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Review

Toxicity, metabolism, and impact of mycotoxins on humans and animals

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Abstract

The worldwide contamination of foods and feeds with mycotoxins is a significant problem. Mycotoxins are secondary metabolites of molds that have adverse effects on humans, animals, and crops that result in illnesses and economic losses. Aflatoxins, ochratoxins, trichothecenes, zearelenone, fumonisins, tremorgenic toxins, and ergot alkaloids are the mycotoxins of greatest agro-economic importance. Some molds are capable of producing more than one mycotoxin and some mycotoxins are produced by more than one fungal species. Often more than one mycotoxin is found on a contaminated substrate. Factors influencing the presence of mycotoxins in foods or feeds include environmental conditions related to storage that can be controlled. Other extrinsic factors such as climate or intrinsic factors such as fungal strain specificity, strain variation, and instability of toxigenic properties are more difficult to control. Mycotoxins have various acute and chronic effects on humans and animals (especially monogastrics) depending on species and susceptibility of an animal within a species. Ruminants have, however, generally been more resistant to the adverse effects of mycotoxins. This is because the rumen microbiota is capable of degrading mycotoxins. The economic impact of mycotoxins include loss of human and animal life, increased health care and veterinary care costs, reduced livestock production, disposal of contaminated foods and feeds, and investment in research and applications to reduce severity of the mycotoxin problem. Although efforts have continued internationally to set guidelines to control mycotoxins, practical measures have not been adequately implemented. © 2001 Elsevier Science Ireland Ltd. All rights reserved.

Keywords: Mycotoxins; Aflatoxins; Ruminants; Non-ruminants

1. Introduction

Mycotoxins are produced mainly by the mycelial structure of filamentous fungi, or more specifically, the molds. Mycotoxins are secondary metabolites that have no biochemical significance

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in fungal growth and development (Moss, 1991). Toxigenic molds are known to produce one or more of these toxic secondary metabolites. It is well established that not all molds are toxigenic and not all secondary metabolites from molds are toxic. Examples of mycotoxins of greatest public health and agroeconomic significance include aflatoxins (AF), ochratoxins (OT), trichothecenes, zearalenone (ZEN), fumonisins (F), tremorgenic toxins, and ergot alkaloids. These toxins account for millions of dollars annually in losses worldwide in human health, animal health, and condemned agricultural products (Shane, 1994; Vasanthi and Bhat, 1998). Factors contributing to the presence or production of mycotoxins in foods or feeds include storage, environmental, and ecological conditions. Often times most factors are beyond human control.

In 1993, the WHO-International Agency for Research on Cancer (WHO-IARC, 1993a,b) evaluated the carcinogenic potential of AF, OT, trichothecenes, ZEN, and F. Naturally occurring AF were classified as carcinogenic to humans (Group 1) while OT and F were classified as possible carcinogens (Group 2B). Trichothecenes and ZEN, however, were not classified as human carcinogens (Group 3). The health hazards of mycotoxins to humans (Peraica et al., 1999) or animals (D'Mello and MacDonald, 1997) have been reviewed extensively in recent years.

Factors affecting the magnitude of toxicity of humans or animals consuming mycotoxin-contaminated foods or feeds, respectively, include species, mechanisms/modes of action, metabolism, and defense mechanisms. In early studies on AF, species specific acute toxicities were reported. The LD_{50} were 0.4, 1, and 500 mg/kg for ducklings, rats, and sheep, respectively (Wogan, 1966). Mechanisms and modes of mycotoxin action are only beginning to shed light on the interspecies and sometimes individual variations in toxic endpoints. For example, AF are known to bind DNA and induce mutagenic and carcinogenic effects in rats (Croy et al., 1978; Bennett et al., 1981; Foster et al., 1983; Muench et al., 1983). However, thymic depression, decreased T-cell function, and cellular immunity are the modes of AF action in bovines (Paul et al., 1977), ovines (Fernandez et al., 2000), and porcines (Pang and Pan, 1994).

Metabolism and defense mechanisms are important factors in understanding mycotoxin toxicity in specific species or individual animals. Specificity of such mechanisms are well demonstrated in the significant difference between ruminants and non-ruminants in handling mycotoxins. Ruminants have generally been more resistant to the adverse effects of mycotoxins (Wogan, 1966; Helferich et al., 1986). In vitro studies have shown the ability of the rumen microbiota to degrade mycotoxins (Ribelin et al., 1978; Kiessling et al., 1984; Swanson et al., 1987). Understanding the metabolic pathways of mycotoxins in ruminants and non-ruminants could enable researchers and public health officials to gain insight on how to assess the associated risks of mycotoxin exposure in various species. The first objective of this review was to identify the key mycotoxins commonly found in foods or feeds, demonstrate their negative effects on humans or animals, and explain their modes of action. The second objective was to explore the potential role of ruminants in detoxification of mycotoxins.

2. Mycotoxins in foods and feeds

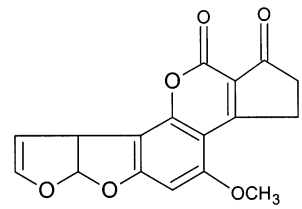
Mycotoxicoses in humans or animals are characterized as food or feed related, non-contagious, non-transferable, non-infectious, and non-traceable to microorganisms other than fungi. Clinical symptoms usually subside upon removal of contaminated food or feed (Robb, 1990). Although there are over 300 mycotoxins that have been isolated and chemically characterized (Betina, 1984), research has focused on those forms causing significant injuries to humans and their farm or companion animals. These include AF, OT, trichothecenes, ZEN, F, and ergot alkaloids. There have also been recent concerns over other toxins such as citrinin and sterigmatocystin.

Mycotoxins that adversely affect human or animal health are found mainly in post-harvest crops such as cereal grains or forages. These toxins are produced by saprophytic fungi during storage or by endophytic fungi during plant growth. Mycotoxins are generally lipophilic (except for FB) and, therefore, they tend to accumulate in the fat frac-

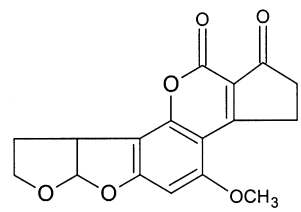
tion of plants and animals. Examples of fungal species and mycotoxins of biological and economical significance in animal agriculture are presented (D'Mello and MacDonald, 1997) in Table 1. In general, mycotoxins are categorized by fungal species, structure, and(or) mode of action. It should be noted, however, that a single species of fungi may produce one or several mycotoxins and individual mycotoxins may be produced by different fungal species. For example, AF are produced by several fungal species, have numerous structural variations, and have different modes of action depending on the target animal (Eaton et al., 1994).

2.1. Aflatoxins

Aflatoxins (A-flavus-toxins) are the most studied (>5000 publications) group of mycotoxins and are produced by different species of the genus *Aspergillus*. They were initially isolated and identified as the cause of the Turkey X disease (i.e. hepatic necrosis) in 1960 (Asao et al., 1963). Aflatoxins (Fig. 1 [AFB_1 and AFB_2], Fig. 2 [AFG_1 and AFG_2], and Fig. 3 [AFM_1 and AFM_2; metabolites found in milk]) are dihydrofuran or tetrahydrafurano moieties fused to a coumarin ring. There are over 20 isolated AF derivatives produced by several fungal species. For example, *Aspergillus flavus* produces AFB_1 and AFB_2



Aflatoxin B_1



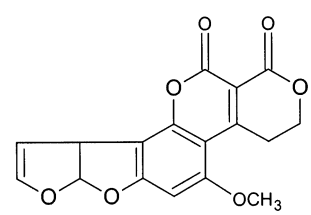
Aflatoxin B_2

Fig. 1. Chemical structure of aflatoxin B (AFB_1 and AFB_2).

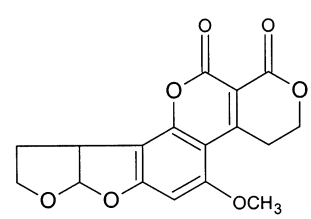
Table 1
Examples of fungal species and mycotoxins of biological and economical significance in animal agriculture (D'Mello and MacDonald, 1997)

Fungal species	Mycotoxin
Aspergillus flavus and *A. parasticus*	Aflatoxins
A. ochraceus, *Penicillium viridicatum*, and *P. cyclopium*	Ochratoxin A
Fusarium culmorum, *F. graminearum*, and *F. sporotrichioides*	Deoxynivalenol
F. sporotrichioides and *F. poae*	T-2 toxin
F. sporotrichioides, *F. graminearum*, and *F. poae*	Diacetoxyscirpenol
F. culmorum, *F. graminearum*, and *F. sporotrichioides*	Zearalenone
F. proliferatum, *F. verticillioides*	Fumonisins
Acremonium coenophialum	Ergopeptine alkaloids
A. lolii	Lolitrem alkaloids

D'Mello and MacDonald, 1997.

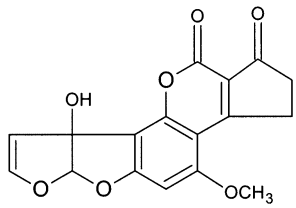


Aflatoxin G_1

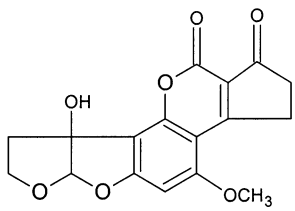


Aflatoxin G_2

Fig. 2. Chemical structure of aflatoxin G (AFG_1 and AFG_2).



Aflatoxin M_1



Aflatoxin M_2

Fig. 3. Chemical structure of aflatoxin M (AFM_1 and AFM_2).

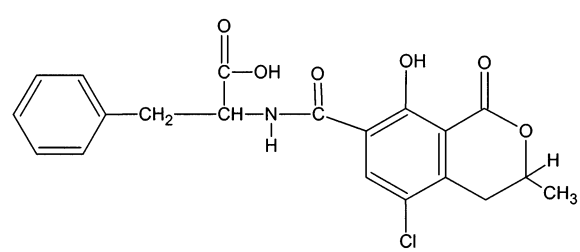
whereas *A. parasiticus* produces AFB_1, AFB_2, AFG_1, and AFG_2 (D'Mello and MacDonald, 1997). These AF, like many other heterocyclic compounds, fluoresce and are distinguished by their fluorescing properties. Both AFB_1 and AFB_2 fluoresce blue and AFG_1 and AFG_2 fluoresce yellow-green under ultraviolet light (Sargeant, 1963). Variations in the magnitude of toxicity exist among AF. For example, AFB_1 is the most toxic in both acute and chronic aflatoxicoses whereas AFM_1 (i.e. a metabolite in milk) is as acutely hepatotoxic as AFB_1 but not as carcinogenic (Carnaghan et al., 1963). These investigators illustrated the various relative potencies of different AF and reported LD_{50} values of 0.36, 0.78, 1.70, and 3.44 mg/kg of ducklings consuming AFB_1, AFG_1, AFB_2, or AFG_2, respectively. Such findings were later confirmed in vitro (Terao and Ueno, 1978) and in vivo (Cole and Cox, 1981). In these studies, the magnitudes of toxicity of AFG_2, AFB_2, and AFG_1 were found to be 10, 20, and 50% of that for AFB_1, respectively. Despite its acute toxicity and carcinogenic potential, AFM_1 is considered a detoxification product (Neal et al., 1998).

2.2. Ochratoxins

Ochratoxins are metabolites of both *Aspergillus* and *Fusarium* species which are chemically described as 3,4-dihydromethylisocoumarin derivatives linked with an amide bond to the amino group of L-β-phenylalanine (Cole and Cox, 1981). These compounds are known for their nephrotoxic effects (renal damage) in poultry (Lanza et al., 1980; Manning and Wyatt, 1984). They also are acutely toxic in rats (Wannemacher et al., 1991) and mice (Carlton and Tuite, 1977) and may promote tumors in humans (Krogh, 1978). Ochratoxin A (OTA, Fig. 4) is the most toxic compound of this group. It was first isolated from *A. ochraceus* (van der Merwe et al., 1965) and was later shown as a secondary metabolite of *Penicillium* species in temperate climates (Smith and Ross, 1991).

2.3. Trichothecenes

Trichothecenes are compounds containing sesquiterpene rings characterized by a 12,13-epoxy-trichothec-9-ene nucleus. They have different constituents on positions 3, 4, 7, 8, and 15 of the molecule. Trichothecenes are mainly produced by several *Fusarium* species (e.g. *F. sporotrichioides*, *F. graminearum*, *F. poae*, and *F. culmorum*) and can be produced by members of other genera such as *Myrothecium* (Tamm and Breitenstein, 1984) and *Trichothecium* (Jones and Lowe, 1960). Trichothecenes include T-2 toxin (Fig. 5), diacetoxyscirpenol (DAS, Fig. 6), deoxynivalenol (known as DON (Fig. 7) or vomitoxin), and nivalenol. Both T-2 toxin and DAS are the most



Ochratoxin A

Fig. 4. Chemical structure of ochratoxin A (OTA).

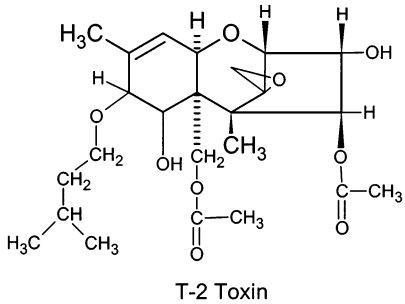


Fig. 5. Chemical structure of T-2 toxin.

toxic and are soluble in non-polar solvents (e.g. ethyl acetate and diethyl ether) whereas DON and its parent compound nivalenol are soluble in polar solvents such as alcohols (Trenholm et al., 1986).

2.4. Zearalenone

Zearalenone (Fig. 8) is a phytoestrogenic compound (Diekman and Green, 1992) known as 6-(10-hydroxy-6-oxo-*trans*-1-undecenyl)-β-resorcylic acid μ-lactone. It is a metabolite primarily associated with several *Fusarium* species (i.e. *F. culmorum*, *F. graminearum*, and *F. sporotrichioides*) with *F. graminearum* being the species most responsible for the estrogenic effects commonly found in farm animals (Marasas, 1991). Alcohol metabolites of ZEN (i.e. α-zearalenol and β-zearalenol) are also estrogenic (Cheeke, 1998a).

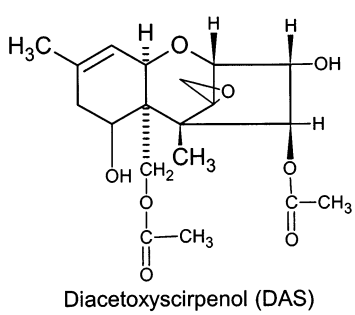


Fig. 6. Chemical structure of diacetoxyscirpenol (DAS).

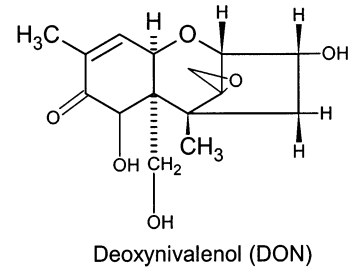


Fig. 7. Chemical structure of deoxynivalenol (DON).

2.5. Fumonisins

Fumonisins (B_1 and B_2, Fig. 9) are cancer-promoting metabolites of *F. proliferatum* and *F. verticillioides* that have a long-chain hydrocarbon unit (similar to that of sphingosine and sphinganine) which plays a role in their toxicity (Wang et al., 1992). Fumonisin B_1 (FB_1) is the most toxic and has been shown to promote tumor in rats (Gelderblom et al., 1988) and cause equine leukoencephalomalacia (Marasas et al., 1988) and porcine pulmonary edema (Harrison et al., 1990). The naturally co-occurring aminopentol isomers (formed by base hydrolysis of the ester-linked tricarballylic acid of FB_1) have been suggested to exert toxic effects due to their structural analogy to sphingoid bases (Humpf et al., 1998).

2.6. Moniliformin

Moniliformin (i.e. a potassium or sodium salt of 1-hydroxycyclobut-1-ene-3,4-dione, Fig. 10) is produced by several *Fusarium* species (mainly *F. proliferatum*) and is usually found on the corn kernel. It can be transferred to next generation crops and survive for years in the soil (Guzman and Casteel, 1994). Although both FB_1 and

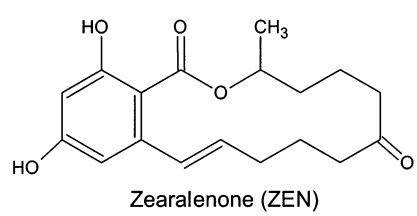


Fig. 8. Chemical structure of Zearalenone (ZEN).

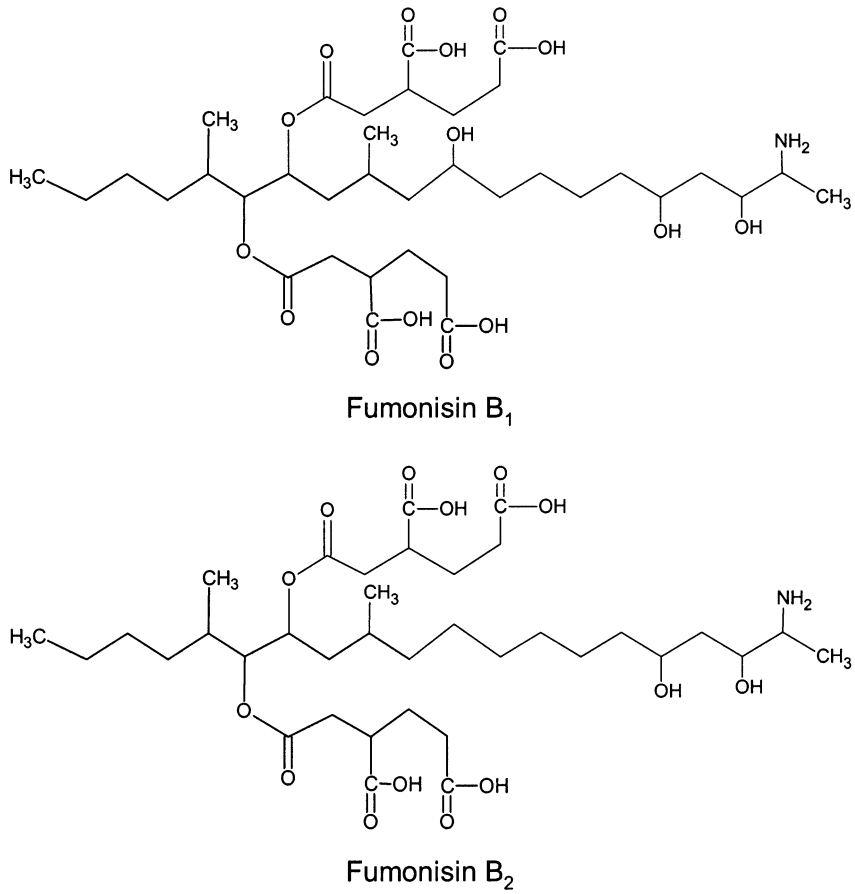


Fig. 9. Chemical structure of Fumonisins B_1 and B_2.

moniliformin are produced by the same fungal species (*F. proliferatum*) no structural resemblance (Figs. 9 and 10) is found between the two toxins. Both toxins are shown in several FDA studies to be ubiquitous in corn in the US (Price et al., 1993).

2.7. Endophytic tremorgens and ergot alkaloids

Several colonizing toxigenic fungal species (e.g. *Acremonium lolii*, *A. coenophialium*, *Claviceps purpurea*, and *Penicillium* spp.) may thrive exclusively on live forages. These endophytic fungi thrive in the tissues of several grasses (e.g. tall fescue) and may have a mutualistic relationship with their hosts. The mycotoxins produced may protect the grass against consumption by herbivores while the plant serves as a host. *A. lolii* is an endophytic fungus that thrives on perennial ryegrass and produces indole-terpene neurotoxins (Miles et al., 1992) called tremorgens such as lolitrem B toxin (Fig. 11). Perennial ryegrass staggers in livestock have been linked to lolitrem B toxin from *A. lolii*

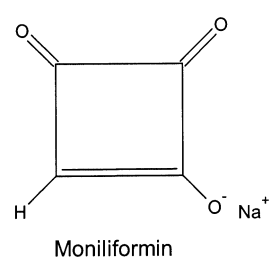


Fig. 10. Chemical structure of moniliformin.

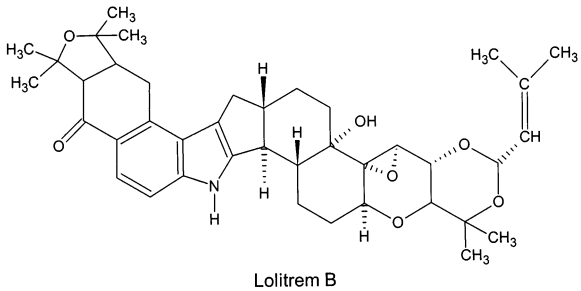


Fig. 11. Chemical structure of lolitrem B toxin.

and to other tremorgens (e.g. penetrem B and verruculogen) produced by several *Penicillium* species including *P. crustosum* and *P. verruculosum*, respectively (Miles et al., 1992). Fescue foot, summer fescue toxicosis, and fat necrosis in cattle have been linked to tall fescue with growth of *A. coenophialium*.

Other *Acremonium* species and *C. purpurea* are also responsible for sleepygrass toxicosis in livestock which is a result of ergot alkaloids (e.g. ergotamine, ergostine, and ergocristine) produced by these fungal species. Ergot alkaloids are structurally related to the hallucinogenic drug known as lysergic acid diethyl amide. Ergotamine (Fig. 12) and other lysergic acid amide derivatives in the perennial grass *Stipa robusta* are commonly associated with toxicoses in sheep (D'Mello and MacDonald, 1997). The two types of ergotism known in humans and animals are convulsive and gangrenous (Cheeke, 1998a).

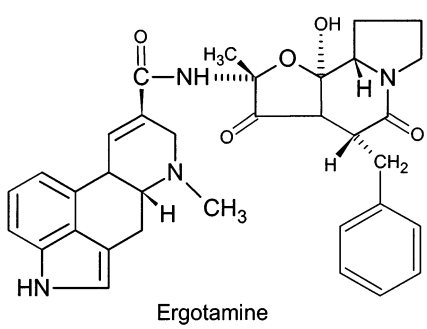


Fig. 12. Chemical structure of ergotamine.

3. Negative effects of mycotoxins on non-ruminants

Early studies on the effects of acute aflatoxicosis indicated various toxicities in different animal species (Wogan, 1966). In monogastrics, variable responses have been shown with all mycotoxins (Cheeke, 1998a). For example, pigs have been shown to be very sensitive to T-2 toxin, DON (Friend et al., 1992), and ZEN (Biehl et al., 1993). Poultry also are adversely affected by both T-2 and DON but are very resistant to the estrogenic effects of ZEN (Cheeke, 1998a). Various degrees of mycotoxicoses from natural sources occur in different animal species because of the wide range of feed ingredients used and the differences among and within species. Experiments and case studies on mycotoxicoses in non-ruminant species have been summarized in the following sections.

3.1. Poultry

Early investigations (1960) of the sudden death of 100 000 turkey poults consuming groundnuts in England linked AF (from *A. flavus*) to acute hepatic necrosis and hyperplasia of the bile ducts of the intoxicated birds (Newberne and Butler, 1969). Chickens have been shown to bruise and hemorrhage from AF (Tung et al., 1971). Injection of AFB_1 alone or in combination with OTA showed impaired development and increased mortality in chicken embryos (Edrington et al., 1995). In India, high levels of AF (ranging from 0.2 to ≥ 1 mg/kg) in combination with other mycotoxins (i.e. OTA and/or trichothecenes) in poultry feed have resulted in diseases such as hepatitis, salmonellosis, coccidiosis, and infectious bursal disease (Jand et al., 1995). These investigators also indicated the presence of mycotoxin-contaminated feed at 29 (78.3%) and 23 (82.1%) of the broiler and layer farms tested, respectively.

The negative effects of mycotoxins on chicken performance have been demonstrated in numerous studies. For example, feeding a high level (3.5 mg/kg of feed) of an AF mixture (i.e. 79% AFB_1, 16% AFG_1, 4% AFB_2, and 1% AFG_2) to broilers reduced their body weight and increased their liver and kidney weights (Smith et al., 1992).

Aflatoxins also increased blood urea-N and decreased serum levels of total protein, albumin, triglycerides, and phosphorus. Feeding OTA (0.3–1 mg/kg of feed) to broilers (Bitay et al., 1979) reduced glycogenolysis and resulted in a dose-dependent glycogen accumulation in the liver. These negative metabolic responses were attributed to inhibition of cyclic adenosine 3′, 5′ monophosphate-dependent protein kinase and were reflected in decreased efficiency of feed utilization and teratogenic malformations (Bitay et al., 1979). The activities of other enzymes (e.g. alkaline phosphatase, acid phosphatase, lactate dehydrogenase, and succinate dehydrogenase) in several organs (e.g. heart, liver, spleen, and pancreas) of 1-week-old chicks also were altered by ingesting feed contaminated with *F. roseum*. Such change in enzyme activity resulted in metabolic and cellular respiratory disorders, reduced body weight gain, and tissue necrosis (Beri et al., 1991).

The negative impact of AF on the immune response was investigated in vitro (Neldon-Ortiz and Qureshi, 1992). In this study, chicken peritoneal macrophages were exposed to various levels of AFB_1 alone (i.e. 5, 10, and 20 μg/ml of culture medium) or with microsomal mixed function oxidase (AFB_1 levels were 0.01, 0.1, 0.5, 1, and 5 μg/ml) that activates AFB_1 to its toxic form (i.e. AFB_1-epoxide). The exposure to AFB_1 alone resulted in a dose-dependent reduction in macrophage adherence potential and an increase in cell damage. The activated form of AFB_1 (i.e. incubated with mixed function oxidase) also induced similar responses and caused morphological alterations and reduced phagocytosis.

Fusarium mycotoxins have been shown to adversely effect poultry. In addition to reduced feed intake and body weight gain, buccal-oral ulceration and plaque formation were observed (Hoerr et al., 1982) when 7-day-old chicks were given T-2 toxin (4 or 16 mg/kg of feed) or DAS (4 or 16 mg/kg of feed). Similar effects were also observed in 1-day–3-week-old chicks consuming T-2 toxin at 6 mg/kg of feed (Kubena et al., 1994) and in 24–25-week-old hens consuming DAS at 20 mg/kg of feed (Brake et al., 2000). Interestingly, fertility was increased in hens (67–69-week-old) and decreased in roosters (25–27-week-old) when DAS was fed at ≤ 5 and 10 mg/kg of feed, respectively (Brake et al., 1999).

3.2. Pigs

Swine are among the most sensitive species to mycotoxins. For example, Southern and Clawson (1979) have demonstrated that total AF at 385 μg/kg of feed was close to the maximum tolerance level for finishing pigs. In their study, feeding diets containing various levels (i.e. 0.02, 0.385, 0.75, and 1.48 mg/kg of feed) of AF (mostly AFB_1 with small amounts of AFG and AFB_2) reduced average daily gain linearly at 0.385 mg/kg of feed or higher while feed efficiency was decreased only at the highest level (i.e. 1.48 mg/kg of feed). Liver weights (as a percentage of body weight) were increased by feeding AF at 0.385 mg/kg of feed or higher, but hepatocellular lesions were only found in pigs receiving the highest AF level (i.e. 1.48 mg/kg of feed). Progression of aflatoxicosis also was evaluated in barrows given graded levels (1, 2, 3, or 4 mg/kg of feed) of AF (Harvey et al., 1988). Body weight gains were decreased in a linear fashion (from 7 kg for the control group to 4, 3, 2.5, and 0.2 kg for barrows receiving 1, 2, 3, or 4 mg of AF/kg of feed, respectively) over a 4-week feeding period. In a study by Huff et al. (1988), swine response to AF (2 mg/kg of feed), OTA (2 mg/kg of feed), or both was evaluated. Compared with the control group, body weight gains were reduced by 26, 24, and 52% for animals consuming diets containing AF, OTA, or both, respectively. It was concluded, therefore, that both toxins have an additive effect on swine weight gain. The reduction in growth rate of swine consuming mycotoxins (Southern and Clawson, 1979; Harvey et al., 1988; Huff et al., 1988) may have been due to decreased protein synthesis as indicated by the lower serum concentrations of albumin, total protein, and urea-N (Harvey et al., 1988; Lindemann et al., 1993). It should be noted, however, that OTA can cause an increase in body weight due to increased water retention (Glavitis and Vanyi, 1995). Additional symptoms of ochratoxicosis in swine include anorexia, faintness, uncoordinated movement, and increased water intake and urination (Glavitis

and Vanyi, 1995). Swine also are sensitive to other mycotoxins such as fumonsins and ergot alkaloids. Fumonisin B_1, for example, has been shown to cause pulmonary edema and heart and respiratory dysfunctions (Diaz and Boermans, 1994). Symptoms of porcine pulmonary edema included dypsnea, cyanosis, and death (Osweiler et al., 1992; Diaz and Boermans, 1994). It was suggested that hepatotoxicity can induce membrane fragments to be released into the bloodstream of the pig and, therefore, cause intense macrophage response in the lungs (Cheeke, 1998a). Recently, another mode of action has been suggested at the heart level due to Ca^{++} channel interference by sphingosine (Constable et al., 2000). Ergot alkaloids were reported to cause staggers and to compromise lactation in swine (Diekman and Green, 1992).

The swine immune response to AF has been inconsistent. Miller et al. (1978) reported decreased lymphocyte blastogenic response to mitogens, reduced macrophage migration, and depressed delayed hypersensitivity when AF was fed at 0.4 to 0.8 mg/kg of feed for 10 week. Other studies, however, have shown that swine humoral immune response was not altered by feeding mixed AF at levels ranging from 0.4 to 0.8 mg/kg of feed (Miller et al., 1981) to acutely toxic levels as high as 500 mg/kg of feed (Panangala et al., 1986). In vitro studies have confirmed that the immunosuppression caused by AF (140 or 280 μg/kg of feed) only occurs at the cellular and not the humoral level (van Heugten et al., 1994). Other in vitro studies (Pang and Pan, 1994) have shown inhibition of DNA synthesis in porcine lymphocytes when AFB_1 was added to the medium at various levels (0.1–10 000 ng/ml of medium).

Mycotoxic porcine nephropathy is a serious disease commonly associated with pigs consuming feed contaminated with OTA. In addition to the enlarged and pale kidneys (with vascular lesions and white spots), morphological changes include damage proximal tubules, atrophy of tubular epithelium, renal fibrosis, and hyalinization of glomeruli (Krogh, 1991). Kidney accumulation of OTA (from feed) was shown to be dose-dependent (Krogh et al., 1974) when pigs were exposed to various levels (0.2, 1, and 4 μg/kg of feed) for an extended period of time (3–4 months).

Negative effects of the mycotoxin ZEN on swine reproductive function have been demonstrated (Cantley et al., 1982; Etienne and Jemmali, 1982; Long and Diekman, 1984, 1986; Flowers et al., 1987; Diekman and Green, 1992). Pigs have been shown to draw the toxic forms of ZEN back from the circulating glucuronide conjugate (Biehl et al., 1993). For this reason the estrogenic effects of ZEN have been pronounced and prolonged in pigs. An extensive study in Hungarian farms (Glavitis and Vanyi, 1995) showed swelling of the vulva and mammary glands and occasional vaginal and rectal prolapses in sexually mature gilts consuming feed contaminated with ZEN. Similar and additional reproductive problems also have been demonstrated by adding ZEN to the diets of sows or gilts. For example, feeding gilts 95% pure ZEN (25–100 mg/kg of feed) from weaning through gestation resulted in constant estrus, pseudopregnancy, and subsequent infertility (Chang et al., 1979). Similar effects also were found when lower levels (3.6–20 μg/kg) of ZEN were fed (Cantley et al., 1982; Flowers et al., 1987) for shorter time (5–20 days). Other estrogenic effects of ZEN on gilts or sows included edematous uterus, ovarian cysts, increased follicular maturation and number of stillborns, and decreased fertilization rate (Glavitis and Vanyi, 1995). In the same study, ZEN induced germinal epithelial degeneration and altered sperm formation in boars. Reproductive disorders (e.g. atrophy of the ovaries and uterus, ovarian degeneration, and glandular dysfunction of the endometrium) also have been reported when sows were exposed to feed contaminated with T-2 toxin. Signs of prenatal T-2 toxicosis (e.g. glandular dysfunction of the endometrium, gastrointestinal edema, and hematopoesis leading to death) were also observed in suckling piglets.

3.3. Horses

The history of myctoxicosis and poisoning in equine has been reviewed by Asquith (1991). In a case study, mature horses consuming AFB_1-contaminated feed (58.4 μg/kg) were jaundiced and

anorexic before death (Greene and Oehme, 1976). Post-mortem examinations revealed enlarged livers, kidney damage, and lesions of bile-duct hyperplasia. In other cases (Asquith and Edds, 1981), equine aflatoxicosis has been characterized by depression, lameness, and death. Post-mortem examinations revealed subcutaneous and enteric hemorrhage, enlarged kidneys, enlarged necrotic livers, and hepatic, nephritic, and myocardial lesions. Studies with ponies have shown damage in the skeletal muscles and heart along with liver dysfunction when acute lethal doses of AFB_1 were administered (Asquith and Edds, 1981). Post-mortem examination of horses consuming corn contaminated with a mixture of AF (AFB_1, AFB_2, and AFM_1 at 114, 10, and 6 μg/kg, respectively) revealed severe hepatic lesions (Vesonder et al., 1991).

The greatest mycotoxin risks to equine identified thus far are the toxins produced by *F. moniliforme* which has been implicated in equine leukoencephalomalacia and acute neurotoxicity (Placinta et al., 1999). These diseases were attributed to consumption of corn contaminated with FB_1 and moniliformin toxins. Symptoms of equine leukoencephalomalacia include ataxia, paresis, apathy hypersensitivity, impaired locomotor function, necrosis of cerebral white matter, and lesions in the cerebral cortex. Bean-hulls poisoning is another mycotoxin-related disease that has been known in Hokkaido (Japan) for 7 decades because of the availability of bean hulls as a cheap source of feed and bedding for horses (Asquith, 1991). Clinical symptoms include central nervous system dysfunction, rapid heartbeat, diminished ocular reflexes, and death. Ueno et al. (1972) demonstrated that 60% of the fungal isolates causing such problems belonged to *F. sporotrichoides* and the toxins produced were T-2 toxin and DAS.

3.4. Dogs and cats

The effects of mycotoxins on companion animals are severe and can lead to death. As early as 1952, a case of hepatitis in dogs was directly linked to consumption of moldy food (Devegowda and Castaldo, 2000). Following the discovery of AF (Asao et al., 1963), the agent responsible for the 1952 case was identified as AFB_1 (Newberne et al., 1966) and the symptoms of aflatoxicoses in dogs were elucidated (Newberne et al., 1966; Ketterer et al., 1975). In the case study by Ketterer et al. (1975), three dogs on a farm in Queensland became ill (severe depression, anorexia, and weakness) and died at different times within a month following consumption of a commercial dog food mixed with AF-contaminated bread. The vomitus specimens from one dog contained high levels of AF (100 μg/g of AFB_1 and 40 μg/g of AFG_1). In addition to hepatitis and sudden death in dogs, symptoms of acute aflatoxicoses in both dogs and cats include vomiting, depression, polydipsea, and polyuria. Death usually occurs in 3 days with LD_{50} levels ranging from 0.5 to 1.0 mg/kg in dogs and 0.3 to 0.6 mg/kg in cats depending on the age of the animal (Newberne et al., 1966). Necropsy observations revealed enlarged livers, disseminated intravascular coagulation, and internal hemorrhaging. In subacute aflatoxicosis (at 0.5–1 mg/kg of petfood over 2–3 week), dogs and cats become lethargic, anorexic, and jaundiced (Newberne et al., 1966). This can be followed by disseminated intravascular coagulation and death. Such impaired blood clotting has been reported in other animals such as rats, rabbits, goats, and chickens (Bababunmi et al., 1997). In chronic aflatoxicoses (at 0.05–0.3 mg/kg of petfood over 6–8 week), dogs and cats had clinical signs similar to those for the subacute phase but jaundice was the predominant manifestation. Histopathology of animals with chronic aflatoxicoses revealed shrunken livers with extensive fibrosis (Newberne et al., 1966; Ketterer et al., 1975). The development of accurate screening methods for AF in recent years has led to a vast improvement in the exclusion of AFB_1 from petfood. As a result, there have been only two isolated cases of aflatoxicoses in dogs in the US since 1980 (Rumbeiha, 2000).

Deoxynivalenol is a major health concern for companion animals and it contaminates petfood via corn even after processing (Scott, 1984). In a recent study (Hughes et al., 1998), the effects of dietary DON on dogs and cats were investigated.

Food refusals were noted when DON levels exceeded 4.5 mg/kg of dog food and 7.7 mg/kg of cat food. This observation suggested a higher sensitivity in dogs than in cats. It has been shown that food consumption was significantly reduced at DON levels of 4.5 μg/kg of dog food and of 7.5 μg/kg of cat food. In the same study (Hughes et al., 1999), vomiting was noted at different times in dogs and cats (depending on the mycotoxin dose) during the 14 day of DON feeding. Dogs vomited for 4 or 9 days at DON levels of 8 and 10 μg/kg, respectively, while cats vomited for 10 days at DON level of 10 μg/kg. Due to the variable toxicity responses to DON in dogs and cats, it was suggested that DON levels in petfood should not exceed 0.5 μg/kg (Bird, 2000).

Studies have shown that trichothecenes have adverse effects on dogs and cats. In a study by Borison et al. (1991), T-2 toxin given to cats intravenously at 2 mg/kg resulted in hypovolemia and death. Sub-lethal T-2 toxicity in cats has been shown to lower white blood cell counts (Devegowda and Castaldo, 2000). Daily subcutaneous administration of 0.05 mg/kg reduced white blood cell count after 7 days while oral administration of 0.06 mg/kg reduced white blood cell count after 24 days. Food consumption was significantly reduced at levels of 4.5 μg/kg DON in dog food and 7.5 μg/kg DON in cat food (Hughes et al., 1999).

As with other species, the kidney is the primary target organ of OTA in dogs and cats. In a study with dogs (Kitchen et al., 1977), pacing and vomiting were observed at an OTA dose of 0.2 mg/kg. At doses between 0.2 and 3.0 mg/kg symptoms of intoxication in dogs included anorexia, polydipsea, polyuria, anxiety, prostration, and death. The necropsy findings (Kitchen et al., 1977) included epithelial degeneration (proximal tubules), mucohemorrhagic enteritis (cecum, colon, and rectum) and necrosis of the lymphoid tissues (spleen, tonsil, thymus, and peripheral lymph nodes).

3.5. Rats and mice

Rats have been used extensively for decades as a model for human mycotoxicoses especially with regard to the carcinogenic potential of AF. This model system, however, has been a subject for debate due to the differences in the detoxification mechanisms between rats and humans as shown by cytosolic conjugation of AFB_1 in vitro (Raney et al., 1992).

In early studies, the LD_{50} for AFB_1 were established in rats at various levels such as 0.5–7 mg/kg (Butler, 1964; Wogan and Newberne, 1967) and 6–18 mg/kg (Patterson, 1973) depending on the method of administration (i.v. or oral, respectively). Necropsy results revealed hepatic damage (i.e. lesions, necrosis, and biliary proliferation) similar to that of other species. In another study (Newberne and Butler, 1969), oral administration of AFB_1 at 5 mg/kg for 9 week in rats has resulted in 100% hepatocellular carcinomas. A recent study (Stetinova et al., 1998), has suggested secondary effects of AFB_1 on the gastrointestinal tract due to changes in the liver detoxification mechanisms and possible reductions in nutrient uptake. In contrast to rats, mice are generally resistant to the hepatocarcinogenic effects of AFB_1. This may explain the high level of glutathione-*S*-transferase (GST) activity in mice challenged with AFB_1 (Quinn et al., 1990). Contrary to the hepatocellular carcinomas commonly found in rat studies with AFB_1 (Butler, 1964; Wogan and Newberne, 1967; Patterson, 1973), mice given AFB_1 by intraperitoneal injection at 0.02 mg/kg of body weight for 12 injections over 3 weeks (average 5.6 mg/kg body weight) have expressed pulmonary tumors (Weider et al., 1968).

Other mycotoxins such as FB_1 also have been implicated in hepatic tumor formation in rats (Gelderblom and Snyman, 1991). In this study, 25 rats were given FB_1 at 50 mg/kg of feed in a 26-month experiment. Similar to AFB_1, the liver was the primary target for FB_1 toxicity. Necropsy results showed hepatocellular carcinoma in 66% of the 15 rats examined during the last 8 months of the study.

The negative effects of tricothecenes on rats have been known for decades and, as a result, the rat has been used extensively as a model for trichothecene toxicity tests. Studies with T-2 toxin have shown the LD_{50} of its oral administration to range from 2.8 to 3.8 mg/kg (Kosuri et al., 1971; Bamburg, 1972; Kravchenko et al., 1983). Oral

administration of T-2 toxin at levels ranging from 5 to 25 μg/kg of feed for extended periods (up to 16 week) reduced feed intake in a dose-dependent manner and caused gastric ulcers and thymic depression (Marasas et al., 1969; Hayes and Schiefer, 1980; Rukmini et al., 1980). Suneja et al. (1984a) also demonstrated reduced nutrient uptake and lipid metabolism (i.e. elevated levels of triglycerides, free cholesterol, total phospholipids, and phosphatidyl choline) when rats were given a daily oral dose (1.5 mg/kg) of T-2 toxin. Suneja et al. (1984b) further reported reduced intestinal uptake of glucose and tryptophan and decreased levels of several enzyme systems (i.e. sucrase, lactase, and Na^+-K^+-ATPase) in the intestinal mucosa.

Symptoms of acute T-2 toxicity in rats include lethargy, reduced feed intake, decreased body temperatures, increased number of white blood cells and lymphocytes by 3-fold, hypertension, and finally tachycardia precedes hypotension and death (Wannemacher et al., 1991). In acute rat studies, the LD_{50} of T-2 toxin (i.v.) was reported to range from 0.7 to 0.9 mg/kg (Feuerstein et al., 1985; Fairhurst et al., 1987). The effects of another tricothecene (i.e. DAS) on rats have been investigated (Sato and Ueno, 1977; More et al., 1990) and LD_{50} was established at 7.3 mg/kg when DAS was given orally. Lower doses of DAS have been shown to induce changes in the mucus-producing cells of the fundic glands of the rat stomach (More et al., 1990).

In a study with DON, the effects of contaminated feed (levels ranging from 0.1 to 104 mg/kg) on rats were investigated (Bosch et al., 1989). The toxicity symptoms included gastrointestinal hemorrhaging, hematuria, and death. In vitro studies, however, have shown that neither T-2 nor DON had hemolytic effects on rat erythrocytes exposed to levels as high as 130 μg/ml (Rizzo et al., 1992).

The LD_{50} of orally administered OTA in rats was established at 21 mg/kg with death occurring at 24–72 h following anorexia and depression. Necropsies revealed an inflammation of the gastrointestinal tract tissues and lesions in the liver and kidney (Purchase and Theron, 1968). The carcinogenic potential of OTA in rats has been shown in several chronic studies (Kane et al., 1986; Boorman et al., 1992). For example, OTA carcinogenicity was illustrated (Kane et al., 1986) when rats were fed OTA at 4 mg/kg of feed every 48 h. At the end of the 12-week dosing period, DNA strand breaks were noted in the liver and kidney tissues.

3.6. Humans

In general, effects of mycotoxins on humans are limited to case studies. Similar to rat studies and *Salmonella* assays for mutagenicity, case studies on AF exposures have provided insight to toxicity in humans. Acute AF exposures have been associated with epidemics of acute toxic hepatitis in areas of China and Africa with death rates ranging from 10 to 60% (Bhat and Krishnamachari, 1977). Studies on an individual attempting suicide by ingesting purified AF have demonstrated that single doses are not as effective in humans as long-term doses (Willis et al., 1980). Toxicity symptoms in a young woman, who attempted suicide with AF in the amounts of 5.5 mg over 2 days and 35 mg over 2 weeks (6 months from the initial dose), included transient nonpruritic macular rash, nausea, and headache. The woman recovered completely and had no significant signs of liver injuries when examined 14 years later (Willis et al., 1980). Results of this case and others suggested that extended subacute doses, as seen in dietary exposures in certain countries, may be required for inducing the lethal acute toxic effects (Willis et al., 1980; Peraica et al., 1999).

The largest risk of AF to humans is usually the result of chronic dietary exposure. Such dietary AF exposures have been associated with human hepatocellular carcinomas, which may be compounded by hepatitis B virus. Approximately 250 000 deaths are caused by hepatocellular carcinomas in China and Sub-Saharan Africa annually (Groopman et al., 1992) and are attributed to risk factors such as high daily intake (1.4 μg) of AF (Wild et al., 1992) and high incidence of hepatitis B (Kensler et al., 1991; Wild et al., 1992). Aflatoxins have been found in tissues of children suffering from Kwashiorkor and Reye's syndrome and were thought to be a contributing factor to these diseases (Becroft and Webster, 1972). Reye's

syndrome, which is characterized by encephalopathy and visceral deterioration, results in liver and kidney enlargement and cerebral edema (Becroft and Webster, 1972; Blunden et al., 1991).

Another potential human carcinogen is OTA which was implicated as the causative agent in epithelial tumors of the upper urinary tract in the Balkan regions (Krogh, 1978). The condition is known as Balkan Endemic Nephropathy. Despite the seriousness of the problem, studies have not completely identified the mechanism or extent of the carcinogenic potential of OTA in humans (Fink-Gremmels, 1999).

Prior to the discovery and implementation of modern milling practices, *Fusarium* species have been implicated in several human outbreaks of mycotoxicoses. Cereal grains contaminated with *F. sporitrichoides* and *F. poae* were implicated in alimentary toxic aleukia in Russia from 1932 to 1947 (Gajdusek, 1953). Symptoms included mucous membrane hyperaemia, esophageal pain, laryngitis, asphyxiation, gastroenteritis, and vertigo. Both DON and ZEN from toxic *Fusaria* have been linked to scabby grain toxicoses in the US, China, Japan, and Australia (Bilgrami and Choudhary, 1998). Symptoms included nausea, vomiting, and diarrhea. Fumonisin B_1 was associated with an illness outbreak in India with symptoms of acute onset of abdominal pain and diarrhea (Bhat and Krishnamachari, 1977). Fumonisins also have been implicated in esophageal cancer in China (Yoshizawa et al., 1994). However, with limited causal relationships and the presence of several confounding factors, data compiled by the International Agency for Research on Cancer were not conclusive for F carcinogenicity in humans (Casegnaro and Wild, 1995).

Tricothecenes have been suggested as potential biological warfare agents. For example, T-2 toxin was implicated as the chemical agent of 'yellow rain' used against the Lao Peoples Democratic Republic from 1975 through 1981 (Peraica et al., 1999). In an investigation of similar biological warfare agents in Cambodia from 1978 to 1981, T-2 toxin, DON, ZEN, nivalenol, and DAS were isolated from water and leaf samples collected from the affected areas (Watson et al., 1984; Peraica et al., 1999). Clinical symptoms preceding death included vomiting, diarrhea, hemorrhage, breathing difficulty, chest pain, blisters, headache, fatigue, and dizziness. In addition to nephritic congestion, autopsy findings included necrosis of the lining of the stomach and upper small intestine, lungs, and liver. It should be noted, however, that the origin of the samples of yellow rain is still a subject of debate. For example, one theory attributed the source of illnesses to unidentified endemic factors because the yellow rain was found to be a native bee fecal material devoid of mycotoxins (Seeley et al., 1985).

Ergot alkaloids cause gangrenous and convulsive forms of ergotism in humans (King, 1979; Taubert and Seboxa, 1990). Symptoms of the gangrenous form induced by *C. purpurea* ergot alkaloids have included edema of the legs, followed by parasthesias and gangrene of the tendons and death (King, 1979). Symptoms of the convulsive form have included gastrointestinal distress, followed by central nervous system dysfunction characterized by twitching, convulsions, and paralysis (Tulpule and Bhat, 1978; Peraica et al., 1999)

4. Negative effects of mycotoxins on ruminants

Ruminants such as cattle, sheep, goats, and deer are less known for their sensitivity to the negative effects of mycotoxins than are non-ruminants. However, production (milk, beef, or wool), reproduction, and growth can be altered when ruminants consume mycotoxin-contaminated feed for extended periods of time. Beef cattle, dairy cattle, sheep, goats, and deer are among ruminants that have been investigated.

4.1. Cattle

Aflatoxins have been shown to negatively effect production, immune system function, and rumen metabolism in cattle. Increasing AF in cattle feed to levels such as 10, 26, 56.4, 81.1, and 108.5 μg/kg has been shown to significantly reduce feed intake at each level in a dose-dependent manner (Choudhary et al., 1998). In a 155-day feeding

trial, AFB_1 (600 μg/kg) was shown to depress feed efficiency and rate of gain in steers (Helferich et al., 1986). Decreased feed efficiency in cattle has been attributed to compromised ruminal function by reducing cellulose digestion, volatile fatty acid (i.e. acetate, propionate, and butyrate) production and rumen motility (Cook et al., 1986; Helferich et al., 1986; Diekman and Green, 1992). Several mechanisms of bovine immunosuppression by AFB_1 have been illustrated in vitro. Paul et al. (1977) demonstrated that AFB_1 suppressed mitogen-induced stimulation of peripheral lymphocytes. In another study (Bodine et al., 1984), AFB_1 was shown to inhibit bovine lymphocyte blastogenesis. In a study by Cook et al. (1986), radiotelemetry was used to measure rumen motility in cattle and the results showed that AF administration (200–800 μg/kg) slowed rumen motility in a dose-dependent manner (Cook et al., 1986). Ochratoxins, on the other hand, do not cause significant toxicity to cattle when fed alone in naturally occurring doses. Barley naturally-contaminated with OTA (390–540 μg/kg) and low levels of AFB_1 (12–13 μg/kg) did not induce any significant clinical symptoms in 12-week-old calves. The absence of a toxic effect may have been due to the ruminal microbial degradation and detoxification (Patterson et al., 1981).

Aflatoxins also affect the quality of milk produced by dairy cows and result in carry-over of AFM_1 from AF-contaminated feed (Applebaum et al., 1982; Veldman et al., 1992). In the study by Applebaum et al. (1982), 10 ruminally-canulated lactating Holstein cows were given AFB_1 (13 mg per cow daily) via the rumen orifice for 7 days. Levels of AFM_1 in the milk of the treated cows ranged from 1.05 to 10.58 ng/l. The AFB_1-treated cows also had a significant reduction in milk yield. In another study (Veldman et al., 1992), the carry-over rate was shown to be higher (6.2 vs. 1.8) in early lactation (2–4 week) when compared with late lactation (34–36 week).

The T-2 toxin is also believed to induce immunosuppression in cattle (Black et al., 1992) by decreasing serum concentrations of IgM, IgG, and IgA (Mann et al., 1983), neutrophil functions and lymphocyte blastogenensis (Mann et al., 1984), and the response of lymphocytes to phytohemagglutinin (Mann et al., 1984). This toxin was also shown to induce necrosis of lymphoid tissues (Buening et al., 1982). Bovine infertility and abortion in the final trimester of gestation also have resulted from consumption of feed contaminated with T-2 toxin (Placinta et al., 1999). Calves consuming T-2 toxin at 10–50 mg/kg of feed have demonstrated ulcers in the abomesum and sloughing of the papilla in the rumen (Cheeke, 1998a). A case investigation of dairy cattle fed moldy corn containing 1 mg/kg T-2 toxin resulted in hemmorhagic syndrome (Hsu et al., 1972). With the exception of T-2 toxin, cattle have not been adversely affected by tricothecenes (Helferich et al., 1986). Neither DON nor DAS are known to affect cattle health or performance in the feedlot (Dicostanzo et al., 1996). Charmley et al. (1993) has shown that DON at levels as high as 6 mg/kg of feed had no adverse effects on milk yield and did not show evidence of carry-over into milk. Zearelenone has been suggested as a causative agent of infertility, reduced milk production, and hyperestrogenism in cattle (D'Mello and MacDonald, 1997).

Fescue foot, hyperthermia, and fat necrosis in cattle have been linked to consumption of tall fescue parasitized with *Acremonium coenophialum* (Cheeke, 1998b). Fescue foot in cattle has been shown to derive from vasoconstriction and gangrene in the hooves and tail due to the relaxation of smooth muscles caused by ergot alkaloids (Rhodes et al., 1991). Hyperthermia (summer fescue toxicosis) in cattle has been characterized by symptoms of weight loss, salivation, and heat stress (Howard et al., 1992). Fat necrosis in cattle is a condition in which areas of the fat are hardened resulting in constriction of internal organs, reduced serum cholesterol, and elevated serum amylase (Cheeke, 1998b). Cattle consuming tall fescue contaminated with endophytic fungi such as *A. lolii* also have shown symptoms of staggers, excitability, increased rectal temperature, increased respiration rate, and loss of body weight (Ross et al., 1989).

4.2. Sheep

Early studies suggested sheep as the most resistant species to mycotoxicosis. An early review by Wogan (1966) illustrated a high LD_{50} (500 mg/kg) in ovines fed AF. However, lower LD_{50} of AF (e.g. 2 mg/kg) have been established (Miller and Wilson, 1994) by injection of AF in sheep. Newberne and Butler (1969) also described sheep as the most resistant species to AF. Several studies, however, have shown notable effects of AF on sheep. Harvey et al. (1995) showed that feeding diets contaminated with AF (79% AFB_1, 16% AFG_1, 4%% AFB_2, and 1% AFG_2) to ewe lambs (2.5 mg/kg or 5.0 mg/kg of feed for 35 days) resulted in hepatotoxicity. In another study (Fernandez et al., 1997), lambs fed AF at 2.5 mg/kg of feed daily for 21 days showed symptoms of clinical aflatoxicosis including hepatic and nephritic lesions, altered mineral metabolism, and increased size and weight of the liver and kidney. Another study (Ramos et al., 1996) with the same daily dose of AF (2.5 mg/kg of feed) examined the plasma mineral concentrations on day 1, 2, 4 and 8 of the initial dose. On day 4 of intoxication, significant reductions in plasma mineral concentrations were detected for Ca (2.39 vs. 2.06 mM), P (2.95 vs. 2.50 mM), Mg (0.88 vs. 0.77 mM), K (4.40 vs. 3.81 mM), and Zn (13.2 vs. 11.6 μM). The resulting mineral deficiencies due to aflatoxicosis were attributed to lower feed intake and to the liver and kidney malfunctions as a result of AF intoxication. Exposure of lambs to AF (2.5 mg/kg of feed for 3 week) revealed changes in extrinsic coagulation factors as determined by increased fibrinogen concentration (Fernandez et al., 1995).

In a recent study (Fernandez et al., 2000), 5-month-old lambs were given feed contaminated with AF (2 mg/kg of feed) for 37 days. Average daily gain on day 35 of feeding AF was significantly reduced from 125 to 79 g and the exposed lambs showed decreased cellular immunity. After allowing for a clearance period of 30 days, the AF-exposed lambs had average daily gain and cellular immunity similar to those for the controls. Mechanisms for cellular immune response to AF in sheep have not been elucidated. Contrary to in vitro findings indicating inhibition of bovine lymphocyte blastogenesis by AF (Bodine et al., 1984), Edrington et al. (1994) could not prove that ovine mitogen-induced lymphocyte blastogenesis occurs in vivo.

Fusaria mycotoxins at high doses also appear to have some negative effects on sheep (Harvey et al., 1986, 1995). Exposing sheep to DON (15.6 mg/kg of feed) for 28 days had no effects on average daily gain, hemacytology parameters, or liver function (Harvey et al., 1986). However, weight loss (-0.6 vs. 2.4 kg/day) was reported after 34 days of feeding DAS (5 mg/kg of feed) to lambs. Further weight loss (-2.7 vs. 2.4 kg/day) also was reported at 34 days of feeding lambs same level of DAS in combination with AF (2.5 mg/kg of feed) suggesting a synergistic effect (Harvey et al., 1995).

It has been suggested that high dietary levels (12 mg/kg of feed) of ZEN for extended periods of time (10 days) may affect reproductive performance of sheep negatively by reducing fertility and ovulation rates (Dicostanzo et al., 1996). Fumonisins at high doses (11.1–45.5 mg/kg of body weight) have been demonstrated as acutely and fatally nephrotoxic and hepatotoxic in lambs (Edrington et al., 1995). It should be noted, however, that such experimental levels have not been found in F-contaminated feeds.

Sheep also have been affected by ryegrass toxicosis, which has resulted in tremors, decreased productivity, and in some cases death (D'Mello and MacDonald, 1997). Perennial ryegrass staggers have been observed in sheep consuming ryegrass contaminated with *A. lollii*. Symptoms have included shaking with loss of coordination and inability to walk (Cheeke, 1998b). Staggers have been demonstrated when *A. lolii*-contaminated ryegrass had lolitrem B toxin at levels of 2.0–2.5 mg/kg (DiMenna et al., 1992).

4.3. Other ruminants

Ruminants other than cattle and sheep have shown variable resistance to mycotoxins. Levels of F at 95 mg/kg of feed offered to weanling goats had no effects on body weight gain and did not show any noticeable signs of toxic effects (Gurung

et al., 1998). Signs of toxic effects were only detected through serum profile and sphingolipid analysis. In a study with white-tailed deer fawn fed 800 mg/kg AF over an 8-week-period (Quist et al., 1997), acute injuries in the liver were indicated by increased serum bile acid concentrations and hepatic lesions.

5. Modes of mycotoxin actions

5.1. Aflatoxins

It is well established that AFB_1 is both carcinogenic and cytotoxic. For example, synthesis of both RNA and DNA was inhibited when AF (5 mg/kg of feed) was given to rats over a 6-week-period (Butler and Neal, 1977). The activated AFB_1 metabolite (i.e. AFB_1-8,9-epoxide) forms a covalent bond with the N7 of guanine (Lillehoj, 1991) and forms AFB_1-N7-guanine adducts in the target cells (Bailey, 1994). The results are $G \rightarrow T$ transversions, DNA repair, lesions, mutations, and subsequently tumor formation (Foster et al., 1983). Hepatocellular carcinomas in humans have been linked to $G \rightarrow T$ transversion at codon 249 of the p53 tumor suppressor gene (Wang and Groopman, 1999). The reactive epoxide can also be hydrolyzed to AFB_1-8,9-dihydrodiol which ionizes to form a Schiff's base with primary amine groups in the proteins (Raney et al., 1992). The short-lived epoxide AFB_1 has also been associated with coagulopathy due to reduced synthesis of vitamin K and other clotting factors as a result of sub-lethal intoxication of animals (Bababunmi et al., 1997). With regard to the cytotoxic effects, AFB_1 has been shown to induce lipid peroxidation in rat livers leading to oxidative damage to hepatocytes (Shen et al., 1995). A more recent study (Bonsi et al., 1999), has demonstrated that AFB_1 can inhibit cyclic nucleotide phosphodiesterase activity in the brain, liver, heart, and kidney tissues.

5.2. Ochratoxins

Mechanisms of OTA toxicity have been experimentally attributed to the open lactone moiety which is structurally analogous to mitochondrial enzymes active sites and competitively binds substrate (Xiao et al., 1996). Studies on the mode of OTA action on cellular respiration indicated competitive inhibition of ATPase (Meisner and Chan, 1974), succinate dehydrogenase, and cytochrome C oxidase (Wei et al., 1985) in rat liver mitochondria. Wei et al. (1985) have attributed the cellular damage caused by OTA to hydroxyl radical formation and lipid peroxidation. In the spleen, OTA was found to disrupt protein synthesis by competitive inhibition of phenylalonyl-tRNA synthase which is a result of the phenylalanine moeity of OTA (Cheeke, 1998a). Recent in vitro studies with human (Schwerdt et al., 1999) and canine (Gekle et al., 2000) kidney cells have demonstrated the stimulatory effect of OTA (100 nmol/l) on extracellular protein kinase and caspase which results in apoptosis. In another in vitro study (Dorrenhaus et al., 2000), exposing human urothelial cells to OTA (50–500 nmol/l) demonstrated induction of unscheduled DNA synthesis with subsequent repair and lesions.

5.3. Tricothecenes

Cytotoxicity of trichothecenes has been attributed to their potent inhibition of protein, RNA, and DNA synthesis (Liao et al., 1976) and has been experimentally linked to the 12,13-epoxytricothecene nucleus (Ueno, 1977). The active sites were suggested to be the 9-ene moeity and the metabolic esters formed by the parent hydroxyls (Ueno and Yamakawa, 1970). When trichothecene binds to active polysomes and ribosomes, the peptide linkages are interrupted, the initiation and termination sequences are diminished, and the ribosomal cycle is disrupted (Ueno, 1977).

Other toxic effects of trichothecenes include disruption of membrane transport and function, suppression of the immune response, and abnormal blood function. For example, the negative effects of T-2 toxin on cell membrane function were explained by disrupting the transport of amino acids, nucleotides, and glucose and activity of Ca–K channel (Bunner and Morris, 1988). Khachatourians (1990) demonstrated that mito-

chondrial electron transport is also inhibited by T-2 toxin as a result of suppression of succinate dehydrogenase activity. Lipid peroxidation through generation of free radicals during T-2 metabolism has also been suggested as a mode of trichothecene action in rat liver (Suneja et al., 1989). With regard to the immune response, proliferation of human lymphocytes in cultures was shown to be inhibited by T-2 toxin, DON, and DAS (Johannisson et al., 1999). In another in vitro study where murine peritoneal macrophages were used (Ayral et al., 1992), DON and DAS were shown to inhibit the phagocytic activity, microbicidal activity, and superoxide anion production. The actions of trichothecenes at the hematological level have been illustrated in several studies (Faifer et al., 1992; Lautraite et al., 1997; Rio et al., 1997; Shinozuka et al., 1997). For example, T-2 toxin reduced granulocyte-macrophage colony-forming cells in the bone marrow of mice (Faifer et al., 1992). In another study (Lautraite et al., 1997), DON inhibited granulo-monocytic progenitors. Both T-2 toxin and DAS also inhibited human erythrobalstic progenitors (Rio et al., 1997). In another study (Shinozuka et al., 1997), T-2 toxin induced apoptosis in hematopoietic and lymphoid mice tissues.

5.4. Zearalenone

Zearalenone has been known for its estrogenic effects on animals. It binds to estrogen receptors influencing estrogen dependent transcription in the nucleus (Kolb, 1984). Receptor binding by ZEN has been shown to inhibit the binding estrogenic hormones in rat mammary tissues (Boyd and Wittliff, 1978). Recent studies (Ahamed et al., 2001; Withanage et al., 2001) have demonstrated the potential for ZEN to stimulate growth of human breast cancer cells containing estrogen response receptors.

5.5. Fumonisins

Fumonisins are both cytotoxic and carcinogenic to animals. The modes of such actions, however, are not completely understood. However, Wang et al. (1991) demonstrated that FB_1 disrupts sphingolipid metabolism by inhibiting sphingosine *N*-acyltransferase (ceramide synthase) in rat liver microsomes. It also has been shown that FB_1 inhibits other intracellular enzymes including protein phosphatases and arginosuccinate synthetase (Jenkins et al., 2000). Therefore, FB_1 exerts its cytotoxicity by inhibiting sphingolipid metabolism, protein metabolism, and the urea cycle. The carcinogenic role of FB_1 has been linked to the accumulation of sphingoid bases that cause unscheduled DNA synthesis (Schroeder et al., 1994), alteration of signaling by cAMP (Huang et al., 1995) and protein kinase C (Yeung et al., 1996), and disruption of normal cell cycling (Ramljak et al., 2000).

5.6. Moniliformin

The cytotoxic action of moniliformin was attributed to the inhibition of pyruvate dehydrogenase (Gathercole et al., 1986). Moniliform also has been shown to increase cardiac permeability in young rats and ducklings, suggesting a mechanism for inducing Keshan disease in humans (Zhang and Li, 1989). Using rat cardiac tissues (Chen et al., 1990), moniliformin has been shown to inhibit other enzymes including glutathione peroxidase and glutathione reductase. It was suggested, therefore, that free radical metabolism in the heart by these crucial enzymes was compromised.

5.7. Endophytic tremorgens and ergot alkaloids

Ergot alkaloids and tremorgens are known for their negative effects neuroreceptors. The primary effect of ergot alkaloids is stimulation of smooth muscle (Cheeke, 1998a). Ergot alkaloids bind alpha-adrenoreceptors and further inhibit beta-adrenoreceptors which results in vasoconstriction (Kolb, 1984). Ergot alkaloids also have been shown to inhibit prolactin secretion in humans (Silvestrini et al., 1978) and animals (Kolb, 1984). This effect was attributed to stimulation of the dopamine receptors, which regulate prolactin. Ergot alkaloids, which are similar to biogenic amines, have also been shown to act on biogenic amine receptors and, therefore, affect neurotrans-

misson (Cheeke, 1998b). The exact mechanism of lolitrem toxicity in livestock ryegrass staggers has not been identified. Possible mechanisms with sheep were suggested as inhibition of amino acid neurotransmitters (Mantle, 1983) or stimulation of choline receptors (McLeay et al., 1999).

6. Metabolism of mycotoxins in animal tissues

As with toxicity testing, most studies focusing on mycotoxin metabolism have been on AF. In general, great variations among species, and in some cases individual animals, exist with regard to AF metabolism. Factors influencing AF metabolism include species, sex, age, health, and diet. The pure form of AFB_1 is not mutagenic and its biotransformation in mammalian tissues is primarily accomplished by microsomal cytochrome P450 monooxygenases. The P450 enzymes and their subfamilies are found at different concentrations in most tissues of various animal species with abundance generally in the liver (Eaton et al., 1994). Four metabolic pathways for AFB_1 include *O*-dealkylation to AFP_1, ketoreduction to aflatoxicol, epoxidation to AF-B_1-8,9-epoxide (acutely toxic, mutagenic, and carcinogenic), and hydroxylation to AFM_1 (acutely toxic), AFP_1, AFQ_1, or AFB_{2a} (all relatively non-toxic). Similar to the pathways identified for polycyclic aromatic hydrocarbons (PAH), bioactivation of AFB_1 to AFB_1-8,9-epoxide has been linked to prostglandin-H-synthase and cytosolic lipooxygenases in lipid-hydroperoxide dependent reactions as well as P450 enzymes (Harvey et al., 1995). Various forms of P450 serve different biotransformation capacities depending on the animal species. In general, activation of AFB_1-8,9-epoxide is accomplished by enzymes in the P450 subfamilies 1A, 2B, 2C, and 3A. The isoform CYP1A2, which is PAH-inducible, is demonstrated to have the greatest binding affinity in humans (Massey et al., 1995). Detoxification of AFB_1-8,9-epoxide and AFM_1 in mammalian tissues is carried out via conjugation by glutathione (GSH), and catalyzed by GST (Massey et al., 1995; Longouet et al., 1998). Alternatively AFB_1-8,9-epoxide is hydrolyzed to a dihydrodiol (Massey et al., 1995; Longouet et al., 1998).

Activation and detoxification efficiencies in an animal species determine individual AF toxicities. Activation of AFB_1 has been shown with high capacity in mixed function oxidase systems of the nasal and tracheal mucosa of swine (Larrson and Tjalve, 1996). However, studies with hogs in AF-contaminated regions of the US resulted in a low occurrence of AF in detectable amounts in hepatic tissues (Honstead et al., 1992), which explained the occurrence of upper respiratory cancers as opposed to hepatocellular carcinomas in AF-exposed porcines. In another study (Kuilman et al., 1998), bovine hepatocytes metabolized AFB_1 to predominately AFM_1 but there were also measurable amounts of AFB_1 epoxide, AFB_1 dihydrodiol, and AFB_1-GSH conjugates. Aflatoxicol was not detected in the hepatocyte cultures although it was detected in earlier studies in cows' plasma, erythrocytes, and milk which was probably a result of the ruminal microbial degradation. Studies have shown that the GSH-GST detoxification mechanism is relatively low in humans compared with rats, mice, or rabbits (Edrington et al., 1995; Massey et al., 1995). Further studies have demonstrated variations in P450 levels, P450 isoform function, and GST activity among species (Ball and Coulombe, 1991) and within tissues of a single species (Larrson et al., 1994)

The AFM_1 was carried from AFB_1 into milk at a conversion rate ranging from 0.5 to 5% when cows consumed AFB_1-contaminated feed (Applebaum et al., 1982; Bodine and Mertens, 1983; Manorama and Singh, 1995; Skrinjar et al., 1992; Veldman et al., 1992; Chopra et al., 1999). Biotransformation of AFB_1 in the cow's liver and the corresponding AFM_1 levels in the milk depend on several factors including milk yield, microsomal mixed function oxidase activity, and presence or absence of bacterial mastitis in the udder (Chopra et al., 1999). According to two studies (Veldman et al., 1992; Chopra et al., 1999), normal carry-over was about 0.4–0.6% and daily AFB_1 intakes of ≥ 70 μg in cows resulted in greater than the regulatory limit (0.05 μg/l of AFM_1) in milk accepted in most countries.

There have been attempts to demonstrate that AF detoxification in ruminants may be enhanced by altering the diet in ruminants with different

specific protein sources (e.g. fish meal) or supplemental amino acids (e.g. methionine) to enhance metabolism. However, the adverse effects of AF on lambs were not altered when soybean meal was replaced with fish meal in the diet (Edrington et al., 1994).

Compared with AF, there are fewer studies and less published information on metabolic biotransformations of tricothecenes. In Germany, a study with swine demonstrated phase 1 hydrolysis and oxidation of tricothecenes at C-3 or C-4, followed by phase 2 glucuronidation (Baur, 1995). Earlier studies with rats and rabbits have indicated that tricothecenes (i.e. T-2 toxin and DAS) were deacylated at C-4 by a nonspecific microsomal carboxyesterase (Ohta et al., 1977). In the case of T-2 toxin, its HT-2 metabolite is cytotoxic. On the other hand, DON was assumed to undergo a direct deoxygenation at the oxide ring to form a double bond resulting in a non-toxic excretable metabolite. The mechanism of the metabolic process for DON, however, was not elucidated at that time (Bhatnagar et al., 1991). Baur (1995) illustrated that DON was slowly metabolized in swine via de-epoxidation or glucuronidation. The intestinal microbiota of rats has been credited with assisting in the metabolism and detoxification of the tricothecenes T-2 toxin, DAS, and DON (Swanson et al., 1988; Worell et al., 1989). The T-2 toxin was also hydroxylated via microsomal P450 esterase in intestinal and hepatic tissues of rats, mice, pigs, cows and chickens (Kobayashi et al., 1987).

In vivo studies with rats and in vitro studies with bovine serum have shown that OTA strongly binds to serum albumin (Chu, 1974; Marquardt and Frolich, 1992) which has resulted in slow clearance in several monogastric mammalian species. For example, the half-life of OTA in rats after injection of 0.01 mg/l was found to be 103 h (Li et al., 1997). In another study (Gatlier et al., 1981), the half-life of OTA in pigs was comparable to rats at 90 h, but in chickens the half-life of OTA was only 4 h. Microsomal hydroxylation of OTA also has been demonstrated in rats yielding the relatively non-toxic 4-hydroxy-OTA. Due to the accumulation of OTA in the blood and kidneys of farm animals, especially pigs, there has been a concern over its potential carry-over into meat products, however, significant residues have not been detected (Cheeke, 1998a).

Zearalenone has been shown to be metabolized to zearalenol and zearalanol in animal tissues (Krogh, 1991). The metabolic reduction of ZEN to the isomeric forms of α-zearalenol and β-zearalenol was demonstrated as a function 3α-hydroxysteroid dehydrogenase in the cytosol or organelles of hepatocytes. Although β-zearalenol has been shown to have three times the relative estrogenic effects as ZEN, α-zearalenol has had less of an estrogenic effect (Cheeke, 1998a). Both ZEN and its metabolites are conjugated by glucuronide and sulfate. The toxicity in animal species has been dependent upon the 3α-hydroxysteroid dehydrogenase activity, the isomeric form of metabolite, and the ability to excrete as opposed to recycling the glucuronide conjugate (Patterson, 1977).

Although the ester moeity of FB_1 was shown to be hydrolyzed in the intestine of vervet monkeys, no appreciable metabolism was detected in tissues such as the liver, kidney, or lung (Shephard et al., 1994). Ergot alkaloids can be hydroxylated or dealkylated by P450 enzymes and, therefore, their water solubility, potential conjugation, and excretion are increased (Cheeke, 1998b). New evidence with cattle indicated extensive hydroxylation of ergotamine in the liver (Moubark and Rosenkrans, 2000).

7. Degradation of mycotoxins by the rumen microorganisms

Evidence of ruminal microbial degradation of mycotoxins has been demonstrated in isolated cultures of rumen contents (Hult et al., 1976; Ribelin et al., 1978; Bodine and Mertens, 1983; Kiessling et al., 1984). This interest was derived from early studies revealing the greater resistance of ruminants to the negative effects of several mycotoxins (Wogan, 1966). It should be noted, however, that the rumen function has been shown to be negatively affected by the presence of mycotoxins, suggesting the significant role of the rumen in handling a mycotoxin exposure, biotransforma-

tion of the toxins, and excretion of the metabolites (Cook et al., 1986). Mertens (1979) determined in vitro that the rumen microbiota may be sensitive to AF and that its function can be compromised. The negative effects included decreased cellulose degradation, volatile fatty acid production, ammonia production, and proteolysis. In studies with steers (Helferich et al., 1986) and young lambs (Edrington et al., 1994), ruminal concentrations of volatile fatty acids were not altered by feeding AF.

In an early study with cattle (Hult et al., 1976), rumen metabolism of OTA demonstrated a cleavage of OTA by ruminal protozoa and bacteria. Ribelin et al. (1978) also demonstrated that OTA cultured with rumen microorganisms was degraded to 4-hydroxy-OTA (Ribelin et al., 1978). The rumen protozoa hydrolyzed the peptide bond of OTA in a reaction catalyzed by carboxypeptidase A and chymostrysinogen, rendering less toxic metabolites. The role of ruminal fermentation was apparent when comparing the LOAEL of 13 mg/kg given orally with that of 1 mg/kg given intravenously to cattle (Cheeke, 1998a).

The metabolism of AF, OT, ZEN, T-2 toxin, DON, and DAS was demonstrated (Kiessling et al., 1984) by using cultures of rumen fluid, rumen protozoa, or rumen bacteria. It was demonstrated that 90–100% of the metabolism of OT, ZEN, T-2 toxin, and DAS were achieved by the rumen protozoa and, therefore, they were considered as the most important ruminal microbial population in mycotoxin biodegradation. Considerably lower metabolism of the same mycotoxins was noted in cultures containing only rumen bacteria. Both AF and DON were not metabolized by the rumen microorganisms (Kiessling et al., 1984). In a subsequent study DON was cultured with rumen fluid and was biotransformed to deepoxynivalenol (Swanson et al., 1987). In this study, T-2 toxin was shown to be completely biodegraded to various metabolites when cultured with rumen contents. Contrary to monogastrics, rumen microbiota did not hydrolyze FB_1 in vitro (Caloni et al., 2000).

Although the above mentioned studies have provided insight into the mechanisms of ruminal microbial degradation of mycotoxins, the complete pathways, rates, and extents of such degradation have not been elucidated. In vitro studies have shown that specific strains of *Lactobacillus* bacteria can bind to AFB_1 (El-Nezami et al., 1998). It has not been determined whether factors such as bacterial binding or the presence of other micoorganisms in the motile rumen environment are conducive to AF biodegradation. This evidence indicated that the motile rumen environment could expedite or delay the biotransformation of other mycotoxins such as tricothecenes (Cook et al., 1986).

8. Factors affecting production, contamination of foods and feeds, and toxicity of mycotoxins

A main difficulty in assessing the risk of mycotoxins to human and animal health is the multiplicity of factors affecting the production or presence of mycotoxins in foods or feeds**.** Mere isolation and confirmation of mycotoxigenic fungal species in foods or feeds does not indicate the presence of mycotoxins. Upon development of accurate and sensitive techniques for qualitative and quantitative analysis of mycotoxins, researchers have found that various factors operate interdependently to affect fungal colonization and/or production of the mycotoxins. D'Mello and MacDonald (1997) categorized the factors as physical, chemical, and biological. Physical factors include the environmental conditions conducive to fungal colonization and mycotoxin production such as temperature, relative humidity, and insect infestation. Chemical factors include the use of fungicides and/or fertilizers. Stresses such as drought, an increase in temperature, and an increase in relative humidity may selectively alter colonization and metabolism of mycotogenic fungi and thus alter mycotoxin production (Russell et al., 1991). These researchers also indicated that unseasonable conditions may render crops and forages susceptible to mycotoxin production. Cool and damp springtime weather favor the germination of the sclerotia and thus ergot alkaloid formation in fescue and ryegrass (Cheeke, 1998a).

The biological factors are based on the interactions between the colonizing toxigenic fungal species and substrate. While some plant species are more susceptible to colonization, environmental conditions may increase the vulnerability of other more resistant plant species. The biological factors have been further sub-categorized (Moss, 1991) into intrinsic factors including fungal species, strain specificity, strain variation, and instability of toxigenic properties. Such intrinsic factors underscore the difficulty of risk assessment of mycotoxin exposure based on mold contamination. Species and strain specificity are well described by the numerous mycotoxins produced by two or more fungi. A strain variation refers to a specific culture identity for the same species fungal isolate and how these strains produce mycotoxins in a variable fashion. Finally, the toxigenic properties may vary over time and as the mycoecology changes toxins may be reduced. Several studies have shown that optimal conditions for fungal growth are not necessarily optimum for toxin production. For example, different strains of *A. flavus* have been shown to produce AF at different rates when cultured under similar conditions (Hesseltine et al., 1970).

A. flavus have shown optimum temperatures for colonization to range from 25 to 35 °C with the substrate water activity (A_w) being 0.90 (Smith and Moss, 1985; Smith and Ross, 1991). In a more recent study (Gqaleni et al., 1997), however, AF production by *A. flavus* was optimal at 30 °C and substrate A_w of 0.996. In the same study, AF production was affected by substrate source, incubation time, and the presence of other mycotoxins produced by *A. flavus* (Gqaleni et al., 1997). Strain variations in toxin production also have been reported for *F. poae* and *F. sporitrichoides* (Joffe and Yagen, 1977). The ideal growth conditions for *Fusarium* species including *F. poae* and *F. sporitrichoides* are 21 °C or less and A_w of 0.980–0.995 (Moss, 1991; Dicostanzo et al., 1996). The optimum temperatures for toxin (i.e. T-2 toxin, ZEN, and DON) production by these species are 6–12, 19–20, and 28 °C, respectively (Park et al., 1996). The differences between optimal growth conditions (temperatures ranging from 0 to 31 °C and A_w 0.95) and toxin production (temperatures ranging from 12 to 24 °C) also have been documented for other species such as *P. viridicatum* (Moss, 1991).

In a 3-year study (Wood, 1992) on occurrence of AF in selected US foods (milled corn products, peanuts, and peanut products) and feeds (shelled corn, cottonseed, and cottonseed meal), the unpredictable nature of AF contamination and the difficulty in assessing the extent of such contamination have been documented. Very high levels (greater than the 20 μg/kg FDA regulatory level) of contamination were found in corn harvested from all parts of the US in 1989, 1990, and 1991. In these years, 9.1, 20.7, and 37.1%, respectively, of the corn samples examined before human consumption contained AF at levels >20 μg/kg in the southern states (i.e. Arkansas, Texas, and Oklahoma). The increase in AF contamination in 1990 was attributed to the drought condition. However, the dramatic increase in AF contamination in 1991 could not be explained by environmental factors. In an earlier study by Wood (1989), it was found that cottonseed and cottonseed meal were the primary targets of AF contamination in the southwestern states of Arizona and California. In comparison to the rest of the nation (0 and 28.0 μg/kg for cottonseed and cottonseed meal, respectively), the average values for both states were 37.7 and 43.0 μg/kg of seed and meal, respectively.

In other parts of the world, OT appear to be a more significant problem in animal feeds than AF. For example, a 4-year (1975–1979) study in Poland (Juszkiewicz and Jadwiga, 1992) showed OT contamination levels ranging from 2 to 200 μg/kg in 14, 12, and 18% of the barley, wheat, and rye samples tested, respectively. In this study, only 4% of the corn samples tested positive for AF. In general, the mycotoxin contamination rate across all cereal grains tested (including oats and corn) was 9%. Surveillance of mixed feeds collected from different Spanish farms (19 cattle, 20 pig, 15 rabbit, and 15 poultry farms) revealed a high level of fungal contamination (Abarca et al., 1994). Out of the 69 samples examined, 61 were positive for *Penicillium*, 56 were positive for *Aspergillus*, and 33 were positive for *Fusarium*. Upon culturing, only five out of 37 *Aspergillus* strains (13.5%)

were AF producers, indicating the unpredictability of AF contamination.

In a more practical sense, mycotoxin contamination of foods or feeds may result from inadequate storage and/or handling of harvested products. Prevention and control methods have been prescribed for mitigating mycotoxin contamination of feeds (Harris, 1997). These methods require that feed handlers and grain mill operators keep grain bins clean and store grain at less than 14% moisture. Feed ingredients must be dry, oxygen free, fermented or treated with mold-growth inhibitors. With regard to silage crops, harvesting at the appropriate moisture content and both packing and sealing the silo (to exclude oxygen and allow for desirable anaerobic fermentation) are essential for reducing mycotoxin contamination potential. Chemical treatment and processing are anthropogenic factors that may decrease mycotoxin contamination of foods or feeds. Wet and dry milling processes as well as heat in the cooking process have been shown to reduce AF in foods (Scott, 1984). Heating and roasting have been shown to significantly decrease AF content in corn (Conway et al., 1978; Hale and Wilson, 1979). A review of several studies, however, suggested that processing and pasteurization of milk do not completely destroy mycotoxins (Manorama and Singh, 1995).

Bentonite and aluminosilicate clays used as binding agents have been shown to reduce AF intoxication in pigs (Smith, 1980, 1984; Lindemann et al., 1993; Schell et al., 1993), cattle (Diaz et al., 1997), rats (Smith, 1980; Galey et al., 1987), and poultry (Scheideler, 1993) without causing digestive problems when mixed with AF-contaminated feeds. However, these clays are ineffective against ZEN and F, can alter the nutritional value (by binding trace minerals and vitamins and reducing their bioavailability), and produce dioxins (Devegowda and Castaldo, 2000). Esterified glucomannan (i.e. an organic compound naturally-occurring in yeasts) is a recently discovered alternative with high binding potential to mycotoxins. For example, glucomannan supplementation (at 0.05% of the diet) of dairy cows consuming AF-contaminated feeds reduced AF in milk by 58% (Devegowda and Castaldo, 2000) while similar reductions in AF in milk were achieved at a much higher level (1.1% of the diet) of sodium bentonite supplementation. These data suggested the potential advantage of esterified glucomannan over clays in reducing AF toxicity. Devegowda and Castaldo (2000) have illustrated that esterified glucomannan was successful in binding F and ZEN at high efficiency (67 and 77% of that for AF). Ammoniation of stored crops has been shown to substantially ($>99\%$) reduce AF levels by hydrolyzing the lactone ring (Phillips et al., 1994). It has also been proven effective in reducing the toxicity of OTA (Marquardt and Frolich, 1992).

Recently, the potential role of dietary factors to counteract the toxic effects of mycotoxins have been reviewed (Galvano et al., 2001). The role of antioxidants (Se and Vitamins A, C, and E) and food additives was evaluated. Antioxidant defense mechanisms observed have included free radical scavenging, reduced lipid peroxidation, and general inhibition of the mutagenic process. Galvano et al. (2001) also reviewed the role of food components (fructose, phenolic compounds, coumarins, and chlorophyll) and food additives (piperine, aspartame, cyproheptadine, and allyl sulfides) in reducing the toxicity of various mycotoxins by decreasing toxin formation and enhancing metabolism. For example, phenolic compounds have been shown to metabolically enhance AFB_1 conjugation and elimination (Rompelberg et al., 1996).

The antioxidant ethoxyquin has been recognized as a strong anti-aflatoxigenic agent. Kensler et al. (1986) demonstrated the role of ethoxyquin in rat hepatocytes as induction of conjugating GST. Mendel et al. (1987) confirmed the enhancing effect of ethoxyquin on phase II metabolism in several subcellular components (microsomes, cytosol, and cell membrane) of the rat liver. In another study, gamma glutamyltranspeptidase was induced along with GST (Manson et al., 1997). A more recent study with marmosets has established ethoxyquin as a potential chemoprotective agent against the carcinogenic effects of AFB_1 in humans (Bammler et al., 2000).

Another synthetic dietary chemoprotective agent investigated for its anti-carcinogenic activity

against AFB_1 is 5-(2-pyrizinyl)-4-methyl-1,2-dithiol-3-thione (oltipraz). Oltipraz was found to decrease the binding of AFB_1 to DNA and to increase epoxide hydrase and glucuronide and glutathione levels in rat liver and kidney (Kensler et al., 1985). Oltipraz also has been shown to decrease P450 activation of AFB_1 and to enhance GST activity in human hepatocytes in vitro (Langouet et al., 1995).

9. Economic impact of mycotoxins

There are multiple criteria for assessing the economic impact of mycotoxins on humans and on animal agriculture. Considerations include loss of human and animal life, health care and veterinary care costs, loss of livestock production, loss of forage crops and feeds, regulatory costs, and research cost focusing on relieving the impact and severity of the mycotoxin problem. Formulas for worldwide economic impact have been difficult to develop and, therefore, most reports on economic impact are on a single aspect of mycotoxin exposure or contamination.

The worldwide contamination of foods and feeds with mycotoxins is a significant problem. Studies have shown extensive mycotoxin contamination in both developing and developed countries. In a recent review (Fink-Gremmels, 1999), it was estimated that 25% of the world's crops may be contaminated with mycotoxins. Surveillance studies (Placinta et al., 1999) showed that worldwide contamination of cereal grains and other feeds with *Fusarium* mycotoxins is a global concern. In Yugoslavia, studies on mycotoxigenic fungi in raw milk have indicated that 91% of the samples tested were contaminated (Skrinjar et al., 1995). In the US, a study was conducted in seven Midwestern states in 1988–1989 and found mycotoxins in 19.5% of corn samples assayed prior to any induced environmental stress and 24.7% of the samples following stress induction (Russell et al., 1991). Shane (1994) estimated the 1980 losses due to AF in corn of eight Southeastern states at 97 million dollars with additional 100 million dollars in production losses at hog farms feeding the contaminated corn.

India is a prime example of a country in which the economy is affected heavily by mycotoxins. In a study in the Bihar region from 1985 to 1987 (Ranjan and Sinha, 1991), nearly 51% of the 387 samples tested were contaminated with molds. Of the 139 samples containing AF, 133 had levels above 20 μg/kg. In another study (Phillips et al., 1996), levels as high as 3700 μg/kg of AF were reported in groundnut meal used for dairy cattle. Researchers also found 21 of 28 dairy feed samples from farms in and around Luhiana and Punjab to be contaminated with AFB_1 at levels ranging from 50 to 400 μg/kg (Dhand et al., 1998). It was estimated that 10 million dollars were lost in India's export within a decade due to groundnut contamination with mycotoxins (Vasanthi and Bhat, 1998).

10. Regulation of mycotoxins in foods and feeds

There have been a range of legislative controls on mycotoxins in foods and feeds worldwide. In January 1999, the European Union set the levels of maximum aflatoxins in agricultural commodities at 4 ppb with AFB_1 at 2 ppb (de Koe, 1999; Devegowda and Castaldo, 2000). In the US, the Federal Food Drug and Cosmetic Act Sec. 402 (a) (1) has regulated AF as adulterants in foods and feeds. Action levels were set at 20 ppb in foods and feeds and 0.5 ppb in milk (Price et al., 1993). The Center for Veterinary Medicine of the FDA has been responsible for examining feeds for mycotoxins. Due to unpredictable high levels in feeds, the FDA has historically relaxed these standards (Price et al., 1993). In 1990, the limit of detection for AF in grain and nuts was 1 μg/kg and in milk was 0.05 ng/l (Wood, 1992). As sensitivity for detection of mycotoxins continue to improve, traditionally acceptable levels of AF in foods and feeds have dropped. As of 1994, 77 countries had regulations for AF in foods and feeds, with tolerance levels in foods as high as 50 μg/kg and in feeds as high as 1000 mg/kg (Van Egmond and Dekker, 1995). In Denmark, pig carcasses are confiscated for levels of OTA >25 μg/kg in the liver or kidney (Galvano et al., 2001).

Worldwide, *Fusarium* mycotoxins have been less stringently regulated than AF (D'Mello and MacDonald, 1997). In the US, there has been no legislative control on *Fusarium* mycotoxins with the exception of advisory levels of DON in grains and oilseed by-products (e.g. soybean meal, cottonseed meal, and canola meal) used for feeding cattle or poultry. Legislations for tricothecenes and fumonisins have been long overdue (Placinta et al., 1999). In the US, F are allowed in petfood at levels up to 10 mg/kg, as long as the contaminated grain compromises less than half of the total product (Bird, 2000). Since domesticated animals have a proven sensitivity to DON, it has been suggested that the acceptable levels should not exceed 0.5 mg/kg in petfood or animal feed (Bird, 2000). The Center for Veterinary Medicine current guidelines for DON were set at no more than 4 ppm for wheat added to feed given to swine (Price et al., 1993). As of the date of this publication, there are still several countries that do not have any regulatory controls over mycotoxins and limited technology to mitigate them.

Efforts have continued internationally to establish guidelines to control mycotoxins. The FAO has worked with developing countries to mitigate mycotoxin contamination in foods and feeds. Activities of FAO programs on mycotoxin mitigation have included advisory assistance, technical assistance, and the implementation of tolerance levels for mycotoxins. In an advisory role, the FAO has worked closely with the WHO Joint Expert Committee on Food Additives and Contaminants (JECFA) to ensure proper sampling and analyses of AF. Technical assistance has focused on mycotoxin prevention and control and has involved the introduction of post-harvest technologies to developing countries (Boutrif, 1995).

11. Summary

Although there have been over 300 isolated mycotoxins (Betina, 1984), research has focused on toxins that have caused significant damage to humans, livestock, or companion animals, including AFB_1, OTA, tricothecenes (T-2 toxin, DON, and DAS), ZEN, F, tremorgenic toxins, and ergot alkaloids. All of these groups have been described as structurally unique secondary fungal metabolites with unique biochemical modes of action. For example, AFB_1 (the most widely studied mycotoxin) is bioactivated from its original form to a mutagenic and carcinogenic metabolite (Smith and Ross, 1991). Tricothecenes have been known to cause immunosuppressive effects such as compromised phagocytic action, reduced IgG and IgM levels as well as reduced antibody response. Both T-2 toxin and DON have been shown to inhibit protein and DNA synthesis. Zearalenone has been known for its endocrine disruption by binding to estrogen receptors. Fumonisins have been shown to inhibit sphinganine-*N*-acetyl transferase and thus ceramide synthesis, increasing the intracellular substrate concentration of sphinganine (Fink-Gremmels, 1999).

Early studies (Wogan, 1966) have demonstrated the species specific acute and chronic toxic effects of mycotoxins. Turkey X disease, which prompted the isolation and identification of AF, was the first case study of acute aflatoxicosis (Asao et al., 1963). Porcines have since been shown to be among the most vulnerable species to mycotoxins, and have been especially prone to the estrogenic effects of ZEN (Diekman and Green, 1992.) The greatest mycotoxin risks to equine species identified thus far have been the toxins associated with the genus *Fusarium* (e.g. *F. proliferatum*). Fumonisins produced by this species have been demonstrated as the cause of both leukoencephalomalacia and acute neurotoxicity in equine (Asquith, 1991; Placinta et al., 1999). Fumonisins are classified by the WHO-IARC (1993a,b) as possible human carcinogens (Class 2B). Aflatoxin B_1 has been listed by IARC as a known carcinogen in humans and studies have shown human hepatocellular carcinomas may be compounded by hepatitis B virus and AFB_1 in combination (Kensler et al., 1991). Rat studies have provided most of the information on specific modes of AF action with regard to human carcinogenicity. Companion animals such as dogs and cats have also been adversely affected by mycotoxins.

The observed negative effects of mycotoxins in ruminants have generally been milder, but mycotoxicoses have still been demonstrated clinically, toxicologically, and pathologically. For example, AF have been shown to decrease feed efficiency and thus productivity in cattle (Choudhary et al., 1998). Both AF and ZEN have been shown to decrease productivity in sheep (Dicostanzo et al., 1996; Fernandez et al., 2000).

The key to developing strategies to control mycotoxicoses in individual species has been understanding the metabolic pathways and modes of mycotoxin action. For example, it has been well documented that AF metabolism occurs via microsomal cytochrome P450 monooxygenases found in all tissues but with greater activity and concentrations in the liver. Variable toxicities have been demonstrated based on the microsomal enzyme capacities in different animals (Edrington et al., 1995; Massey et al., 1995). For ZEN, the toxicity in animals has been dependent upon the 3α-hydroxysteroid dehydrogenase activity, the isomeric form of the metabolite, and the ability to excrete as opposed to recycling the glucuronide conjugate (Patterson, 1977; Cheeke, 1998a).

Ruminal degradation of mycotoxins has been recognized for a long time owing to the relative resistance to mycotoxins in ruminant species. Culture isolates from rumen fluids have been shown to metabolize OT, ZEN, T-2 toxin, and DAS (Ribelin et al., 1978; Kiessling et al., 1984). Ruminal protozoa have been demonstrated to play a very important role in the detoxification process in adult cattle (Ribelin et al., 1978; Kiessling et al., 1984).

Despite the ever-increasing understanding of mycotoxins, they still have a continuous and severe economic impact worldwide. Mycotoxins have resulted in losses in human and animal life, in livestock production, in forage crops and other feeds, in health care and veterinary care costs, in regulatory costs, and in research costs. Worldwide regulation on mycotoxins has varied by region. As of 1994, 77 countries had developed regulations for AF in animal feeds and human food, with regulatory levels in foods as high as 50 μg/kg and in feeds as high as 1 mg/kg. Most countries have not regulated other mycotoxins (e.g. OTA, T-2 toxin, DON, DAS, or ZEN) in foods or feeds (Van Egmond and Dekker, 1995).

12. Conclusions

In the introduction to one of the most comprehensive texts on mycotoxins, Lillehoj (1991) emphasized the evolutionary aspects of AF as a model for mycotoxin biosynthesis. In order to survive, fungi such as *A. flavus* had to evolve to withstand environmental challenges and find an ecological niche. Therefore, the production of secondary toxic metabolites has been a survival mechanism for mycotoxigenic fungi. While some fungi and mycotoxins have destroyed the host, others have thrived in mutual harmony with the host, providing protection from herbivory.

Adaptation in higher trophic levels, such as mammalian species, has been crucial to survival as well. In view of the wide range and varying degrees of mycotoxicoses in animals, there have been signs of co-evolutionary aspects of mammalian susceptibilities. Monogastric animals from fowl to domestic companion animals to humans have been susceptible to a number of mycotoxins. Besides the loss in productivity and toxicity tests with very high doses, studies demonstrated a relative resistance of ruminants to the adverse effects of a number of mycotoxins, opening the door for research into adaptive mechanisms in these species. The key to determining how the ruminant species adapted to mycotoxins has been through studies on the metabolic pathways in the rumen environment.

Besides the demonstrated effects of mycotoxins on humans or animals some important aspects of toxicology and control have still resided in the realm of the unknown and unexplored. For example, there has been a general paucity of data on mycotoxins classified as carcinogens in humans by IARC, and currently there is a genuine concern over the carcinogenic potential of OTA and F, for which few regulations exist worldwide. Only with continued research on understanding the effects and modes of mycotoxin action in various species, have regulations and control strategies been forthcoming.

References

Abarca, M.L., Bragulat, M.R., Castella, G., Cabanes, F.J., 1994. Mycoflora and aflatoxin producing strains in animal mixed feeds. J. Food Prot. 57, 256–258.

Ahamed, S., Foster, J.S., Bukovsky, A., Wimalasena, J., 2001. Signal transduction through the ras/Erk pathway is essential for the mycoestrogen zearalenone-induced cell-cycle progression in MCF-7 cells. Mol. Carcinog. 30, 88–98.

Applebaum, R.S., Brackett, R.E., Wiseman, D.W., Marth, E.H., 1982. Response of dairy cows to dietary aflatoxin: feed intake and yield, toxin content, and quality of milk of cows treated with pure and impure aflatoxin. J. Dairy Sci. 65, 1503–1508.

Asao, T., Buchi, G., Abdel-Kader, M.M., Chang, S.B., Wick, E.L., Wogan, G.N., 1963. Aflatoxins B and G. J. Am. Chem. Soc. 85, 1706–1707.

Asquith, R.L., 1991. Mycotoxicoses in horses. In: Smith, J.E., Anderson, R.A. (Eds.), Mycotoxins and Animal Foods. CRC Press, Boca Raton, FL, pp. 679–688.

Asquith, R.L., Edds, G.T., 1981. Investigations in equine aflatoxicosis. Proc. Am. Assoc. Equine Pract. 26, 193–193.

Ayral, A.M., Dubech, N., Le Bars, J., Escoula, L., 1992. In vitro effect of diacetoxyscirpenol and deoxynivalenol on microbicidal activity of murine peritoneal macrophages. Mycopathologia 120, 121–127.

Bababunmi, E.A., Thabrew, I., Bassir, O., 1997. Aflatoxin induced coagulopathy in different nutritionally classified animal species. World Rev. Nutr. Diet 34, 161–181.

Bailey, G.S., 1994. Role of aflatoxin-DNA adducts in the cancer process. In: Eaton, D.L., Groopman, J.D. (Eds.), The Toxicology of Aflatoxins: Human Health, Veterinary, and Agricultural Significance. Academic Press, San Diego, CA, pp. 137–148.

Ball, R.W., Coulombe, R.A., 1991. Comparitive biotransformation of aflatoxin B1 in mammalian airway epithelium. Carcinogenesis 12, 305–310.

Bamburg, J.R., 1972. The biological activities and detection of naturally occurring 12,13-epoxy-delta-tricothecenes, Clin. Toxicol. 495–515.

Bammler, T.K., Slone, D.H., Eaton, D.L., 2000. Effects of dietary olipratz and ethoxyquin on aflatoxin B1 biotransformation in non-human primates. Toxicol. Sci. 54, 30–41.

Baur, J., 1995. The metabolism of trichothecenes in swine. Dtsch. Tierarztl. Wochesnchr. 102, 50–52.

Becroft, D.M.O., Webster, D.R., 1972. Aflatoxins and Reye's disease. Br. Med. J. 4, 117.

Bennett, R.A., Essigmann, J.M., Wogan, G.N., 1981. Excretion of an aflatoxin-guanine adduct in the urine of afaltoxin B1-treated rats. Cancer Res. 41, 650–654.

Beri, H.K., Vadehra, D.V., Gupta, J.K., 1991. Proportionate incidence of mycotoxigenic fungi—Fusarium and its effect on ingestion by poultry. J. Food Sci. Technol. 28, 329–331.

Betina, V., 1984. Biological effects of mycotoxins. In: Betina, V. (Ed.), Mycotoxins-Production, Isolation, Separation and Purification. Elsevier, Amsterdam, The Netherlands, pp. 25–36.

Bhat, R.V., Krishnamachari, K.A.V.R., 1977. Follow-up study of aflatoxic hepatitis in parts of Western India. Ind. J. Med. Res. 66, 55–58.

Bhatnagar, D., Lillehoj, E.B., Bennett, J.W., 1991. Biological detoxification of mycotoxins. In: Smith, J.E., Anderson, R.A. (Eds.), Mycotoxins and Animal Foods. CRC Press, Boca Raton, FL, pp. 815–822.

Biehl, M.L., Prelusky, D.B., Koritz, G.D., Hartin, K.E., Buck, W.B., Trenholm, H.L., 1993. Biliary excretion and enterohepatic cycling of zearalenone in immature pigs. Toxicol. Appl. Pharmacol. 121, 152–159.

Bilgrami, K.S., Choudhary, A.K., 1998. Mycotoxins in preharvest contamination of agricultural crops. In: Sinha, K.K., Bhatnagar, D. (Eds.), Mycotoxins, Agriculture, and Food Safety. Marcel Dekker Publishers, New York.

Bird, C., 2000. Detecting and Controlling Mycotoxins in Petfoods, Technical Symposium on Mycotoxins. Alltech, Inc, Nicholasville, KY.

Bitay, F.H., Glavitis, R., Sellyey, G., 1979. Mycotoxins. Magyar Allatorvosak Lapja 34, 417–422.

Black, R.D., McVet, D.S., Oehme, F.W., 1992. Immunotoxicity in the bovine animal. Vet. Hum. Toxicol. 34, 438–442.

Blunden, G., Roch, O.G., Rogers, D.J., Coker, R.D., Bradburn, N., 1991. Mycotoxins in food. Med. Lab. Sci. 48, 271–282.

Bodine, A.B., Mertens, D.R., 1983. Toxicology, metabolism, and physiological effect of aflatoxin in the bovine. In: Diener, U.L., Asquith, R.L., Dickens, J.W. (Eds.), Aflatoxin and *Aspergillus flavus* in Corn. So. Cooperative Ser. Bull. 279. Craftmaster Printers Inc, Opelika, AL, pp. 46–50.

Bodine, A.B., Fisher, S.F., Gangjee, S., 1984. Effect of aflatoxin B1 and major metabolites on phytohemeagglutinin-stimulated lymphoblastogenesis of bovine lymphocytes. J. Dairy Sci. 67, 110–114.

Bonsi, P., Agusti-Tocco, G., Palmery, M., Giorgi, M., 1999. Aflatoxin B_1 is an inhibitor of cyclic nucleotide phosphodiesterase activity. Gen. Pharmacol. 32, 615–619.

Boorman, G.A., McDonald, M.R., Imoto, S., Persing, R., 1992. Renal lesions induce by ochratoxin A exposure in the F344 rat. Toxicol. Pathol. 20, 236–245.

Bosch, U., Mirocha, C.J., Abbas, H.K., di Menna, M., 1989. Toxicity and toxin production by Fusarium isolates from New Zealand. Mycopathologia 108, 73–79.

Borison, H.L., Goodheart, M.L., Thut, D.C., 1991. Hypovalemic shock in acute lethal T-2 mycotoxicosis. Toxicol. Appl. Pharmacol. 108, 107–113.

Boutrif, E., 1995. FAO programmes for prevention, regulation, and control of mycotoxins in foods. Natural Toxins 3, 322–326.

Boyd, P.A., Wittliff, J.L., 1978. Mechanism of Fusarium mycotoxin action in mammary gland. J. Toxicol. Environ. Health 4, 1–8.

Brake, J., Hamilton, P.B., Kittrell, R.S., 1999. Effects of the trichothecene mycotoxin diacetoxyscirpenol on fertility and hatchability of broiler breeders. Poult. Sci. 78, 1690–1694.

Brake, J., Hamilton, P.B., Kittrell, R.S., 2000. Effects of the trichothecene mycotoxin diacetoxyscirpenol on feed consumption, body weight, and oral lesions of broiler breeders. Poult. Sci. 79, 856–863.

Buening, G.M., Mann, D.D., Hook, B., Osweiller, G.D., 1982. The effect of T-2 toxin on bovine immune system: cellular factors. Vet. Immunol. Immunopathol. 3, 411–418.

Bunner, D.L., Morris, E.R., 1988. Alteration of multiple cell membrane functions in L-6 myoblasts by T-2 toxin: an important mechanism of action. Toxicol. Appl. Pharmacol. 92, 113–121.

Butler, W.H., Neal, G.E., 1977. Mode of action and human health aspects of aflatoxin carcinogenesis. Pure Appl. Chem. 49, 1747–1751.

Caloni, F., Spotti, M., Auerbach, H., Op den Camp, H., Grimmels, J.F., Pompa, G., 2000. In vitro metabolism of fumonisin B1 by ruminal microflora. Vet. Res. Commun. 24, 379–387.

Cantley, D.C., Redmer, D.A., Osweiler, G.D., Day, B.N., 1982. Effect of zearalenone mycotoxin on luteal function in gilts. J. Anim. Sci. 55, 104S.

Carlton, W.W., Tuite, J., 1977. Metabolites of *P. viridicatum* toxicology. In: Rodricks, J.V., Hesseltine, C.W., Mehlman, M.A. (Eds.), Mycotoxins in Human and Animal Health. Pathotox, Park Forest South, IL, pp. 525–541.

Carnaghan, R.B.A., Hartley, R.D., O'Kelly, J., 1963. Toxicity and fluorescence properties of the aflatoxins. Nature (London) 200, 1101–1102.

Casegnaro, M., Wild, C., 1995. IARC activities in mycotoxin research. Natural Toxins 3, 327–331.

Chang, K., Kurtz, H.J., Mirocha, C.J., 1979. Effects of the mycotoxin zearalenone on swine reproduction. Am. J. Vet. Res. 40, 1260–1267.

Charmley, E., Trenholm, H.L., Thompson, B.K., Vudathala, D., Nicholson, J.W.G., Prelusky, D.B., Charmley, L.L., 1993. Influence of levels of deoxynivalenol in the diet of dairy cows on feed intake, milk production, and its composition. J. Dairy Sci. 76, 3580–3587.

Cheeke, P.R., 1998a. Mycotoxins in cereal grains and supplements. In: Cheeke, P.R. (Ed.), Natural Toxicants in Feeds, Forages, and Poisonous Plants. Interstate Publishers, Inc, Danville, IL, pp. 87–136.

Cheeke, P.R., 1998b. Mycotoxins associated with forages. In: Cheeke, P.R. (Ed.), Natural Toxicants in Feeds, Forages, and Poisonous Plants. Interstate Publishers, Inc, Danville, IL, pp. 243–274.

Chen, L.Y., Tian, X.L., Yang, B., 1990. A study on the inhibition of rat myocardium glutathione peroxidase and glutathione reductase by moniliformin. Mycopathologia 110, 119–124.

Chopra, R.C., Chabra, A., Prasad, K.S.N., Dudhe, A., Murthy, T.N., Prasad, T., 1999. Carryover of Aflatoxin M1 in milk of cows fed aflatoxin B1 contaminated ration. Ind. J. Anim. Nutr. 16, 103–106.

Choudhary, P.L., Sharma, R.S., Borkhataria, V.N., Desai, M.C., 1998. Effect of feeding aflatoxin B1 on feed consumption through naturally contaminated feeds. Ind. J. Anim. Sci. 68, 400–401.

Chu, F.S., 1974. A comparative study of the interaction of ochratoxins with bovine serum albumin. Biochem. Pharmacol. 23, 1105–1113.

Cole, R.J., Cox, R.H., 1981. Handbook of Toxic and Fungal Metabolites. Academic Press, New York.

Constable, P.D., Smith, G.W., Rottinghaus, E., Haschek, W.M., 2000. Ingestion of fumonisin B1-containing material decreases cardiac contractility and mechanical efficiency in swine. Toxicol. Appl. Pharmacol. 162, 151–160.

Conway, H.F., Anderson, R.A., Bagley, E.B., 1978. Detoxification of aflatoxin contaminated corn by roasting. Cereal Chem. 55, 115–118.

Cook, W.O., Richard, J.L., Osweiller, G.D., Trampel, D.W., 1986. Clinical and pathologic changes in acute bovine aflatoxicosis: rumen motility and tissue and fluid concentrations of aflatoxins B1 and M1. Am. J. Vet. Res. 47, 1817–1825.

Croy, R.G., Essigmann, J.M., Reinhold, V.N., Wogan, G.N., 1978. Identification of the principal afaltoxin B1-DNA adduct formed in vivo in rat liver. Proc. Natl. Acad. Sci. 75, 1745–1749.

D'Mello, J.P.F., MacDonald, A.M.C., 1997. Mycotoxins. Anim. Feed Sci. Technol. 69, 155–166.

de Koe, W.J., 1999. Regulations of the European Union for mycotoxins in foods. Arh. Hig. Rada. Toksikol. 50, 37–46.

Devegowda, G., Castaldo, D., 2000. Mycotoxins: hidden killers in pet foods. Is there a solution? In: Technical Symposium on Mycotoxins. Alltech, Inc, Nicholasville, KY.

Dhand, N.K., Joshi, D.V., Jand, S.K., 1998. Aflatoxins in dairy feeds/ingredients. Ind. J. Anim. Nutr. 15, 285–286.

Diaz, F.J., Boermans, H.J., 1994. Fumonisin toxicosis in domestic animals: a review. Vet. Hum. Toxicol. 36, 548–555.

Diaz, D.E., Blackwelder, J.T., Hagler, W.M. Jr, Hopkins, B.A., Jones, F.T., Anderson, K.L., Whitlow, L.W., 1997. The potential of dietary clay products to reduce aflatoxin transmission to the milk of dairy cows. J. Dairy Sci. 80, 26S.

Dicostanzo, A., Johnston, L.W.H., Murphy, M., 1996. A review of the effects of molds and mycotoxins in ruminants. Prof. Anim. Sci. 12, 138–150.

Diekman, M.A., Green, M.L., 1992. Mycotoxins and reproduction in domestic livestock. J. Anim. Sci. 70, 1615–1627.

DiMenna, M.E., Mortimer, P.H., Prestidge, R.A., Hawkes, A.D., Sprosen, J.M., 1992. Lolitrem B concentrations, counts of *Acremonium lolii* hyphae, and the incidence of ryegrass staggers in lambs on plots of *A. lolii*-infected perennial ryegrass. New Zealand J. Agric. Res. 35, 211–217.

Dorrenhaus, A., Flieger, A., Golka, K., Schulze, H., Albrecht, M., Degen, G.H., Follman, W., 2000. Induction of unscheduled DNA synthesis in primary human urothelial cells by the mycotoxin ochratoxin A. Toxicol. Sci. 53, 271–277.

Eaton, D.L., Ramsdell, H.S., Neal, G.E., 1994. Biotransformation of aflatoxins. In: Eaton, D.L., Groopman, J.D. (Eds.), The Toxicology of Aflatoxins: Human Health, Vet-

erinary, and Agricultural Significance. Academic Press, San Diego, CA, pp. 45–72.
Edrington, T.S., Harvey, R.B., Kubena, L.F., 1994. Effect of aflatoxin in growing lambs fed ruminally degradable or escape protein sources. J. Anim. Sci. 72, 1274–1281.
Edrington, T.S., Harvey, R.B., Kubena, L.F., 1995. Toxic effects of aflatoxin B1 and ochratoxin A, alone and in combination, on chicken embryos. Bull. Environ. Contam. Toxicol. 54, 331–336.
El-Nezami, H., Kankaanpaa, P., Salminen, S., Ahokas, J., 1998. Ability of dairy strains of lactic acid bacteria to bind a common food carcinogen, aflatoxin B1. Food Chem. Toxicol. 36, 321–326.
Etienne, M., Jemmali, J., 1982. Effects of zearalenone (F2) on estrous activity and reproduction in gilts. J. Anim. Sci. 55, 1–10.
Fairhurst, S., Marrs, T.C., Parker, H.C., Scawin, J.W., Swanston, D.W., 1987. Acute toxicity of T-2 toxin in rats, mice, guinea pigs, and pigeons. Toxicology 43, 31–49.
Faifer, G.C., Zabal, O., Godoy, H.M., 1992. Further studies on hematopoietic damage produced by a single dose of T-2 toxin in mice. Toxicology 75, 169–174.
Fernandez, A., Ramos, J.J., Saez, T., Verde, M.T., 1995. Changes in the coagulation profile of lambs intoxicated with aflatoxin in their feed. Vet. Res. 26, 180–184.
Fernandez, A., Belio, R., Ramos, J.J., Sanz, M.C., Saez, T., 1997. Aflatoxins and their metabolites in the tissues, faeces and urine from lambs feeding on an aflatoxin-contaminated diet. J. Sci. Food Agric. 74, 161–168.
Fernandez, A., Hernandez, M., Verde, M.T., Sanz, M., 2000. Effect of aflatoxin on performance, hematology, and clinical immunology in lambs. Can. J. Vet. Res. 64, 53–58.
Feuerstein, G., Glodstein, D.S., Ramwell, P.W., Zerbe, R.L., Lux, W.E., Faden, A.I., Bayorh, M.A., 1985. Cardiorespiratory, sympathetic, and biochemical responses of T-2 toxin in the guinea pig and rat. J. Pharmacol. Exp. Ther. 232, 786–794.
Fink-Gremmels, J., 1999. Mycotoxins: their implications for human and animal health. Vet. Q. 21, 115–120.
Flowers, B., Cantley, T., Day, B.N., 1987. A comparison of the effects of zearalenone and estradiol benzoate on reproduction during the estrous cycle in gilts. J. Anim. Sci. 65, 1576–1584.
Foster, P.L., Eisenstadt, E., Miller, J.H., 1983. Base substitution mutations induced by metabolically activated aflatoxin B1. Proc. Natl. Acad. Sci. 80, 2695–2698.
Friend, D.W., Thompson, B.K., Trenholm, H.L., Boermans, H.J., Hartin, K.E., Panich, P.L., 1992. Toxicity of T-2 toxin and its interaction with deoxynivalenol when fed to young pigs. Can. J. Anim. Sci. 72, 703–711.
Gajdusek, P., 1953. Acute infectious hemorrhagic fevers and mycotoxicoses in the Union of Soviet Socialist Republics. Medical Science Publication No. 2. Walter Reed Army Medical Center, Washington.
Galey, F.D., Lambert, R.J., Busse, M., Buck, W.B., 1987. Therapeutic efficacy of activated charcoal in rats exposed to lethal doses of T-2 toxin. Toxicon 25, 493–499.
Galvano, F., Piva, A., Ritieni, A., Galvano, G., 2001. Dietary strategies to counteract the effects of mycotoxins: a review. J. Food Prot. 64, 120–131.
Gathercole, P.S., Thiel, P.G., Hofmeyr, J.H., 1986. Inhibition of pyruvate dehydrogenase complex by moniliformin. Biochem. J. 233, 719–723.
Gatlier, P., Alvineric, M., Charpenteau, J.L., 1981. The pharmacokinetic profiles of ochratoxin A in pigs, rabbits and chickens. Food. Cosmet. Toxicol. 19, 735–738.
Gekle, M., Schwerdt, G., Freudinger, R., Mildenberger, S., Wilflingseder, D., Pollack, V., Dander, M., Schramek, H., 2000. Ochratoxin A induces JNK activation and apoptosis in MDCK cells at nanomolar concentrations. J. Pharmacol. Exp. Ther. 293, 837–844.
Gelderblom, W.C.A., Snyman, S.D., 1991. Mutagenicity of potentially carcinogenic mycotoxins produced by *Fusarium moniliforme*. Mycotoxin Res. 7, 46–52.
Gelderblom, W.C., Jaskiewicz, K., Marasas, W.F., Thiel, P.G., Horak, R.M., Vleggar, R., Kriek, N.P., 1988. Fumonisins novel mycotoxins with cancer-promoting activity produced by *Fusarium moniliforme*. Appl. Environ. Microbiol. 54, 1806–1811.
Glavitis, R., Vanyi, A., 1995. More important mycotoxicosis in pigs. Magyar Allatorvosak Lapja 50, 407–420.
Gqaleni, N., Smith, J.E., Lacey, J., Gettinby, G., 1997. Effects of temperature, water activity, and incubation time on production of aflatoxins and cyclopiazonic acid by an isolate of *Apsergillus flavus* in surface agar. Appl. Environ. Microbiol. 63, 1048–1053.
Greene, H.J., Oehme, F.W., 1976. A possible case of equine aflatoxicosis. Clin. Toxicol. 9, 251–254.
Groopman, J.D., Zhu, J.Q., Donahue, P.R., Pikul, A., Zhang, L.S., Chen, J.S., Wogan, G.N., 1992. Molecular dosimetry of urinary aflatoxin-DNA adducts in people living in Guangxi autonomous region, People's Republic of China. Cancer Res. 52, 45–52.
Gurung, N.K., Rankins, D.L. Jr, Shelby, R.A., Goel, S., 1998. Effect of fumonisin B1- contaminated feeds on weanling angora goats. J. Anim. Sci. 76, 2863–2870.
Guzman, R.E., Casteel, S.W., 1994. Fumonisin mycotoxins: their origin and effects on livestock. Prof. Anim. Sci. 10, 124–129.
Hale, O.M., Wilson, D.M., 1979. Performance of pigs on diets containing heated or unheated corn with or without aflatoxin. J. Anim. Sci. 48, 1394–1400.
Harris, B. Jr, 1997. Minimizing mycotoxin problems. Feed Manag. 48, 27–28.
Harrison, L.R., Colvin, B.M., Greene, J.T., Newman, L.E., Cole, J.R. Jr., 1990. Pulmonary edema and hydrothorax in swine produced by fumonisin B_1, a toxic metabolite of Fusarium moniliforme. J. Vet. Diagn. Invest. 2, 217–221.
Harvey, R.B., Kubena, L.F., Corrier, D.E., Witzel, D.A., Phillips, T.D., Heidelbaugh, N.D., 1986. Effects of deoxynivalenol in a wheat ration fed to growing lambs. Am. J. Vet. Res. 47, 1631–1632.
Harvey, R.B., Huff, W.E., Kubena, L.F., Corrier, D.E., Phillips, T.D., 1988. Progression of aflatoxicosis in growing barrows. Am. J. Vet. Res. 49, 482–487.

Harvey, R.B., Edrington, T.S., Kubena, L.F., Elissalde, M.H.C.D.E., Rottinghaus, G.E., 1995. Effect of aflatoxin and diacetoxyscirpenol in ewe lambs. Bull. Environ. Contam. Toxicol. 54, 325–330.

Hayes, M.A., Schiefer, H.B., 1980. Comparative toxicity of dietary T-2 toxin in laboratory rats and mice. J. Appl. Toxicol. 6, 207–212.

Helferich, W.G., Garrett, W.N., Hsieh, D.P.H., Baldwin, R.L., 1986. Feedlot performance and tissue residues of cattle consuming diets containing aflatoxins. J. Anim. Sci. 62, 691–696.

Hesseltine, C.W., Shotwell, O.L., Smith, M., Ellis, J.J., Vandegraft, E., Shannon, G., 1970. Production of various aflatoxins by strains of the *Aspergillis flavus* series. Proc. 1st U.S.-Japan Conf. Toxic Microorg, Washington, DC.

Hoerr, F.J., Carlton, W.W., Yagen, B., Joffe, A.Z., 1982. Mycotoxicosis caused by either T-2 toxin or diacetoxyscirpenol in the diet of broiler chicks. Fundam. Appl. Toxicol. 2, 121–124.

Honstead, J.P., Dreesen, D.W., Stubblefield, R.D., Shotwell, O.L., 1992. Aflatoxins in swine tissues during drought conditions: an epidemiologic study. J. Food Prot. 55, 182–186.

Howard, M.D., Muntifering, R.B., Bradley, N.W., Mitchell, G.E. Jr, Lowry, S.R., 1992. Voluntary intake and ingestive behavior of steers grazing Johnstone or endophyte-infected Kentucky-31 tall fescue. J. Anim. Sci. 70, 1227–1237.

Hsu, I.C., Smalley, E.B., Strong, F.M., Ribelin, W.E., 1972. Identification of T-2 toxin in moldy corn associated with a lethal toxicosis in dairy cattle. Appl. Microbiol. 24, 682–690.

Huang, C., Dickman, M., Henderson, G., Jones, C., 1995. Repression of protein kinase C and stimulation of cyclic AMP response elements by fumonisin, a fungal encoded toxin which is a carcinogen. Cancer Res. 55, 1655–1659.

Huff, W.E., Kubena, L.F., Harvey, R.B., Doerr, J.A., 1988. Mycotoxin interactions in poultry and swine. J. Anim. Sci. 66, 2351–2355.

Hult, K., Teiling, A., Gatenback, S., 1976. Degradation of ochratoxin A by the ruminant. Appl. Environ. Microbiol. 32, 443–444.

Hughes, D.M., Gahl, M.J., Graham, C.H., Grieb, S.L., 1999. Overt signs of toxicity to dogs and cats of dietary deoxynivalenol. J. Anim. Sci. 77, 693–700.

Humpf, H.U., Schmelz, E.M., Filmore, F.I., Vesper, H., Vales, T.R., Wang, E., Menaldino, D.S., Liotta, D.C., Merrill, A.H. Jr, 1998. Acylation of naturally occurring and synthetic 1-deoxysphinganines by ceramide synthase. J. Biol. Chem. 273, 19060–19064.

Jand, S.K., Singh, P.P., Singh, A., 1995. Observations on occurrence of poultry diseases associated with mycotoxins in feed. Ind. J. Anim. Sci. 65, 1063–1067.

Jenkins, G.R., Tolleson, W.H., Newkirk, D.K., Roberts, D.W., Rowland, K.L., Saheki, T., Kobayashi, K., Howard, P.C., Melchior, W.B., 2000. Identification of fumonisin B1 as an inhibitor of argininosuccinate synthetase using fumonisin affinity chromatography and in vitro kinetic studies. J. Biochem. Mol. Toxicol. 14, 320–328.

Joffe, A.Z., Yagen, B., 1977. Comparitive study of the yield of T-2 toxin produced by *Fusarium poae*, *F. spritrichoides*, and *F. spiritrichoides* var. trininctum strains from different sources. Mycopathologia 60, 93–100.

Johannisson, A., Bjokhag, B., Hansson, W., Gadhasson, I.L., Thuvander, A., 1999. Effects of four trichothecene mycotoxins on activation marker expression and cell proliferation of human lymphocytes in culture. Cell Biol. Toxicol. 15, 203–215.

Jones, E.R.H., Lowe, G., 1960. The biogenesis of tricothecen. Chem. Soc. J. 63, 3959–3962.

Juszkiewicz, T., Jadwiga, P.P., 1992. Occurance of mycotoxins in animal feeds. J. Environ. Pathol. Toxicol. Oncol. 11, 211–215.

Kane, A., Creppy, E.E., Roth, A., Roschenthaler, R., Dirheimer, G., 1986. Distribution of the [^{3}H]-label from low doses of radioactive ochratoxin A, and evidence of for DNA single strand breaks caused in the liver and kidneys. Arch. Toxicol. 58, 219–224.

Kensler, T.W., Egner, P.A., Trush, M.A., Bueding, E., Groopman, J.D., 1985. Modification of aflatoxin B1 binding to DNA in vivo in rats fed phenolic antioxidants, ethoxyquin and a dithiothione. Carcinogenesis 6, 759–763.

Kensler, T.W., Egner, P.A., Davidson, N.E., Roebuck, B.D., Pikul, A., Groopman, J.D., 1986. Modulation of aflatoxin metabolism, aflatoxin-N7-guanine formation, and hepatic tumorigenesis in rats fed ethoxyquin: role of induction of glutathione-*S*-transferase.

Kensler, T.W., Davis, E.F., Bolton, M.G., 1991. Strategies for chemoprotection against aflatoxin induced liver cancer. In: Eaton, D.L., Groopman, J.D. (Eds.), The Toxicology of Aflatoxins: Human Health, Veterinary, and Agricultural Significance. Academic Press, San Diego, pp. 281–306.

Ketterer, P.J., Williams, E.S., Blaney, B.J., Connole, M.D., 1975. Canine aflatoxicosis. Austr. Vet. J. 51, 355–357.

Khachatourians, G.C., 1990. Metabolic effects of trichothecene T-2 toxin. Can. J. Physiol. Pharmacol. 68, 1004–1008.

Kiessling, K.H., Pettersson, H., Sandholm, K., Olsen, M., 1984. Metabolism of aflatoxin, ochratoxin, zearalenone, and three tricothecenes by intact rumen fluid, rumen protozoa, and rumen bacteria. Appl. Environ. Microbiol. 47, 1070–1073.

King, B., 1979. An outbreak of ergotism in Wollo, Ethiopia. Lancet 1, 1411.

Kitchen, D.N., Carlton, W.W., Tuite, J., 1977. Ochratoxin A and citrinin induced nephrosis in beagle dogs. II. Pathology. Vet. Pathol. 14, 261–262.

Kobayashi, J., Horikoshi, T., Ryu, J.C., Tashiro, F., Ishii, K., Ueno, Y., 1987. The cytochrome P-450-dependent hydroxylation of T-2 toxin in various animal species. Food Chem. Toxicol. 25, 539–544.

Kolb, E., 1984. Recent knowledge on the mechanism of action and metabolism of mycotoxins. Z. Gesamte. Inn. Med. 39, 353–358.

Kosuri, N.R., Smalley, E.B., Nichols, R.E., 1971. Toxicologic studies of *Fusarium tricinctum* (corda) Snyder et Hansen from moldy corn. Am. J. Vet. Res. 32, 1843–1850.

Kravchenko, L.V., Khvylya, S.I., Avreneva, L.I., Morozov, I.A., Tutelyan, V.A., 1983. Effect if T-2 toxin on organ ultrastructure and activity of organelle-specific enzyme activity in rats. Cytologia 25, 1264–1269.

Krogh, P., 1978. Causal association of mycotoxic nephropathy. Acta Pathol. et Microbiol. Scand. 269, 1S–28S.

Krogh, P., 1991. Porcine nephropathy associated with ochratoxin A. In: Smith, J.E., Anderson, R.A. (Eds.), Mycotoxins and Animal Foods. CRC Press, Boca Raton, FL, pp. 627–645.

Krogh, P., Axelson, N.H., Elling, F., Gyrd-Hansen, N., Hald, B., Hyldgaard-Jensen, J., Larsen, A.E., Madsen, A., Mortensen, H.P., Moller, T., Petersen, O.K., Ravnskov, U., Rostgaard, M., Aaluund, O., 1974. Experimental porcine nephropathy. Changes of renal function and structure induced by ochratoxin A-contaminated feed. Acta Pathol. Microbiol. Scand. 0, 1S–21S.

Kubena, L.F., Smith, E.E., Gentles, A., Harvey, R.B., Edrington, T.S., Phillips, T.D., Rottinghaus, G.E., 1994. Individual and combined toxicity of T-2 toxin and cyclopiazonic acid in broiler chicks. Poult. Sci. 73, 1390–1397.

Kuilman, M.E.M., Maas, R.F.M., Judah, D.J., Gremmels, J., 1998. Bovine hepatic metabolism of aflatoxin B1. J. Agric. Food. Chem. 46, 2707–2713.

Langouet, S., Coles, B., Morel, F., Becquemont, L., Beaune, P., Guengerich, F.P., Ketterer, B., Guillouzo, A., 1995. Inhibition of CYP1A2 and CYP3A4 by oltipraz results in reduction of aflatoxin B1 metabolism in human hepatocytes in primary culture. Cancer Res. 55, 5574–5579.

Lanza, G.M., Washburn, K.W., Wyatt, R.D., 1980. Variation with age in response of broilers to aflatoxin. Poult. Sci. 59, 282–288.

Larrson, P., Busk, L., Tjalve, H., 1994. Hepatic and extrahepatic bioactivation and GSH conjugation of aflatoxin B1 in sheep. Carcinogenesis 15, 947–955.

Larrson, P., Tjalve, H., 1996. Bioactivation of aflatoxin B1 in the nasal and tracheal mucosa in swine. J. Anim. Sci. 74, 1672–1680.

Lautraite, S., Parent-Massin, D., Rio, B., 1997. In vitro toxicity induced by deoxynivalenol (DON) on human and rat granulomonocytic progenitors. Cell. Biol. Toxicol. 13, 175–183.

Liao, L.L., Grollman, A.P., Horwitz, S.B., 1976. Mechanism of action of the 12,13- epoxytrichothecene, anguidine, an inhibitor of protein synthesis. Biochim. Biophys. Acta 454, 273–284.

Li, S., Marquardt, R.R., Frohlich, A.A., Vitti, T.G., Crow, G., 1997. Pharmacokinetics of ochratoxin A and its metabolites in rats. Toxicol. Appl. Pharmacol. 145, 82–90.

Lillehoj, E.B., 1991. Aflatoxin: an ecologically elicited activation signal. In: Smith, J.E., Anderson, R.A. (Eds.), Mycotoxins and Animal Foods. CRC Press, Boca Raton, FL, pp. 119–139.

Lindemann, M.D., Blodgett, D.J., Kornegay, E.T., Schurig, G.G., 1993. Potential ameliorators of aflatoxicosis in weanling/growing swine. J. Anim. Sci. 71, 171–178.

Long, G.G., Diekman, M.A., 1984. Effect of purified zearalenone on early gestation in gilts. J. Anim. Sci. 59, 1662–1670.

Long, G.G., Diekman, M.A., 1986. Characterization of effects of zearalenone in swine during early pregnancy. Am. J. Vet. Res. 47, 184–187.

Longouet, S., Johnson, W.W., Guillouzo, A., Guengerich, F.P., 1998. Detoxication of aflatoxin B1 as a model for carcinogen metabolism. In Vitro Mol. Toxicol. 11, 95–101.

Mann, D.D., Beuning, G.M., Hook, B., 1983. Effects of T-2 toxin on bovine serum proteins. Am. J. Vet. Res. 44, 1751–1759.

Mann, D.D., Buening, G.M., Osweiller, G.D., Hook, B.S., 1984. Effect of subclinical levels of T-2 toxin on the bovine cellular immune system. Can. J. Comp. Med. 43, 308–312.

Manning, R.O., Wyatt, R.D., 1984. Effect of cold acclimation of broiler chicks on susceptibility to acute aflatoxicosis. Poult. Sci. 63, 24S.

Manorama, Singh, R.S., 1995. Mycotoxins in milk and milk products, J. Dairying, Foods Home Sci. 14, 101–107.

Manson, M.M., Ball, H.W., Barrett, M.C., Clark, H.L., Judah, D.J., Williamson, G., Neal, G.E., 1997. Mechanism of action of dietary chemoprotective agents in rat liver: induction of phase I and II drug metabolizing enzymes and aflatoxin B1 metabolism. Carcinogenesis 18, 1729–1738.

Mantle, P.G., 1983. Amino acid neurotransmitter release from cerebrocortical synaptosomes of sheep with severe ryegrass staggers in New Zealand. Res. Vet. Sci. 34, 373–375.

Marasas, W.F.O., 1991. Toxigenic Fusaria. In: Smith, J.E., Anderson, R.A. (Eds.), Mycotoxins and Animal Foods. CRC Press, Boca Raton, FL, pp. 119–139.

Marasas, W.F.O., Bamburg, J.R., Smalley, E.B., Strong, F.M., Ragland, W.K., Degurse, P.E., 1969. Toxic effects on trout mice and rats of T-2 toxin produced by the fungus *Fusarium tricinctum* (Cd.) Snyd. et Hans. Toxicol. Appl. Pharmacol. 15, 471–482.

Marasas, W.F., Kellerman, T.S., Gelderblom, W.C., Coetzer, J.A., Thiel, P.G., van der Lugt, J.J., 1988. Leukoencephalomalacia in a horse induced by fumonisin B_1 isolated from *Fusarium moniliforme*. Onderstepoort J. Vet. Res. 55, 197–203.

Marquardt, R.R., Frolich, A.A., 1992. A review of recent advances in understanding ochratoxicosis. J. Anim. Sci. 70, 3968–3988.

Massey, T.E., Stewart, R.K., Daniels, J.M., Liu, L., 1995. Biochemical and molecular aspects of mammalian susceptibility to aflatoxin B1 carcinogenicity. Proc. Soc. Exp. Biol. Med. 208, 213–227.

McLeay, L.M., Smith, B.L., Munday-Finch, S.C., 1999. Tremorgenic mycotoxins paxilline, penitrem and lolitrem B, the non-tremorgenic 31-epilolitrem B and electromyographic activity of the reticulum and rumen of sheep. Res. Vet. Sci. 66, 119–127.

Meisner, H., Chan, S., 1974. Ochratoxin A, an inhibitor of mitochondrial electron transport systems. Biochemistry 13, 2795–2800.

Mendel, H.G., Manson, M.M., Judah, D.J., Simpson, J.L., Green, J.A., Forrester, L.M., Wolf, C.R., Neal, G.E., 1987. Metabolic basis for the protective effect of the antioxidant ethoxyquin on aflatoxin B1 hepatocarcinogenesis in the rat. Cancer Res. 47, 5218–5223.

Mertens, D.R., 1979. Biological effects of mycotoxins upon rumen fermentation and lactating dairy cows. In: Interactions of Mycotoxins in Animal Production. National Academy Press, Washington, DC, pp. 118–136.

Miles, C.O., Wilkins, A.L., Gallagher, R.T., Hawkes, A.D., Munday, S.C.T.N.R., 1992. Synthesis and tremorgenecity of paxitrols and lolitrol: possible biosynthetic precursors of lolitrem B. J. Agric. Food Chem. 40, 234–236.

Miller, D.M., Wilson, D.M., 1994. Veterinary diseases related to aflatoxin. In: Eaton, D.L., Groopman, J.D. (Eds.), The Toxicology of Aflatoxins: Human Health, Veterinary, and Agricultural Significance. Academic Press, San Diego, pp. 347–364.

Miller, D.M., Stuart, B.P., Crowel, W.A., Cole, R.J., Goven, A.J., Brown, J., 1978. Aflatoxin in swine: its effect on immunity and relationship to salmonellosis. Am. Assoc. Vet. Lab. Diagn. 21, 135–142.

Miller, D.M., Stuart, B.P., Crowel, W.A., 1981. Experimental aflatoxicosis in swine: morphological and clinical pathological results. Can. J. Comp. Med. 45, 343–351.

More, J., Gatlier, P., Eeckhoutte, C., 1990. Effect of low dosese of a tricothecene mycotoxin (diacetoxyscirpenol) on rat gastric glycoproteins: a histochemical study. Toxicol. Lett. 50, 173–178.

Moss, M.O., 1991. The environmental factors controlling mycotoxin formation. In: Smith, J.E., Anderson, R.A. (Eds.), Mycotoxins and Animal Foods. CRC Press, Boca Raton, FL, pp. 37–56.

Moubark, A.S., Rosenkrans, C.F., 2000. Hepatic metabolism of ergot alkaloids in beef cattle by cytochrome P450. Biochim. Biophys. Res. Commun. 274, 746–749.

Muench, K.F., Misra, R.P., Humayun, M.Z., 1983. Sequence specificity in aflatoxin B1-DNA interactions. Proc. Natl. Acad. Sci. 80, 6–10.

Neal, G.E., Eaton, D.L., Judah, D.J., Verna, A., 1998. Metabolism and toxicity of aflatoxins M1 and B1 in human-derived in vitro systems. Toxicol. Appl. Pharmacol. 151, 152–158.

Neldon-Ortiz, D.L., Qureshi, M.A., 1992. The effects of direct and microsomal activated aflatoxin B1 on chicken peritoneal macrophages in vitro. Vet. Immunol. Immunopathol. 31, 61–76.

Newberne, P.M., Butler, W.H., 1969. Acute and chronic effects of aflatoxin on the liver of domestic and laboratory animals: a review. Cancer Res. 29, 236–236.

Newberne, P.M., Russo, R., Wogan, G.N., 1966. Acute toxicity of aflatoxin B1 in the dog. Pathol. Vet. 3, 337–340.

Ohta, M., Ishii, K., Ueno, Y., 1977. Metabolism of trichothecene mycotoxins. I. Microsomal deacylation of T-2 toxin in animal tissues. J. Biochem. 82, 1591–1598.

Osweiler, G.D., Ross, P.F., Wilson, T.M., Nelson, P.E., Witte, S.T., Carson, T.L., Rice, L.G., Nelson, H.A., 1992. Characterizations of an epizootic of pulmonary edema in swine associated with fumonisin in corn screenings. J. Vet. Diagn. Invest. 4, 53–59.

Panangala, V.S., Giambrone, J.J., Diener, U.L., Davis, N.D., Hoerr, F.J., Mitra, A., Schultz, R.D., Wilt, G.R., 1986. Effects of aflatoxin on the growth, performance, and immune responses of weanling swine. Am. J. Vet. Res. 47, 2062–2067.

Pang, V.F., Pan, C.Y., 1994. The cytotoxic effects of aflatoxin B1 on swine lymphocytes in vitro. J. Chin. Soc. Vet. Sci. 20, 289–301.

Park, J.J., Smalley, E.B., Chu, F.S., 1996. Natural occurrence of Fusarium mycotoxins in field samples from the 1992 Wisconsin corn crop. Appl. Environ. Microbiol. 62, 1642–1648.

Patterson, D.S.P., 1973. Metabolism as a factor in determining the toxic action of the aflatoxins in different animal species. Food Cosmet. Toxicol. 11, 287–294.

Patterson, D.S.P., 1977. Metabolism of aflatoxin and other mycotoxins in relation to their toxicity and the accumulation of residues in animal tissues. Pure Appl. Chem. 49, 1723–1731.

Patterson, D.S.P., Shreeve, B.J., Roberts, B.A., Berrett, S., Brush, P.J., Glancy, E.M., 1981. Effect on calves of barley naturally contaminated with ochratoxin A and groundnut meal contaminated with low concentration of aflatoxin B1. Res. Vet. Sci. 31, 213–218.

Paul, P.S., Johnson, D.W., Mirocha, C.J., Soper, F.F., Thoen, C.C., Muscoplat, C.C., Weber, A.F., 1977. In vitro stimulation of bovine peripheral blood lymphocytes: suppression of phytomitogen and specific antigen lymphocyte responses by aflatoxin. Am. J. Vet. Res. 38, 2033–2035.

Peraica, M., Radic, B., Lucic, A., Pavlovic, M., 1999. Toxic effects of mycotoxins in humans. Bull. World Health Org. 77, 754–763.

Phillips, T.D., Clement, B.A., Park, D.L., 1994. Approaches to the reduction of aflatoxin. In: Eaton, D.L., Groopman, J.L. (Eds.), The Toxicology of Aflatoxins. Academic Press, San Diego, pp. 365–381.

Phillips, S., Wareing, P., Ambika, D., Shantanu, P., Medlock, V., 1996. The mycoflora and incidence of aflatoxin, zearalenone and sterigmatocystin in dairy feed and forage samples from Eastern India and Bangladesh. Mycopathologia 133, 15–21.

Placinta, C.M., D'Mello, J.P.F., MacDonald, A.M.C., 1999. A review of worldwide contamination of cereal grains and animal feed with *Fusariam* mycotoxins. Anim. Feed Sci. Technol. 78, 21–37.

Price, W.D., Randall, R.A., McChesney, D.G., 1993. Naturally occurring toxins in feedstuffs: Center for Veterinary Medicine perspective. J. Anim. Sci. 71, 2556–2562.

Purchase, I.F.H., Theron, J.J., 1968. The acute toxicity of ochratoxin A to rats. Food. Cosmet. Toxicol. 6, 479–483.

Quinn, B.A., Crane, T.L., Kocal, T.E., Best, S.J., Cameron, R.G., Rushmore, T.H., Farber, E., Hayes, M.A., 1990.

Protective activity of different hepatic cytosolic glutathione-*S*-transferases against DNA-binding metabolites of aflatoxin B1. Toxicol. Appl. Pharmacol. 105, 351–363.

Quist, C.F., Howerth, E.W., Fischer, J.R., Wyatt, R.D., Miller, D.M., Nettles, V.F., 1997. Evaluation of low-level aflatoxin in the diet of white-tailed deer. J. Wildlife Dis. 33, 112–121.

Ramljak, D., Calvert, R.J., Wiesenfeld, P.W., Diwan, B.A., Catipovic, B., Marasas, W.F., Victor, T.C., Anderson, L.M., Gelderblom, W.C., 2000. A potential mechanism for fumonisin B(1)- mediated hepatocarcinogenesis: cyclin D1 stabilization associated with activation of Akt and inhibition of GSK-3beta activity. Carcinogenesis 21, 1537–1546.

Ramos, J.J., Fernandez, A., Saez, T., Sanz, M.C., Marca, M.C., 1996. Effect of aflatoxicosis on blood mineral constituents of growing lambs. Small Ruminant Res. 21, 233–238.

Raney, K.D., Meyer, D.J., Ketterer, B., Harris, T.M., Guengerich, F.P., 1992. Glutathione conjugation of aflatoxin B-1 exo- and endo-epoxides by rat and human glutathione-*S*-transferases. Chem. Res. Toxicol. 5, 470–478.

Ranjan, K.S., Sinha, A.K., 1991. Occurrence of mycotoxigenic fungi and mycotoxins in animal feed from Bihar, India. J. Sci. Food Agric. 56, 39–47.

Rhodes, M.T., Paterson, J.A., Kerley, H.E., Garner, H.E., Laughlin, M.H., 1991. Reduced blood flow to peripheral and core body tissues induced by endophyte infected tall fescue. J. Anim. Sci. 69, 2033–2043.

Ribelin, W.E., Fukushima, K., Still, P.E., 1978. The toxicity of ochratoxins to ruminants. Can. J. Comp. Med. 42, 172–177.

Rio, B., Lautraite, S., Parent-Massin, D., 1997. In vitro toxicity of trichothecenes on human erythroblastic progenitors. Hum. Exp. Toxicol. 16, 673–679.

Rizzo, A.F., Atroshi, F., Hirvi, T., Saloniemi, H., 1992. The hemolytic activity of deoxynivalenol and T-2 toxin. Natural Toxins 1, 106–110.

Robb, J., 1990. Effects of mycotoxins on animal performance. In: Haresign, W., Cole, D.J.A. (Eds.), Recent Advances in Animal Nutrition. Butterworths, London, pp. 61–76.

Rompelberg, C.J., Evertz, S.J., Bruijntjes-Rozier, G.C., van den Heuvel, P.D., Verhagen, H., 1996. Effect of eugenol on the genotoxicity of established mutagens in the liver. Food Chem. Toxicol. 34, 33–42.

Ross, A.D., Bryden, W.L., Bakua, W., Burgess, L.W., 1989. Induction of heat stress in beef cattle by feeding the ergots of *Claviceps pupurea*. Austr. Vet. J. 66, 247–249.

Rukmini, C., Prasad, J.S., Rao, K., 1980. Effects of feeding T-2 toxin to rats and monkeys. Food Cosmet. Toxicol. 18, 267–269.

Rumbeiha, W.K., 2000. Clinical implications of mycotoxicosis in companion animals, Technical Symposium on Mycotoxins. Nicholasville, KY. 10-3-2000.

Russell, L., Cox, D.F., Larsen, G., Bodwell, K., Nelson, C.E., 1991. Incidence of molds and mycotoxins in commercial animal feed mills in seven midwestern states 1988–1989. J. Anim. Sci. 69, 5–12.

Sargeant, K.C.R.B.A.A.R., 1963. Chemistry and origin of aflatoxins, Chemical Ind. London, 53–55.

Sato, N., Ueno, Y., 1977. Comparative toxicities of tricothecenes. In: Rodericks, J.V., Hesseltine, C.W., Mehlman, M.A. (Eds.), Mycotoxins in Human and Animal Health, pp. 295–307.

Scheideler, S.E., 1993. Effects of various types of aluminosilicates and aflatoxin B1 on aflatoxin toxicity, chick performance and mineral status. Poult. Sci. 72, 282–288.

Schell, T.C., Lindemann, M.D., Kornegay, E.T., Blodgett, D.J., 1993. Effects of feeding aflatoxin-contaminated diets with and without clay to weanling and growing pigs performance, liver function and mineral metabolism. J. Anim. Sci. 71, 1209–1218.

Schwerdt, G., Freudinger, R., Mildenberger, S., Silbernagl, S., Gekle, M., 1999. The nephrotoxin ochratoxin A induces apoptosis in cultured human proximal tubule cells. Cell Biol. Toxicol. 15, 405–415.

Schroeder, J.J., Crane, H.M., Xia, J., Liotta, D.C., Merrill, A.H., 1994. Disruption of sphingolipid metabolism and stimulation of DNA synthesis by fumonisin B1. A molecular mechanism for carcinogenesis associated with *Fusarium moniliforme*. J. Biol. Chem. 269, 3475–3481.

Scott, P.M., 1984. Effects of processing on mycotoxins. J. Food Prot. 41, 489–492.

Seeley, T.D., Nowicke, J.W., Meselson, M., Guillemin, J., Akratanakul, P., 1985. Yellow rain. Scientific Am. 253, 128–137.

Shane, S.H., 1994. Economic issues associated with aflatoxins. In: Eaton, D.L., Groopman, J.D. (Eds.), The Toxicology of Aflatoxins: Human Health, Veterinary, and Agricultural Significance. Academic Press, San Diego, pp. 513–527.

Shen, H.M., Ong, C.N., Shi, C.Y., 1995. Involvement of reactive oxygen species in aflatoxin B1-induced cell injury in cultured rat hepatocytes. Toxicology 99, 115–123.

Shephard, G.S., Thiel, P.G., Sydenham, E.W., Vleggaar, R., Alberts, J.F., 1994. Determination of the mycotoxin fumonisin B1 and identification of its partially hydrolysed metabolites in the faeces of non-human primates. Food Chem. Toxicol. 32, 23–29.

Shinozuka, J., Li, G., Kiatipattanasakul, W., Uetsuka, K., Nakayama, H., Doi, K., 1997. T-2 toxin-induced apoptosis in lymphoid organs of mice. Exp. Toxicol. Pathol. 49, 387–392.

Silvestrini, F., Liuzzi, A., Chiodini, P.G., 1978. Effect of ergot alkaloids on growth hormone and prolactin secretion in humans. Pharmacology. 16, 78S–87S.

Skrinjar, M., Stubblefield, R.D., Vujicic, I.F., Stojanovic, E., 1992. Distribution of aflatoxin- producing moulds and aflatoxins in dairy cattle feed and raw milk. Acta Microbiol. Hung. 39, 175–179.

Skrinjar, M., Danev, M., Dimic, G., 1995. Investigation on the presence of toxigenic fungi and aflatoxins in raw milk. Acata Alimentaria 24, 395–402.

Smith, T.K., 1980. Influence of dietary fiber, protein and zeolite on zearalenone toxicosis in rats and swine. J. Anim. Sci. 50, 278–285.

Smith, T.K., 1984. Spent canola oil bleaching clays: potential for treatment of T-2 toxicosis in rats and short term inclusion in diets for immature swine. Can. J. Anim. Sci. 64, 725–732.

Smith, J.E., Moss, M.O., 1985. Mycotoxins: Formation, Analysis and Significance. Wiley, Chichester.

Smith, J.E., Ross, K., 1991. The toxigenic Aspergilli. In: Smith, J.E., Anderson, R.A. (Eds.), Mycotoxins and Animal Foods. CRC Press, Boca Raton, FL, pp. 101–118.

Smith, E.E., Kubena, L.F., Braithwaite, R.B., Harvey, R.B., Phillips, T.D., Reine, A.H., 1992. Toxicological evaluation of aflatoxin and cyclopiazonic acid in broiler chickens. Poult. Sci. 71, 1136–1144.

Southern, L.L., Clawson, A.J., 1979. Effects of aflatoxins on finishing swine. J. Anim. Sci. 49, 1006–1011.

Stetinova, V., Grossmann, V., Kvetina, J., 1998. Changes in the gastrointestinal tract, cardiovascular function and some drug metabolizing processes in rats and guinea-pigs intoxicated with aflatoxin B1. Polish J. Pharmacol. 50, 135–141.

Suneja, S.K., Ram, G.C., Wagle, D.S., 1984a. Effect of T-2 toxin administration to rats on lipid metabolism in liver. Toxicol. Lett. 22, 113–118.

Suneja, S.K., Ram, G.C., Wagle, D.S., 1984b. Effects of T-2 toxin on glucose and tryptophan uptake and intestinal mucosa enzymes. Toxicon 23, 39–44.

Suneja, S.K., Wagle, D.S., Ram, G.C., 1989. Effect of oral administration of T-2 toxin on glutathione shuttle enzymes, microsomal reductases and lipid peroxidation in rat liver. Toxicon 27, 995–1001.

Swanson, S.P., Nicoletti, J., Rood, H.D. Jr, Buck, W., Cote, L.M., 1987. Metabolism of three trichothecene mycotoxins, T-2 toxin, diacetoxyscirpenol, and deoxynivalenol, by bovine rumen microorganisms. J. Chromatogr. 414, 335–342.

Swanson, S.P., Helaszek, C., Buck, W.B., Rood, H.D., Haschek, W.M., 1988. The role of intestinal microflora in metabolism of tricothecene mycotoxins. Food Chem. Toxicol. 26, 823–830.

Tamm, C., Breitenstein, W., 1984. The biosynthesis of mycotoxins. In: Steyn, P.S. (Ed.), Mycotoxins: A Study in Secondary Metabolism. Academic Press, New York, pp. 69–91.

Taubert, G., Seboxa, T., 1990. Histological examination of blood vessels in peripheral gangrene among patients with sporadic outbreak of ergotism. Trop. Geogr. Med. 42, 58–62.

Terao, K., Ueno, Y., 1978. Morphological and functional damage to cells and tissues. In: Uraguchi, K., Yamazaki, M. (Eds.), Toxicology, Biochemistry and Pathology of Mycotoxins. Wiley, New York, pp. 189–210.

Trenholm, H.L., Friend, D.W., Hamilton, R.M.G., Thompson, B.K., Hartin, K.E., 1986. Incedence and toxicology of deoxynivalenol as an emerging mycotoxin problem. In: Proc. VI International Conf. on the Mycoses. Pan American Health Organization, Washington, DC.

Tulpule, P.G., Bhat, R.V., 1978. Food toxins and their implications in human health. Ind. J. Med. Res. 68, 99S–108S.

Tung, H.T., Smith, J.W., Hamilton, P.B., 1971. Aflatoxicosis and bruising in the chicken. Poult. Sci. 50, 795–800.

Ueno, Y., 1977. Mode of action of trichothecenes. Pure Appl. Chem. 49, 1737–1745.

Ueno, Y., Yamakawa, H., 1970. Cytotoxicity of trichothecenes. Jpn. J. Exp. Med. 40, 385–390.

Ueno, Y., Ishii, K., Sakai, K., Kanaeda, S., Tsunodo, H., 1972. Toxicological approaches to the metabolites of Fusaria. IV. Microbial survey of "bean-hulls poisoning of horses" with isolation of toxic trichothecenes, neosalaniol and T-2 toxin of *Fusarium solani* M-1-1. Jpn. J. Exp. Med. 42, 187–203.

Van Egmond, H.P., Dekker, W.H., 1995. Worldwide regulations for mycotoxins in 1994. Natural Toxins 3, 332–336.

van der Merwe, K.J., Steyn, P.S., Fourie, L., Scoot, D.B., Theron, J.J., 1965. Ochratoxin A, a toxic metabolite produced by *Aspergillus ochraceus Wilh*. Nature 205, 112–113.

van Heugten, E., Spears, J.W., Coffey, M.T., Kegley, E.B., Qureshi, M.A., 1994. The effect of methionine and aflatoxin on immune function in weanling pigs. J. Anim. Sci. 72, 658–664.

Vasanthi, S., Bhat, R.V., 1998. Mycotoxins in foods-occurrence, health & economic significance & food control measures. Ind. J. Med. Res. 108, 212–224.

Veldman, V., Meijst, J.A.C., Borggreve, J., Heeres-van der Tol, J.J., 1992. Carry-over of aflatoxin from cows' food to milk. Anim. Prod. 55, 163–168.

Vesonder, R., Haliburton, J., Stubblefield, R., Gilmore, W., Peterson, S., 1991. Aspergillus flavus and aflatoxins B1, B2, and M1 in corn associated with equine death. Arch. Environ. Contam. Toxicol. 20, 151–153.

Wang, J.S., Groopman, J.D., 1999. DNA damage by mycotoxins. Mutat. Res. 424, 167–181.

Wang, E., Norred, W.P., Bacon, C.W., Riley, R.T., Merrill, A.H., 1991. Inhibition of sphingolipid biosynthesis by fumonisins. Implications for diseases associated with *Fusarium moniliforme*. J. Biol. Chem. 266, 14486–14490.

Wang, E., Ross, F.P., Wilson, T.M., Riley, R.T., Merrill, A.H. Jr, 1992. Increases in serum sphingosine and sphinganine and decreases in complex sphingolipids in ponies given feed containing fumonisins, mycotoxins produced by *Fusarium moniliforme*. J. Nutr. 122, 1706–1716.

Wannemacher, R.W., Bunner, D.L., Neufeld, H.A., 1991. Toxicity of tricothecenes and other related mycotoxins in laboratory animals. In: Smith, J.E., Anderson, R.A. (Eds.), Mycotoxins and Animal Foods. CRC Press, Boca Raton, FL, pp. 499–552.

Watson, S.A., Mirocha, C.J., Hayes, A.W., 1984. Analysis for tricothecenes in samples from Southeast Asia associated with "yellow rain". Fundam. Appl. Toxicol. 4, 700–717.

Wei, Y.H., Lu, C.Y., Lin, T.N., Wei, R.D., 1985. Effect of ochratoxin A on rat liver mitochondrial respiration and oxidative phosphorylation. Toxicology 36, 119–130.

Weider, R., Wogan, G.N., Shimkin, M.B., 1968. Pulmonary tumors in strain A mice given injections of aflatoxin B1. J. Natl. Cancer Inst. 40, 1195–1197.

Wild, C.P., Hudson, G.J., Sabbioni, G., Chapot, B., Hall, A.J., Wogan, G.N., Whittle, H., Montesano, R., Groopman, J.D., 1992. Dietary intake of aflatoxins and the level of albumin-bound aflatoxin in peripheral blood in The Gambia, West Africa. Cancer Epidemiol. Biomarkers Prev. 1, 229–234.

Willis, R.M., Mulvihill, J.J., Hoofnagle, J.H., 1980. Attempted suicide with purified aflatoxin. Lancet 1, 1198–1199.

Withanage, G.S., Murata, H., Koyama, T., Ishiwata, I., 2001. Agonistic and antagonistic effects of zearalenone, an etrogenic mycotoxin, on SKN, HHUA, and HepG2 human cancer cell lines. Vet. Hum. Toxicol. 43, 6–10.

Wogan, G.N., 1966. Chemical nature and biological effects of the aflatoxins. Bacteriol. Rev. 2, 460–470.

Wogan, G.N., Newberne, P.M., 1967. Dose-response characteristics of aflatoxin B1 carcinogenesis in the rat. Cancer Res. 27, 2370–2376.

Wood, G., 1989. Aflatoxins in domestic and imported foods and feeds. J. Assoc. Offic. Anal. Chem. 72, 543–548.

Wood, G., 1992. Mycotoxins in foods and feeds in the United States. J. Anim. Sci. 70, 3941–3949.

World Health Organization International Agency for Research on Cancer (IARC), 1993a. Toxins derived from *Fusarium moniliforme*: fumonisins B1 and B2 and fusarin C. IARC Monographs on the Evaluation of Carcinogenic Risks to Humans 56, 445–462.

World Health Organization International Agency for Research on Cancer (IARC), 1993. IARC Monographs on the Evaluation of Carcinogenic Risks to Humans.

Worell, N.R., Mallett, A.K., Cook, W.M., Baldwin, M.C.P., Shepherd, M.J., 1989. The role of gut micro-organisms in the metabolism of deoxynivalenol administered to rats. Xenobiotica 19, 25–32.

Xiao, H., Srinivasa, M., Marquardt, R.R., Li, S., Vodela, J.K., Frohlich, A.A., Kemppainen, B.W., 1996. Toxicity of ochratoxin A, its opened lactone form, and several of its analogs: structure-activity relationships. Toxicol. Appl. Pharmocol. 137, 182–192.

Yeung, J.M., Wang, H.Y., Prelusky, D.B., 1996. Fumonisin B1 induces protein kinase C translocation via direct interaction with diacylglycerol binding site. Toxicol. Appl. Pharmacol. 141, 178–184.

Yoshizawa, T., Yamashita, A., Luo, Y., 1994. Fumonisin occurrence in corn from high- and low-risk areas for human esophageal cancer in China. Appl. Environ. Microbiol. 60, 1626–1629.

Zhang, H., Li, J.L., 1989. Study on toxicological mechanism of moniliformin. Wei Sheng Wu Xue Bao 29, 93–100.