}$.

Apples are contaminated more frequently than other fruits. According to different sources, patulin content in apples varies from 20 to 17,700 $\mu\text{g}/\text{kg}$. In the FRG and Canada patulin has been found in 50% of the studied apples, in some cases its concentration reached 18,000 $\mu\text{g}/\text{kg}$.

Patulin was found in apple juice in the USA, Canada, the FRG, the GDR, Finland, France (Table 3). The content of patulin in other types of juice - pear, quince and grapes in different studies was found to vary from 30 to 4,500 $\mu\text{g}/\text{kg}$.

It was shown that citrus fruits and some vegetables possess natural resistance to infestation with patulin's producents. These, specifically, include potato, onions, radishes, eggplant, colliflower, pumpkin and hoarse radish.

Different representatives of the wide spread genus Fusarium are producents of mycotoxins and they include trichothecenemycotoxins and zearalenone.

Trichothecenemycotoxins comprise a group of compounds incorporating more than 40 metabolites of different species of Fusarium, Cephalosporium, and Myrothecium. This group contains as natural contaminants of food products and feeds the following main mycotoxins: T-2 toxin, deoxinivalenol (vomitoxin), nivalenol, and diacetoxiskirpenol.

Table 3
 Frequency and level of patulin contamination of apple juice
 in some countries (different literature sources)

Country	Mode of manufacturing of apple juice	Number of examined samples	Number of samples with patulin, %	Maximum contamination level (ug/l)
USA	Commercial	136	37	440
		95	4	25000
		13	61	300
		40	57	more than 300
Canada	Commercial	12	8	1000
The FRG	Commercial	72	29	150
The GDR	Commercial	274	84	more than 400
		106	40	200
		36	20	300
France	Commercial	7	70	300
	Household	8	50	300
Finland	Commercial	24	0	-
	Household	20	40	1000
Sweden	Commercial	66	37	54
Norway	Commercial	140	25	220

The main information on the spread of trichothecene mycotoxins under natural conditions is summed up in Table 4 which shows that trichothecene mycotoxins are found particularly often in maize, barley and wheat.

Besides isolation of fungi producers from peanuts, fodder grain and different formular feeds was reported for trichothecene mycotoxins in Japan, Korea, India, the USA, the USSR, Sweden, and Hungary.

Zearalenone: sources and spread

Zearalenone is a metabolite of F. graminearum, F. tricinctum, F. moniliforme and F. oxysporum. The best temperature for its formation is 12-14°C. Under laboratory conditions the toxic strains of Fusarium produce a range of derivatives of zearalenone but only zearalenone occurs as a natural contaminant of food products and feeds.

Zearalenone possesses a strong estrogenic and teratogenic action and is a serious problem for livestock husbandry in many countries while its ability to accumulate in tissues of farm animals renders it a mycotoxin which is potentially dangerous for man's health.

Under natural conditions zearalenone occurs primarily as a contaminant of grain products. It has been found in considerable concentrations in maize, barley, wheat and also in oats, sorghum, different feeds, in walnuts in the USA, Canada, Australia, Japan, South Africa and in many European countries.

It should be stressed that fungi of the F. graminearum genus frequently affect maize in the field and are the cause of the rot of cobs and stock. Thus, maize contamination of

Table 4

Level of contamination with trichothecene mycotoxins of feeds and food of vegetative origin in some countries (different literature sources)

Mycotoxin	Country	Product	Contamination level ($\mu\text{g}/\text{kg}$)
T-2 toxin	USA	Maize	2.0
	Yugoslavia	Maize	up to 5.0
	India	Maize	4.0
	France	Maize	0.02
	Canada	Barley	25.0
	India	Saffor Sorghum	0.5 -
	USA	Combined feed	0.076
Desoxinivalenol (vomitorin)	USA	Maize	0.5-10.7; 1.8
	Canada	Maize	0.47-0.89; 7.9
		Wheat	0.1-0.6
		Barley	0.24-0.43
		Oats	0.08-0.11
	South Africa	Maize	0.25-4.0; 2.5-7.4
	Austria	Maize	1.3; 7.9
	France	Maize	0.6
	Japan	Barley	up to 40.4; 7.3
		Wheat	up to 4.7
Diacetoxiskir- penol	FRG	Oats	3.0; 15.0
	FRG	Maize	31.5
	India	Maize	up to 14.0
Saffor		1.0	
Nivalenol	USA	Combined feed	0.5
	France	Maize	4.28
	Japan	Barley	up to 36.9
		Wheat	up to 7.8

zearalenone may occur both in the field and in storage.

In the USA, for instance, examination of maize contaminated with zearalenone showed that in 1967-1969 from 1 to 2% of samples of the maize studied contained zearalenone. In 1972, 17% of the samples proved to be contaminated and in 1973 up to 10% of maize contained zearalenone at a concentration ranging from 0.4 to 5.0 $\mu\text{g}/\text{kg}$.

There are reports about detecting in maize and oats, alongside with zearalenone (at a concentration of 25 and 135 $\mu\text{g}/\text{kg}$), zearalenol in amounts reaching 4.0 $\mu\text{g}/\text{kg}$. It should be stressed that the estrogenic activity of zearalenol is from 3 to 4 times higher than the activity of zearalenone.

Table 5 shows levels of feed contamination with zearalenone which was the cause of a hyperestrogenic syndrome in pigs in the USA.

Cases of hyperestrogenism in pigs caused by maize contaminated with zearalenone has been registered in 1963, 1968, 1969, 1972 and 1974. Thus, in 1972 more than 50% of maize samples proved to be infected with a zearalenone producer -- Fusarium graminearum, 42% of samples contained zearalenone in amounts ranging from 0.7 to 37.5 $\mu\text{g}/\text{kg}$. Approximately 3% of the samples of the 1976 harvest contained zearalenone in concentrations ranging from 0.043 to 10.0 $\mu\text{g}/\text{kg}$. According to information for 1982 70.7% of samples of feeds in Yugoslavia contained zearalenone in amounts from 0.2 to 20.0 $\mu\text{g}/\text{kg}$. This includes 83.3% of the studied samples of maize (88.3% of samples of combined feed for pigs and 100% of the examined samples of balanced fodder for cattle.

Table 5

Zearalenone contamination level in feeds which caused hyperestrogens effect in pigs in some states of the USA (5)

Feed	State	Contamination level ($\mu\text{g}/\text{kg}$)
Maize grain	Minnesota	0.1-0.15
Dry rations of sows	Vancouver	0.15
Rations for piglings	Vancouver	0.066
Dry rations of sows	Vancouver	0.15 0.25
Maize grain	Vancouver	0.20
Rations during lactation	Vancouver	1.00
Rations during pregnancy	Vancouver	0.5
	Minnesota	2.5-5.5
Sesame oil	Minnesota	1.5
Maize grain	Ohio	0.12
Mixed fodder maize	Ohio	0.12
Maize grain	Minnesota	6.4
Mixed feed in brickets	Minnesota	6.8

Level of contamination of feeds with mycotoxins produced by microscopic fungi of the Fusarium genus and the symptoms of the toxicosis caused by them in animals (15)

Detected mycotoxin	Contamination level (µg/kg)	Type of feed	Symptoms of intoxication
Diacetoxyscirpenol	500 380	Balanced fodder "-"	Intestinal haemorrhages in pigs
Desoxynivalenol, zearalenone	1800 250	Maize grain	Feed refusal in pigs
Desoxynivalenol, zearalenone	1000 175	Maize grain	Feed refusal in pigs
Desoxynivalenol, zearalenone	100 1750	Maize grain	Feed refusal in pigs
Desoxynivalenol, zearalenone	40-60 3600	Balanced fodder in brickets	Feed refusal and stool with blood in pigs
T-2 toxin, zearalenone	76 700	Balanced fodder	Stool with blood in neat cattle
Desoxynivalenol, zearalenone	1000 500	Mixed feed	Vomiting in dogs
Desoxynivalenol, zearalenone	1000 traces	Balanced fodder	Feed refusal in pigs
Zearalenone	6400	Maize grain	Feed refusal in pigs
Zearalenone	1500	Sesame meal	Diarrhea in turkeys
T-2 toxin	300	Balanced fodder	Vomiting and diarrhea in pigs
Zearalenone, desoxynivalenol	210 70	Bricketed food	Death of dogs
Zearalenone	250	Bricketed food	Feed refusal, decrease in lactation of cows
Zearalenone	500	Bricketed food	Gasterenteritis and haemorrhages in rabbits
Zearalenone, desoxynivalenol	10 1000	Maize grain	Slow growth in cattle

The study of cases of fusariotoxicosis in pigs and milch-
ing cows in Hungary, zearalenone was found in feeds and maize
at concentrations reaching 80.0 $\mu\text{g}/\text{kg}$. In France, in one of
the studies conducted in 1974, the content of zearalenone
reached 170 $\mu\text{g}/\text{kg}$ in 85% of samples of maize.

Zearalenone has been also found in maize in India (up to
16.0 $\mu\text{g}/\text{kg}$), and also in South Africa at concentrations reach-
ing 12.8 $\mu\text{g}/\text{kg}$.

There are reports about detection of zearalenone in 11%
of samples of beverages, porrage and beer of local production
in Swaziland at concentrations from 8.0 to 53.0 $\mu\text{g}/\text{kg}$ and
also in 12% of samples out of 140 samples of beer in Lesoto,
at concentrations from 0.3 to 2.0 $\mu\text{g}/\text{l}$.

It should be borne in mind that one and the same food
products and feeds may contain several mycotoxins produced
by microscopic fungi of the Fusarium genus. Mirocha et al.
(1980) showed that the feeds which caused alimentary toxicoses
in animals often contained both trichothecene mycotoxins and
zearalenone (Table 6).

In conclusion of this part we should note that some
zearalenone derivatives are now used as growth stimulants for
farm animals.

Conclusion

This lecture has taken up very vast material on mycoto-
xins as natural contaminants of food products and feeds. The
information given here shows convincingly that mycotoxins
are ubiquitous. Indeed, mycotoxins are found in most coun-
tries in all continents (Table 7). It should be also borne in

mind that mycotoxin may contaminate all main foodstuffs (Table 8). It is also important that intensive trade among countries and continents facilitates greatly the spread of mycotoxins and mycotoxicoses. There are all grounds to believe that the problem of mycotoxins is a global one and involves interests of all countries.

Table 7

Geography of mycotoxin occurrence in the world

Continent	Countries where mycotoxins have been found as natural contaminants of food and feeds
Europe	Austria, Bulgaria, Great Britain, Hungary, GDR, Greece, Denmark, Ireland, Spain, Italy, Norway, Poland, Portugal, USSR, Finland, France, FRG, Czechoslovakia, Switzerland, Sweden, Yugoslavia.
Asia	Bangladesh, Burma, Vietnam, India, Indonesia, Iraq, Iran, Pakistan, Thailand, Turkey, the Philippines, Sri Lanka, Japan.
Africa	Angola, Upper Volta, Gabon, Gambia, Ghana, Benin, Egypt, Zambia, Lesoto, Mali, Malawi, St. Mauritius, Madagascar, Mozambique, Niger, Nigeria, Kenya, Swaziland, Senegal, Sudan, Togo, Tunisia, Uganda, SAR.
America	Argentina, Brazil, Guatemala, Canada, Columbia, Mexico, Nicaragua, Salvador, USA.
Australia	

Table 8
Food and feed contaminated with mycotoxins under natural conditions

Group of products	Type of products
Cereal grains	Maize, wheat, barley, oats, rye, rice, sorghum, millet, bread and bakery products.
Oil-bearing crops	Peanuts and products of processing (meal, oil, cake), cotton seed and products of processing (oil, meal, cake), sunflower, soya, copra, sesame.
Nuts	Pistachio, brazil nuts, filberts, almonds, cashew, pecan, walnuts, Aleppo pine nuts, chestnuts.
Legumes	Beans, peas, etc., coffee and cocoa beans.
Fruits, berries	Apples, pears, apricots, peaches, bananas, mazzarad cherry, raspberry, oranges, figs, etc., dry fruit, sea buckthorn.
Vegetables	Tomatoes, red pepper.
Livestock products	Meat, internal organs of farm animals, milk and dairy products, meat and eggs of poultry, fish and other marine products.
Beverages	Beer, wine, fruit and vegetable juices, paste, jam, marmelade, etc.
Feeds of farm animals and poultry	Combined feeds, hay, straw, etc.

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