Mycotoxins and Immunotoxicity

Neil E. Forsberg, Professor
Department of Animal Sciences
Oregon State University

Mycotoxins and immunity. Fungi produce a variety of secondary metabolites which function as mycotoxins. At high concentrations, mycotoxins directly affect specific organs or tissues including the liver, kidney, oral and gastric mucosa, brain, or reproductive tract (1). In these cases, the signs of disease often are clearly apparent and attributable to impairment of specific tissues. At lower concentrations, effects of mycotoxins may be more subtle. They reduce the growth rate of young animals and some interfere with mechanisms of resistance and immune function, making the animals more susceptible to infections. These effects on immunity and resistance are difficult to recognize because the signs of disease are associated with the infection rather than with the mycotoxin that predisposed the animal to infection (1).

Approximately 400 mycotoxins have been isolated and many are immunosuppressive. The feed-borne immunosuppressive mycotoxins include aflatoxins, fumonisins, gliotoxin, ochratoxins, patulin and trichothecenes. The molds responsible for production of these toxins are listed in Table 1.

Table 1. Mycotoxin classes, specific toxins and mold species commonly detected in livestock feeds

Mycotoxin class	Examples of specific toxins	Mold species
Aflatoxins	aflatoxin B1 aflatoxin G1	*Aspergillus flavus* and *A. parasiticus*
Fumonisins	fumonisin B1 fumonisin B2 fusarin C moniliformin	*Fusarium verticilloides, F. moniliforme* and *F. proliferatum*
Gliotoxin	gliotoxin	*Gliocladium, A. fumigatus,* and *Candida albicans*
Ochratoxins	ochratoxins A and B	*Aspergillus ochraceus, Penicillium verrucosum*
Patulin	Patulin	various *Penicillium* and *Aspergillus species*
Trichothecenes	-Group A: (T2 toxin, diacetoxyscirpenol) -Group B: (nivalenol, vomitoxin/DON) -Group C/D trichothenes	various *Fusarium, Stachybotrys* and *Myrothecium* species

The immune system. The immune system consists of two distinct, yet interacting, systems. These include the innate and the adaptive systems. The innate system consists of structural, chemical and enzymatic barriers to pathogen entry, activities of phagocytic cells (e.g., macrophages, monocytes and neutrophils which sequester and digest pathogens) and natural killer (NK) cells which recognize and kill pathogen-infected cells and cancer cells. The adaptive system consists of B-lymphocytes (humoral immunity), T-lymphocytes (cell-mediated immunity) and antigen presenting cells. Activities of all aspects of the immune system are intricately coordinated by a mixture of signaling molecules which include cytokines, chemokines and specific receptors on immune cells.

The complexity of the immune system implies that there are dozens, possibly hundreds, of methods by which immune function may be assessed. The literature is rich with examples of how mycotoxins compromise immune function; however, the diverse approaches available to quantify “immune function” make it difficult to compare studies and to assess relative importance of mycotoxins as immunosuppressive agents. Because of this, prominent scientists (2) have recommended an establishment

of standards for assessing effects of mycotoxins on immune function. Their recommendations for assessment of innate, humoral and cellular immunity are provided in Table 2 (2).

Table 2. Recommendations by which specific aspects of the immune system may be assessed.

Immunologic parameter	Recommended assessment strategy
-innate immunity	-NK cell activity -macrophage and neutrophil function
-humoral immunity	-antibody response to sheep red blood cells (SRBC)
-cellular immunity	-lymphoproliferative responses to T-cell antigens -delayed-type hypersensitivity responses

In the remainder of this review, summaries of the immunosuppressive activities of individual mycotoxins are provided. The limitation of the data set is that inconsistent methods for assessing immune function have been utilized. However, the strength of the data set is that, for many mycotoxins, immunosuppression and immunomodulation are consistently observed.

Aflatoxins. Aflatoxins have clear negative effects on all aspects of the immune system of poultry, swine and ruminants. In fact, treatment of females with aflatoxin results in transmission via the egg (poultry) and from the sow to the piglet and thereby reduces immune function in offspring. Fewer studies have been completed on effects of aflatoxins on immune functions of ruminant animals but, in general, data indicate impairment. For example, aflatoxin-fed lambs have reduced bacteriostatic activity in serum and reduced *in vivo* cellular immunity (3). Aflatoxins cause failure in the acquired immunity system of lambs by decreasing antibody production (4). In dairy animals, feeding of AFB1 reduces appetite, body weight and milk yield (5). Bacterial populations in the milk increase during aflatoxin consumption (5). Aflatoxin suppresses the lymphocyte response to mitogens and the lymphocyte response of *M. bovis*-infected animals to specific antigen is significantly suppressed by aflatoxin (6).

Fumonisins. The fumonisins are unusual because they exert some species-specific, non-immunotoxic effects on horses (leukoencephalomalacia) and swine (pulmonary edema). However, their immunotoxic effects are not species-specific. The fumonisins also specifically inhibit humoral, cellular and innate aspects of the immune system. Compared to swine, poultry are relatively resistant to fumonisins. However, chickens are more sensitive to fumonisin B1 whereas turkeys are more sensitive to fumonisin B2. In poultry, fumonisins reduce thymus weight, reduce antibody responses to sheep red blood cells and to immunization against *B. abortus* and Newcastle disease (2). Fumonisins also reduce macrophage number, phagocytotic ability of macrophages and decrease total white blood cell counts in poultry. In pigs, fumonisins reduce lymphocyte blastogenesis and titers against pseudorabies (2). In ruminant species, fumonisins have less effect than in swine and horses. In ruminants, fumonisins are reported to reduce neutrophil migration and mitogen-activated lymphocyte blastogenesis (7).

Gliotoxin. This toxin is produced by *A. fumigatus* and *C. albicans*, molds which infect immunocompromised animals. Substantial *in vitro* data indicate that gliotoxin is immunotoxic; however, these observations have not yet been fully supported by *in vivo* studies. *In vitro*, gliotoxin reduces neutrophil migration and phagocytosis and is cytotoxic to lymphocytes (8). It has been used successfully as an immunosuppressive adjunct in transplants. Gliotoxin inhibits one interesting aspect of the innate immune system: the mucociliary system (9) of the lung which facilitates removal of air-borne pathogens (such as *A. fumigatus*).

Ochratoxins. Ochratoxin A (OA) causes porcine nephropathy and may be the cause of a fatal human kidney disease in the Balkans (2). Ochratoxin A exerts suppressive effects on all aspects of immunity. In poultry, OA reduces serum immunoglobulins, monocyte phagocytic activity, lymphocyte proliferation and causes delayed hypersensitivity reactivity. Studies have reported that OA increases susceptibility to *S. typhimurium* and *E. Coli* infection in chickens. Ochratoxin A reduces mitogen-stimulated proliferation of

bovine peripheral blood mononuclear cells *in vitro* (10). A recent study investigated cytogenetic and cytotoxic effects of both OA and zearalenone (Z: an estrogenic *Fusarium* mycotoxin) on bovine lymphocytes (10). Both OA and Z caused bovine chromosome aberrations and sister chromatid exchanges. Ochratoxin A-induced programmed cell death was not limited to bovine lymphocytes, as comparable data were demonstrated in a human leukemic T-cell line (11).

Patulin. Immunotoxic properties of patulin have been investigated primarily in laboratory (rodent) species. Specific immunotoxic and immunomodulatory actions of patulin include increased splenic T lymphocytes, depressed serum IgG, depressed delayed hypersensitivity responses and increased neutrophil numbers (1). Patulin increased resistance to *C. albicans* (1). Extrapolation of rodent doses to humans has led authors to conclude that patulin would not likely alter immunity (2) and potential effects in livestock are not known.

Trichothecenes. Trichothecenes are a large group (>180) of compounds (including T2 toxin and deoxynivalenol) which are potent protein synthesis inhibitors. They specifically target mitotic tissues including those with important immune functions (e.g., bone marrow, lymph nodes, spleen, thymus and intestinal mucosa). Depending upon dose and exposure regimen, trichothecenes may be either immunosuppressive or immunostimulatory (2). Repetitive exposure to trichothecenes increases susceptibility to pathogens including *Mycobacterium*, *Candida*, *Cryptococcus*, *Listeria*, *Salmonella*, *Aspergillus* and herpes simplex Type 1 (2). T2 toxin results in a 100,000-fold reduction in LD50 for *Salmonella* in mice (2). In contrast, T2 toxin increases resistance to mastitis-causing infections and *Listeria* in mice. Other effects of T2 toxin on the immune system have included impaired lymphocyte proliferation. The effects of T2 toxin and diacetoxyscirpenol on this parameter are greater than those caused by the Group B trichothecenes (e.g., vomitoxin and nivalenol).

Summary. This review has summarized details on immunomodulation and immunotoxicity by some of the better-known mycotoxins. It is essential to also recognize that mycotoxins, in addition to their immunomodulatory and immunotoxic actions, impair other aspects of animal physiology and health. A partial summary of these impairments is presented in Table 3.

Table 3. Effects of common mycotoxins on animal physiology and performance

Mycotoxin	Known impairments
Aflatoxins	Hepatotoxicity, cancer, bile duct hyperplasia, intestinal hemorrhage, GI dysfunction, decreased feed efficiency, reduced egg and milk production, embryonic death, prolonged clotting.
Deoxynivalenol (DON) or vomitoxin	Reduced feed intake, lower milk production, elevated milk somatic counts, reduced reproductive efficiency, feed refusal in pigs.
Fumonisins	Neurological problems in horses, pigs and poultry. Reduced weight gains in feeder cattle.
Ochratoxins	Decreased body weight, feed intake and blood proteins. Bile duct hyperplasia in poultry. Nephropathies in many species. Impaired semen quality in swine. Potentially carcinogenic.
T2 toxin	Feed refusal, production losses, gastroenteritis, intestinal hemorrhage, death, vomiting, abortion.
Zearalenone	Estrogenic responses, abortion, reduced feed intake, decreased milk production, vaginitis, vaginal secretions, poor reproductive performance, mammary gland enlargement, atrophy of testis and ovaries, poor feed intake

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