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Overview of Mycotoxins in Food and Animal Feed: Recent Advances

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Abstract

Mycotoxins, toxic metabolites produced by certain fungi, pose significant and multifaceted threats to human and animal health, agriculture, and international trade. This review aims to provide a comprehensive overview of mycotoxins, focusing on their classification, occurrence, and impact on human and animal health. The review encompasses an analysis of existing literature on mycotoxin contamination using relevant information gathered from databases such as PubMed, Scopus, and Web of Science, with keywords including mycotoxin contamination, aflatoxins, ochratoxins, fumonisins, zearalenone, trichothecenes, deoxynivalenol, crops, food, and feed. The search aimed to capture existing literature on key mycotoxins of global concern and factors influencing their presence in crops, food, and feed. These contaminants can cause mycotoxicosis through chronic or acute exposure. Biotic and abiotic factors influence mycotoxin contamination, and establishing global regulatory limits is crucial for mitigation. A holistic management approach, involving all relevant stakeholders and encompassing pre-harvest, harvest, and post-harvest strategies, is necessary for controlling mycotoxin contamination and enhancing public health and international trade.

Keywords

Mycotoxins, Health Hazard, Food & Feed, Future Trends, Emerging Mycotoxins

1. Introduction

General Overview of Mycotoxins

The occurrence of mycotoxin-producing fungi in foods and feeds has become a significant global concern [1]. Mycotoxins are toxic metabolites produced by fungi such as Aspergillus, Fusarium, and Penicillium, which can lead to life-threatening health conditions, collectively known as mycotoxicosis [2]. The health implications of mycotoxins are profound, as they are carcinogenic, teratogenic, nephrotoxic, mutagenic, immunotoxic, and allergenic [3]-[6]. These toxic compounds can cause acute and chronic toxic effects on the cardiovascular system, liver, and central nervous system of humans and animals [7] [8].

The key mycotoxins of major concern include aflatoxins, fumonisins, ochratoxin A, zearalenone, and trichothecenes. These mycotoxins are known to contaminate a wide range of agricultural products, including cereals, nuts, spices, cocoa, and coffee beans [3] [9]. The global impact of mycotoxin contamination is substantial, with approximately 4.5 - 5 billion individuals facing a significant risk of long-term exposure to harmful mycotoxins [10]. This risk is particularly pronounced in underdeveloped countries, where limited resources and infrastructure lead to poor agricultural practices, inadequate storage, and a lack of regulatory enforcement, resulting in higher mycotoxin contamination rates.

The challenges posed by mycotoxin contamination are multifaceted, contributing to food loss, reduced food availability, and heightened risks of hunger and malnutrition. Furthermore, the carry-over of mycotoxins in livestock products, such as meat, can lead to chronic or acute toxicity in humans [6] [11]. Given the severity of these issues, it is crucial to understand the classification and occurrence of mycotoxins, their toxicological effects, methods for their detection, and strategies for prevention. This review, titled "Overview of Mycotoxins in Food and Animal Feed: Recent Advances", aims to provide a comprehensive overview of these aspects, offering insights into the current state of mycotoxin research and future directions for mitigating this global health threat.

2. Classification and Occurrence of Mycotoxins

Mycotoxins, which are secondary metabolites of fungi, can be broadly classified based on their chemical structure and biological activities (**Table 1**). As reported by [12], the occurrence of mycotoxins in Sub-Saharan Africa indicates that aflatoxins (43.75%) are the most prevalent, followed by fumonisin (21.87%), ochratoxin A (12.5%), zearalenone (9.38%), and deoxynivalenol (6.25%).

Globally, mycotoxin surveys revealed high occurrence rates of fumonisin (64%), deoxynivalenol (65%), and zearalenone (48%), underscoring the widespread occurrences of these mycotoxins globally [13]. Mycotoxins have significant impacts on agricultural economies, and human and animal health. Understanding the biological activities of these mycotoxins is crucial for developing effective mitigation strategies and regulatory frameworks suitable for their control.

Table 1. Classification of mycotoxins based on chemical structures and biological activities.

Mycotoxins	Classification based on chemical structure	Classification based on biological activities	References
Aflatoxins (B_1, B_2, G_1, G_2)	Polyketides	Hepatotoxic, carcinogenic, immunotoxic, cytotoxic	[6]
Ochratoxin (A, B, C)	Polyketides	Cytotoxic, immunotoxic, nephrotoxic, carcinogenic	[14]
Fumonisin (B_1, B_2)	Polyketides	Cytotic, neurotoxic, carcinogenic	[14]
Deoxynivalenol	Peptide-derived	Gastrointestinal	[15]
Zearalenone	Terpenoid-derived	Estrogenic, nephropathic, carcinogenic	[16]
Patulin	Phenolic	Carcinogenic, mutagenic, neurotoxic, immunotoxic	[17]
Nivalenol	Peptide-derived	Gastrointestinal	[15]
Enniatins	Cyclic hexadepsipeptides	Cytotoxic, antibacterial, anthelmintic, phytotoxic, insecticidal	[18]
Beauvericin	Cyclic hexadepsipeptides	Antibacterial, antifungal, apoptosis, phytotoxic	[18]

3. Mycotoxins of Concern in Food and Animal Feed

Although a wide range of mycotoxins has been discussed, this study will concentrate on the following key types.

3.1. Aflatoxin

Aflatoxins (Afs), being secondary metabolites of certain strains of fungi such as *Aspergillus flavus* and *A. parasiticus*, and the less common *A. nomius* [19], are very dangerous to health when consumed. The name "aflatoxin" was derived from the name of the genus and species of the microorganism from which it was first identified (*i.e.*, "a" from Aspergillus and "fla" from flavus). The B and G classes indicate the blue and green light fluorescence emitted by these metabolites when viewed under ultraviolet (UV) light, and the sub-types 1 and 2 refer to the major and minor compounds, respectively. *A. flavus* is a producer of the type B aflatoxin, while the other two fungal species produce both types of aflatoxins, B and G [20].

Aflatoxins contaminate a wide variety of important foodstuffs including cereals, and aflatoxins could cause cancer, behave as mutagens, teratogens, and can be immunosuppressive. Aflatoxins can cause both acute toxicity and chronic carcinogenicity in humans and animals [21]. Aflatoxin B1 is classified by the International Agency of Research on Cancer (IARC) as a Group 1 carcinogen, with high risks for hepatocellular carcinoma (HCC) in individuals exposed to aflatoxins, while AFM1 is listed in Group 2B; possibly carcinogenic to humans [22]. Acute toxicosis, though rare in developed countries, is common in some developing countries such as Africa [23]. The toxic effects of AFB1 are principally due to the binding of bioactivated AFB1-8,9-epoxide to cellular macromolecules, particularly mitochondrial and nuclear nucleic acids and nucleoproteins, resulting in general cytotoxic effects [21]. Due to the toxicity of aflatoxins to humans and animals, many countries have established regulations and regulatory limits for their

control in foods meant for human and animal consumption [24].

Recent studies have highlighted the importance of understanding the molecular mechanisms of aflatoxin toxicity, including the role of miRNAs in regulating gene expression [25]. The classification of aflatoxin B1 as a Group 1 carcinogen by [26] underscores the need for stringent regulations and controls to minimize exposure. Novel approaches, such as the use of nanotechnology-based detection methods [24], may enhance our ability to detect and mitigate aflatoxin contamination.

3.2. Ochratoxin

Ochratoxins are products of moulds, most especially *Aspergillus* and *Penicillium.* Among the ochratoxin family A, B, and C, ochratoxin A (OTA) is the most dangerous to health [27]. Cereals such as maize, oats, barley, wheat, groundnuts, dried fruits, and coffee are the main sources of OTA contamination. These were reported to have been infected by *A. ochraceus, A. niger, A. carbonarius, A. flavus, A. auricomus, A. glaucus, A. melleus, A. alliaceus, P. verrucosum, and P. virridicatumn* [21].

The International Agency for Research on Cancer (IARC) has classified OTA as a group 2B carcinogen because of its nephrotoxic, hepatotoxic, immunosuppressive, teratogenic, and carcinogenic effects on animals and humans [26]. Several countries have established regulatory bodies for the detection and control of OTA because of its toxicity and frequent occurrence in feed and foodstuffs [28]. The average range/limit for OTA is between 3 - 50 μg/kg [28]. The European Commission (EC) regulation maximum limits for OTA in unprocessed cereals are 5 μg/kg and 3 μg/kg for unprocessed cereal products as published in Commission Regulation No. 1881/2006 [29].

Ochratoxin A (OTA) can negatively impact the productivity of food-producing animals by decreasing feed efficiency, leading to weight loss, and reducing egg production [30]. Due to its fat-soluble nature, OTA tends to accumulate in animal tissues, particularly in pigs [31]. OTA disrupts the activity of the enzyme phenylalanine hydroxylase in the kidneys and liver, hindering proper protein synthesis at the molecular level. Furthermore, OTA also inhibits the synthesis of RNA and DNA [32], leading to a range of harmful effects on animal health and productivity.

3.3. Fumonisin

Fumonisins are produced by several species of Fusarium, which include *F. verticillioides, F. proliferatum,* and *F. nygamai*. The most commonly produced toxin is Fumonisin B1, which is of *Fusarium verticillioides* origin. The *F. verticillioides* is of great economic importance and can be found in maize. Fumonisins, produced by Fusarium species, interfere with sphingolipid metabolism [33] and are associated with various syndromes in animals, including leukoencephalomalacia in horses and hepatotoxicity in rats [34]; pulmonary oedema and hydrothorax in pigs [35]; carcinogenicity and apoptosis [36] in the liver of rats. This, if consumed in large amounts, may also lead to cancer of the lungs in humans [37]. It can also

lead to serious diseases in rats, horses, and cattle and could cause liver, kidney, and lung damage in rats, and equine leukoencephalomalacia, a neurological disease, in horses [38]. Fumonisins are also linked to pulmonary inflammation in swine and liver cancer in rodents [38].

3.4. Zearalenone

Zearalenone (ZEN), like fumonisin, is also produced by some species of Fusarium, the most common being *F. graminearum*, and others such as *Fusarium crookwellense, Fusarium culmorum*, and *Fusarium equiseti*, may also be a cause. Cereal crops such as maize are the main target [39]. Biologically, they cause reproductive problems in animals and also affect oestrogen, resulting in abortion, infertility, and other reproductive malformations in many animal species such as sheep, cattle, and pigs. It has lesser toxicity compared to fumonisin in swine, cattle, and sheep [40].

Zearalenone is absorbed by the intestinal tract and undergoes metabolism, leading to the production of some metabolites, some of which are even more potent than their parent toxin [41]. ZEN binds to estrogen receptors to cause hyperestrogenic effects, thereby disrupting endocrine systems [42]. Increased levels of ZEN may induce toxicity, which may give rise to oxidative stress and cytotoxicity. Zearalenone affects reproductive systems, based on the research on pigs, leading to hyperplasia of the reproductive tract, infertility, and hormonal dysfunctions. For humans, exposure to ZEN is linked to the development of breast cancer and endometrial hyperplasia. Conclusively, ZEN could affect various organs such as the liver, kidneys, immune cells, and intestines [39].

3.5. Trichothecenes

Trichothecenes are mycotoxins produced by a number of fungal genera, including Fusarium, Myrothecium, Trichothecium, Trichoderma, and Cephalosporium among others [21]. The name "trichothecenes" was given as a result of the first member of the family identified. The almost 180 different structures are grouped into four distinct groups, A to D. Cereals such as maize, oats, barley, etc. are the primary source of trichothecene. The type-A and type-B trichothecenes are less toxic and are commonly circulated by means of the mentioned crops. The type-C and type-D are more toxic and rarely occur in food and feed.

The genus Fusarium comprises numerous field fungi adapted to infect plants, including wheat, corn, barley, oats, and forages. Fusarium affects plants in temperate regions, and this has led to most outbreaks.

Trichothecenes prevent protein build-up and are toxic to molds, bacteria, plants, and animals [43]. Trichothecenes are established food and feed contaminants, and their consumption may lead to irritation of the intestines, which may result in alimentary hemorrhage, vomiting, and diarrhea. Direct contact with trichothecenes can cause dermatitis, while chronic exposure in animals can lead to immune system suppression [38].

3.6. Patulin

Patulin, produced by Aspergillus, Penicillium, and Byssochlamys fungi, is toxic to both plants and animals. It is present in rotten apples and apple products, grains, and other foods. Despite the antibacterial, antiviral, and antiprotozoal properties of patulin, it is reported to be toxic to both plants and animals [44]. The toxicity of patulin has masked its clinical uses as antibiotics [45]. Patulin is associated with the following acute symptoms in animal species: liver, spleen, and kidney damage, and immune system toxicity, while in humans it is responsible for nausea, gastrointestinal disorders, and vomiting.

The US FDA limits patulin to 50 ppb as an action level in food for human consumption. The EU committee has set a maximum level of 50 ppb for fruit juices and concentrated fruit juices, 25 ppb for solid apple products, and 10 ppb for juices and foods consumed by babies [46].

3.7. Deoxynivalenol

Deoxynivalenol (DON) is produced by fungi of the genus Fusarium, and they can be found in several grains and products of animal origin. Ingestion of food contaminated by DON may cause nausea, emesis, diarrhoea, abdominal and head pain, dizziness, fever, and eventually death. The severity of the disease depends on the concentration of the ingested toxin. The chronic effects of DON are mainly felt on the immune, reproductive, and gastrointestinal systems [47] [48]. Ingestion of DON, in addition to its acute effects, can lead to genotoxicity in human lymphocytes, which may later lead to DNA damage by oxidative stress [49]. Deoxynivalenol (DON) exposure is associated with emetic effects in humans, and animal research has demonstrated a range of detrimental effects, including immunosuppression, anorexia, reduced nutritional efficiency, and impaired reproductive capacity [50].

3.8. Enniatin

Enniatins (ENN) are made up of different groups, of which ENB1, despite being less studied, is the most widely relevant and prevalent ENN contaminant of agricultural produce [51]. ENN occurs in cereals such as maize, barley, oats, etc., but to a lesser extent in potatoes, eggs, apples, etc. [52]. In a study conducted on about 4000 food, feed, and unprocessed grain samples in Europe between 2000 and 2013, ENNs were detected in 37%, 68%, and 76% of food, feed, and unprocessed grain samples, respectively [53]. As high as 5720 μg/kg was reported in grains, 980 μg/kg was reported in cereal-based food, and 5441 μg/kg in feed and feed raw materials [54]. [55] reported these contaminants in Nordic countries, which include Norway, Sweden, Iceland, North America (Canada), Asia (Iran, China), as well as Africa (Egypt, Mozambique, Cameroon, etc.). Because of its wide distribution across the globe, it can be deduced that the formation of this toxin by *Fusarium* spp. is not affected by any climatic condition but may be modified, which in turn has an impact on its toxicity [55]. Biological contamination of crops by this fungus re-

sults from the crops' compatibility with the fungus, harvesting, and storage methods and conditions, which make ingestion of the fungus by plants and animals very easy and rampant [51].

Most ENNs exhibit both antifungal and antibacterial characteristics against organisms such as *Candida albicans, Cryptococcus neoformans, Escherichia coli*, *Yersinia enterocolitica*, etc. [18]. Recently, De Felice *et al.*, 2023, reported ENN B_1 to have the ability to permeabilize the lysosomal membrane by destabilizing the LAMP-2 complex at a concentration close to the EC50 (1.5 - 1.7 μmol/L). ENNs have also been found to inhibit drug efflux pumps and enzymes, which has stimulated interest in these molecules for use in medicine. ENNs were proven to inhibit ABC (ATP-binding cassette) transporters and, therefore, have started to be considered for their utility in cancer therapy [52]. Understanding the molecular mechanisms of enniatin toxicity is crucial for developing effective strategies to minimize its impact on human and animal health.

3.9. Beauvericin

Beauvericin (BEA) is composed of cyclic hexadepsipeptides consisting of alternating D-*α*-hydroxy-isovaleryl-(2-hydroxy-3-methylbutanoic acid) and amino acid units. This compound was first isolated from the culture of *Beauverina bassiana*, a pathogen of insects [56], and several other Fusarium species such as *F. acuminatum*, *F. acutatum*, and *F. anthophilum* [57], have been reported to produce beauvericin. Beauvericin was one of the active constituents of B. bassiana, and despite its toxicity to human cells, it was confirmed to have antimicrobial and anti-tumor activities [57].

Beauvericin is used as a commercial insecticidal agent because of the ability of the entomopathogenic fungus to become established and spread widely by insect movement, leading to its efficiency and the assurance that beauvericin production will remain within the threshold limits as stipulated by the EPA [58].

BEA has been reported to be a common contaminant of grains throughout the world, although the results from surveys reported thus far are dominated by data from Europe. Although the reported concentrations are highly variable, the maximum concentration was 520 mg/kg in a sample of visibly infected maize [59].

4. Global Occurrence of Mycotoxins in Food and Feed

Mycotoxins are found in food and feed worldwide, with varying levels of contamination depending on climatic conditions, geographical location, as well as agricultural practices. Food and feed commodities with reported mycotoxin contamination include grains (maize, wheat, rice, barley, etc.), nuts (peanuts, walnuts, almonds, etc.), seeds (sunflower, sesame, etc.), spices (chilli pepper), dried fruits (dates, cocoa, coffee, etc.), and animal feeds (silage, roughages, etc.).

Temperature is a critical factor influencing Ochratoxin A production in agricultural commodities, with optimal temperatures reported for different Aspergillus species: 25˚C - 30˚C for *A. ochraceus*, 10˚C - 20˚C for *A. carbonarius,* and

20˚C - 25˚C for *A. niger* [60]. In hot climatic countries such as South America, South Asia, and Africa, *Aspergillus* species are the major Ochratoxin A producers [61]. Similarly, in moderately temperate countries such as the United States, Canada, and Europe, *Penicillium* genus is the major Ochratoxin A producer [62]. The moisture content of the grains is an important factor needed for fungal growth and Ochratoxin A production.

Fumonisins are ubiquitous in distribution. They can be found on freshly harvested and stored agricultural produce globally. FB1 is found on rice, sorghum, pearl millet, maize and corn-based foods, beans, wheat noodles, curry, chili pickle, cowpea, triticale, asparagus, soya beans, etc. [63]. The occurrence of the mycotoxin in corn samples from 75 countries between January 2014 and June 2017 is shown in Table 2 below.

The occurrence of mycotoxins in feed and food products; Ochratoxin A, Fumonisin, Zeralenone, Deoxynivanelol, Aflatoxin, and the occurrence of the emerging mycotoxins (Beauvericin and Ennatins B) in different regions of the world, their concentrations, and the foods they attack are captured and elucidated in Tables 3-6.

Table 2. Occurrence of mycotoxins in 11,237 corn samples collected from 75 countries from January 2014 to June 2017.

All regions	AF	ZEN	DON	T-2	FUM	OTA
Number of samples tested	8885	9061	8013	4088	6563	3369
Median of Positive (ng/g)	3	76	581	24	1374	2
Average of Positive (ng/g)	22	239	1388	53	2536	17
Maximum (ng/g)	1352	16,495	43,770	976	171,920	889

AF = aflatoxins; ZEN, zearalenone; DON, deoxynivalenol; T-2, T-2 toxin; FUM, fumonisins; OTA, Ochratoxin A [64].

Table 3. OTA occurrence in food products.

Food sample	Country	Number of sample	% occurrence	Maximum (ug/kg)	Mean (ug/kg)
Corn, wheat	Pakistan	40	27.5	360	-
Corn, rice, wheat, and oat-based foods	USA	489	41	9.3	-
Palm dates	Tunisia & Algeria	27	11	6.07	58.7
Grounded sweet pepper	Italy	8	100	-	23.6
Beer	Mainly Portugal	85	10.6	11.25	-
Chicken meat	India	115	41	-	1.41
Red, rosé, and white wine	Serbia	113	52.2	0.134	0.026
Milk	China	120	25.8	18.8	-
Beef burger	Egypt	25	100	-	4.55

Extracted from [63].

Table 4. Occurrence of deoxynivalenol and aflatoxins in food and feed around the world.

Food/Feed	Country	Range (ug/kg)	Detection technique	Reference
		Deoxynivalenol		
Barley/Bakery products	Argetina	2360	HPLC-UV	[65]
	Brazil	310 - 15,500	LC-MS/MS	[66]
	Romania	0 - 4000	ELISA	[67]
	Tunisia	500 - 3600	HPLC	[68]
	Hungary	97 - 3065	HPLC	[69]
Corn	South Korea	3.3 - 232.56	HPLC	[70]
Corn/corm germ meal	China	100 - 4320.9/100 - 4402.7	HPLC	[71]
Corn flour/Courflakes	Serbia	931/878	HPLC	[72]
		Aflatoxins		
Almond	Turkey	1 - 13 (AFB1)	TLC	[73]
Cashew nuts	Brazil	0.60 - 31.50 (Total Afs)	ELISA	[74]
Chilies	United States	<2 (AFB1)	ELISA and TLC	[75]
Corn	Costa Rica	24 (Total Afs)	ELISA and HPLC	[76]
Corn	Zimbabwe	0.75 - 26.6 (AFB1)	HPLC	[77]
Dried Fruits	Pakistan	0.04 - 9.80 (AFB1)	HPLC	[78]
Ginger	Nigeria	0.11 - 9.52 (Total Afs)	HPLC	[79]
Milk (cow)	Iran	0.006 - 0.18 (AFM1)	HPLC	[80]
Nuts	Japan	0.17 - 2.59 (AFB1)	HPLC, HPTLC	[81]

Table 5. Occurrence of mycotoxins in feed samples around the world.

Regions	Sample size (n)	Mycotoxins Concentration (ug/kg)				
		AFB1	FUM	ZEN	DON	OTA
North Europe	1958	3.1	186	35	504	1.9
Central Europe	21,036	1.6	187	40	428	2.8
Southern Europe	3527	2.1	607	44	324	2.6
Eastern Europe	2382	3.4	87	15	153	3.6
North America	5471	8.7	652	102	505	2.4
Central America	367	3.9	929	60	316	2.5
South America	17,332	3.2	1,390	51	344	17
Middle East/North Africa	1075	2.4	347	31	236	3.1
Sub-Saharan Africa	208	23	789	38	352	7.2
South Africa	1077	2.2	266	30	363	2.2
Oceania	1695	2.0	106	105	158	3.6

Continued

South Asia	1136	20	288	37	96	4.6
Southeast Asia	4310	10	573	43	137	3.0
East Asia	13,232	10	810	90	418	2.9
Central Asia	15	1.4	13	1.5	28	22

AFB1 = aflatoxin B1, DON = deoxynivalenol, ZEN = zearalenone, FUM = fumonisin, OTA = ochratoxin A [13].

Table 6. Occurrence of Beauvericin and Enniatins B in food samples around the world.

Commodity	Origin	Positive/total samples	Concentration range (mg/kg)	Reference
Beauvericin				
Maize	USA	2/5	<0.1 - 0.5	[82]
Wheat	Finland	13/14	<0.01	[83]
Barley	Finland	22/22	<0.01 - 0.019	[83]
Rice	Spain	1/1	<0.17 - 11.8	[84]
Wheat	Morocco	7/25	<0.17 - 4	[85]
Ennatins B				
Wheat	Finland	14/14	<0.0004 - 18.3	[83]
Grain-based products	Italy	11/12	<0.0038	[86]
Maize	Tunisia	2/31	<0.145	[87]
Rice	Morocco	21/70	<0.145 - 26.2	[88]
Barley	Spain	2/4	<0.145 - 21.37	[84]

[82] (modified).

5. Toxicological Effects of Mycotoxins

Mycotoxicosis refers to the harmful effects of mycotoxins on the health of humans and animals [89]. The deleterious effect of mycotoxins as carcinogenic, mutagenic, teratogenic, nephrotoxic, or hepatotoxic agents to humans and animals is well reported in the literature. Mycotoxins can induce bone malformation, poor fetal organ development, and suppression of the immune system [90].

Humans and animals are exposed to mycotoxins through various routes, including direct ingestion of contaminated foods and indirect ingestion through consumption of products like meat, milk, and eggs from animals with prior exposure to these toxins [91]. Additionally, exposure to mycotoxins can occur through direct contact with contaminated substances, allowing the toxins to be absorbed through the skin [89]. The severity of mycotoxin exposure depends on the type of mycotoxin(s) consumed, the level of exposure, *i.e.*, the amount of toxins consumed, duration of exposure, the nutritional or physiological status of the host, and the synergistic effects of other compounds and metabolites within the host [92].

Acute and CHRONIC Toxicities of Mycotoxins and Their Health Impacts

Bioaccumulation of mycotoxins due to continuous intake of low levels of mycotoxins in food can result in chronic toxicity [93]. The aflatoxins B1 (AFB1), B2 (AFB2), G1 (AFG1), and G2 (AFG2) are of great toxicological importance due to the resulting metabolite from the hepatic metabolism of these toxins [94]. AFB1 is the most prevalent toxin in human and animal food/feed, and it is also the most dangerous hepatocarcinogen in mammals [95]. Common types of cancer induced by AFB1 include hepatocellular carcinoma (HCC), and liver cancer. AFB1 is metabolized by cytochrome P450 enzymes in the liver into a reactive intermediate called AFB1-8,9-exo-epoxide, which forms DNA adducts. These adducts lead to genetic mutations, particularly in the p53 tumor suppressor gene, resulting in carcinogenesis. The DNA adducts disrupt normal cellular processes, including DNA repair, and increase the risk of liver cancer [96].

Even though DON has been reported to be non-carcinogenic, acute toxicity can lead to severe health implications if contaminated food/feed is consumed over a long period of time (**Table 7**). In addition, cases of chronic toxicity have been reported to affect the gastrointestinal, immune, and reproductive systems [97].

Ochratoxins can disrupt some biological processes in the body, such as the production of protein, metabolism of calcium, lipid peroxidation, metabolism of sugars, and mitochondrial respiration [98]. Exposure of humans and animals to OTA-contaminated foods/feed over a short and long period could result in acute and chronic toxicity with resulting health impact (**Table 7**).

Fumonisins (FMNs) are hepatotoxic, immunotoxic, carcinogenic, and nephrotoxic [99]. FMNs have been reported to have a strong toxic effect on the lungs, liver, and kidneys of rats, cattle, and horses. FMNs caused leukoencephalomalacia (ELEM) in horses and pulmonary inflammation in pigs [100].

Trichothecenes (T-2/HT-2) negatively affect the synthesis of protein, thereby resulting in membrane dysfunction, immunosuppression, and cell death [101]. T-2/ HT-2 toxins have negative effects on the gastrointestinal tract and the epithelium of the skin, thereby resulting in nausea, diarrhea, vomiting, burns, blisters, redness of the skin, and necrosis [90]. Long-term exposure to trichothecenes can weaken the immune system, making individuals more susceptible to infections. In swine, the conversion of T-2 to HT-2 has been shown to have detrimental effects on blood cells and immune function [102].

Zearalenone (ZEA) is similar to an estrogenic hormone which is naturally produced in the body [103]. ZEA has been reported to have toxic effects on the blood, immune system, liver, and DNA [104]. ZEA is a non-steroidal estrogenic mycotoxin and has been reported as the likely cause of hyper-estrogenic syndromes in humans, and reproductive disorders in animals such as cattle, sheep, and swine [105]. Further information on the effects on human health is shown in **Table 7** below.

Table 7. Acute and chronic toxicities of mycotoxins and their health impacts.

Mycotoxins	Causative organism	Associated food/feed crops	Toxicity			
			Acute toxicity	Chronic Toxicity	Health Impact	References
Aflatoxins	*A. flavus* *A. parasiticus*	Maize, peanuts, tree nuts, copra, spices, cottonseed	Liver damage, vomiting, abdominal pain	Liver cancer, immunosuppression, stunted growth in children, chronic liver damage and degeneration, risk of enteropathies and rickets	Human carcinogens, particularly hepatocellular carcinoma (HCC)	[90] [109]
Fumonisins	*Fusarium verticillioides* *F. proliferatum*	Maize	Gastrointestinal issues, liver damage	Esophageal cancer, neural tube defects	Esophageal cancer and neural tube defects in humans, human tumor, throat cancer, grouped as 2B carcinogen, influence the metabolism of cholesterol	[98] [110]
Deoxynivalenol (DON)	*Fusarium graminearum* *F. culmorum* *F. crookwellense*	Wheat: Bread wheat, durum wheat, barley, maize, rice Oats, Rye	Vomiting, diarrhea, nausea, fever, dizziness, abdominal pain, headache, feed refusal, reduced weight gain	Immunosuppression, growth impairment	Feed refusal, vomiting, and immunological effects in animals and humans, acute gastroenteritis, and immunotoxicity	[97]
Ochratoxin A	*Penicillium verrucosum* *A. ochraceus* *A. carbonarius* *A. niger*	Maize, wheat, barley, oats, dried Meats and fruits, coffee, wine	Kidney damage	Kidney disease, cancer	Hepatotoxicity, nephrotoxicity, teratogenesis, and suppression of the immune system	[90] [111]
T-2 toxin, HT-2 toxin, and other Trichothecenes	*F. acuminatum*, *F. poae*, and *F. sporotrichiodes*	Heavily molded grains	Vomiting, abdominal pain, and diarrhea	white blood cell loss (leukopenia), bleeding from the nose and mouth, bone marrow depletion, and fever	Alimentary toxic aleukia (ATA), gastrointestinal hemorrhage, leukocytosis, circulatory shock, reduced cardiac, immunosuppression	[90]
Zearalenone	*F. graminearum*, *F. culmorum*, *F. cerealis*, *F. semitectum*, *F. equiseti*	Contaminated grains, cereals, animal products	Toxic effects on the blood, immune system, liver, and DNA	Immunotoxicity, Hepatoxicity genotoxicity	Cytotoxicity and oxidative stress, hyper-estrogenic syndromes in humans, and reproductive disorders in animals	[112] [113]

6. Regulatory Framework of Mycotoxins

Regulations for mycotoxins in food and feed have become more stringent globally

due to their toxic effects on humans and animals. Over 100 countries had established detailed guidelines to minimize mycotoxin exposure by 2003 [106]. The European Union has set maximum levels for 40 mycotoxins in food, and Codex Alimentarius sets global maximum limits for mycotoxins in food and feed, regulating aflatoxins, ochratoxin A, fumonisins, deoxynivalenol, and zearalenone [107].

The regulatory framework for mycotoxins is well-established in developed countries, with strict limits and enforcement mechanisms in place [105]. Risk assessment is the scientific basis behind mycotoxin regulations, determining the probability of toxicity or adverse reactions from human exposure to food-borne hazards. Regulations are established based on observed or verified reports of toxic effects of mycotoxins [107].

Aflatoxins are known to be carcinogenic and genotoxic; hence, the maximum levels are set very low to reasonably achieve.

Also, the advisory level for DON in finished wheat meant for human consumption was set at 1 mg/kg, and 10 mg/kg for beef, 5 mg/kg for swine, and 5 mg/kg for other animals. JECFA established the tolerable weekly intake level of 100 ng/kg body weight for ochratoxin A [113], compared to 120 ng/kg set by EFSA [115]. Table S1 presents the various maximum regulatory limits for mycotoxins in food in the European Union (EU) countries, Africa, and Europe, as set by the FAO international inquiry and other regulatory bodies globally.

However, countries in Africa and Asia face challenges in mycotoxin regulation, including limited scientific data, research funding, and analytical facilities. This has resulted in increased loss of trade, economic value of agricultural products, and health issues in humans and animals [116]. For example, in Malawi, regulatory limits are only set for peanuts meant for export, while in Nigeria, the initial aflatoxin regulatory limit was adjusted to align with EU limits to increase trade with the region [116]. In contrast, Morocco has comprehensive mycotoxin regulations comparable to EU standards, covering aflatoxins, ochratoxin A, patulin, fumonisins, deoxynivalenol, and zearalenone [116].

The World Health Organization (WHO) and the Joint FAO/WHO Expert Committee on Food Additives (JECFA) play crucial roles in establishing and enforcing mycotoxin limits. JECFA serves as a scientific advisory body to FAO, WHO, and the Codex Alimentarius Commission, evaluating the health risks of mycotoxins and recommending adequate protection measures. WHO encourages national authorities to monitor and ensure that levels of mycotoxins in foodstuffs comply with national and international maximum levels.

The establishment of regulations and control limits for mycotoxins in food and feed is essential to protect international trade and public health. Global harmonization of regulations is necessary to ensure consumer safety and facilitate international trade. The Codex standards and guidelines have been considered acceptable for international trade and assurances of consumer protection and safety.

The exclusive approach to the control of mycotoxins and setting regulatory

guidelines that is achievable in developing countries will need a holistic approach that focuses on the development of an efficient food safety management system. The development of an efficient food safety management system through the setting of regulatory limits must take into consideration the various components of an effective food safety management system such as: the creation of a monitoring program, control strategies through good manufacturing practices, processing, prevention strategies, decontamination through pre-processing or pretreatment, producer-consumer education advocacy, and setting of regulatory limits [118].

7. Emerging Issues and Future Directions

Emerging Mycotoxin-Related Challenges

Emerging mycotoxins (EMs) like nivalenol (NIV), enniatins (ENNs), beauvericin (BEA), diacetoxyscirpenol (DAS), fusaric acid (FUS), patulin (PAT), moniliformin (MON), and sterigmatocystin (STG) contaminate grains and feed commodities globally. These EMs, often overlooked and not routinely monitored, are increasingly recognized for their presence and impact. Fusarium species, such as *F. sambucinum*, *F. subglutinans*, and *F. tricinctum*, produce NIV, ENNs, BEA, DAS, FUS, and MON, while Aspergillus and Penicillium species produce STG and PAT.

Studies have shown that EMs co-occur with regulated mycotoxins, exacerbating adverse effects such as reproductive issues, immune system disorders, and impaired intestinal morphology [118] [119]. For example, enniatin B was found at concentrations of up to 13,335 μg/kg in triticale grown in France [120], while diacetoxyscirpenol was detected at 70 μg/kg in grains from Japan [121]. A recent metadata study revealed that NIV, ENNs, and BEA are the most predominant toxins, frequently contaminating cereal grains [122].

Climate change is significantly influencing the emergence of new mold species, further complicating food safety and biodiversity [123]. Rising temperatures, changing precipitation patterns, and increased CO_2 levels are altering the distribution and prevalence of mycotoxigenic fungi, such as Aspergillus and Fusarium species [124]. These changes are expected to increase mycotoxin contamination in crops, particularly in tropical and subtropical regions. Changing climate patterns are projected to alter host-pathogen interactions, fungal ecology, and mycotoxin production. For example, studies suggest that Aspergillus flavus may outcompete other species, increasing aflatoxin risks in Europe [124]. Similarly, warmer temperatures and drought conditions may favor Fusarium species, leading to increased fumonisin contamination. To manage these emerging risks, effective mitigation strategies are necessary, including cumulative risk assessments, multi-screening methods, and climate-smart agricultural practices.

8. Future Trends and Priorities in Mycotoxin Research and Management

The future of mycotoxin research and management is rapidly evolving, driven by advancements in technology, analytics, and interdisciplinary approaches. Emerg-

ing technologies, such as nanotechnology and advanced filtration systems, are being explored for efficient mycotoxin detection and removal. Real-time monitoring using sensors and AI algorithms is also being developed to enable swift and accurate detection of mycotoxins.

Artificial Intelligence (AI) is revolutionizing mycotoxin detection in food products. Recent studies have shown that AI-powered techniques, such as machine learning (ML), deep learning (DL), and hyperspectral imaging, can accurately detect mycotoxins like aflatoxins, fumonisins, and deoxynivalenol in various crops, including maize, wheat, peanuts, and coffee beans [125] [126]. A recent review by [125] revealed the capacity and efficiency of hyperspectral imaging combined with machine learning for non-destructive mycotoxin detection in cereal grains and nuts. Also, the application of machine learning and deep learning for detecting mycotoxins in food products is trending [126]. These studies further confirm the accuracy and potential for future advancement of these AI tools.

Nanotechnology has emerged as a powerful tool for detecting mycotoxins in food products. Recent studies have shown that nanotechnology-based sensors and assays can detect mycotoxins with high sensitivity and specificity [93] [127]. Gold nanoparticles, magnetic nanoparticles, and quantum dots have been used to detect mycotoxins such as aflatoxins, ochratoxin A, and fumonisins. The application of a graphene-based nanosensor for rapid and sensitive detection of ochratoxin A in food products has been reported [127]. Another study using gold nanoparticle-based sensors detected aflatoxin B1 in maize with a limit of detection of 0.01 ng/mL [93]. Also, a review of nanotechnology-based mycotoxin detection methods highlighted the potential of nanotechnology to improve food safety [128].

Biological detoxification is a promising approach for controlling mycotoxins, involving microorganisms or enzymes to degrade or transform mycotoxins into less toxic compounds [129]-[131]. This method has shown efficacy in reducing mycotoxin concentrations, such as Lactobacillus rhamnosus detoxifying aflatoxin B1 in maize by 90% [131] and Trichoderma harzianum reducing ochratoxin A in coffee by 80% [132], demonstrating the potential of microbes to improve food safety.

Integrated mycotoxin management, which combines multiple strategies such as good agricultural practices, resistant crop varieties, and holistic feed management, is also gaining attention.

Risk assessment and regulatory frameworks are crucial in ensuring consumer safety and facilitating international trade. Strengthening these frameworks and protocols is essential to protect public health and promote global food security. Sustainable solutions, such as cost-effective and environmentally friendly methods for mycotoxin mitigation, are also a priority. Collaboration and knowledge sharing among researchers, industries, and regulatory bodies are vital to address mycotoxin challenges and develop effective strategies for management and mitigation [93]. Further genetic manipulations of toxigenic fungi genomes are promising and may play a significant role in developing resistant crop varieties and

improving mycotoxin management in the future. Microbial interventions, through the metabolic interactions of beneficial microorganisms against virulent fungal strains, may be explored for bioremediation and mycotoxin degradation.

A multidisciplinary approach is necessary to effectively address the menace of mycotoxin contamination globally. Addressing the economic impact of mycotoxin contamination and mitigation strategies will inform policy decisions and ensure that effective solutions are implemented. By working together, researchers and industries can develop effective strategies for mycotoxin management and mitigation, ensuring a safer food supply chain [93].

9. Conclusion

Mycotoxins pose a significant threat to human and animal health, food security, and agricultural economies. The widespread presence of aflatoxins, fumonisins, ochratoxins, zearalenone, and trichothecenes in food and feed commodities underscores the need for effective control, surveillance, and prevention strategies. Climate change and emerging mycotoxins further exacerbate the issue, highlighting the importance of adopting Good Agricultural Practices, proper post-harvest handling, and robust regulatory frameworks. To mitigate the adverse effects of mycotoxins, it is essential to enhance stakeholder education, support farmers in adopting best practices, and prioritize the supply of safe and mycotoxin-free grains and livestock feeds. By working together, we can reduce the risks associated with mycotoxin contamination and promote a healthier and more sustainable food system.

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Conflicts of Interest

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