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Technical Bulletin

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Mycotoxins and their effect on Animal Production

Mycotoxins

Mycotoxins are fungal metabolites that cause an undesirable effect in animals and humans. Mycotoxicosis are diseases caused by exposure to foods or feeds contaminated with mycotoxins (Nelson et al., 1993).

Mycotoxins exhibit a variety of biological effects in animals such as: liver and kidney toxicity, central nervous system abnormalities, estrogenic responses etc. They are organic compounds with low molecular weight and no immuno-geneticity.

In tropical and subtropical climates, the fungi development is enhanced by factors such as high humidity and temperature. Fungi grow and spread well in cereals (corn, wheat, barley, sorghum and rice) and peanuts, in which they generally finds a highly nutritious substrate for their development. Fungi growth and mycotoxin production in cereals can occur in several development phases of maturation, harvest, transportation, processing or storage of grains and forages.

Indeed the Food & Agriculture Organization (FAO) estimates that 25% of world crops are affected, leading to 5-10% of commodities being discarded.

More than 400 mycotoxins, presently known, are produced by approximately one hundred fungi. The main mycotoxins can be divided into three groups (Table 1).

Factors influencing mycotoxin contamination

Molds can grow and mycotoxins can be produced either pre-harvest or post-harvest, during storage, transport, processing, or feeding. Mold growth and mycotoxins

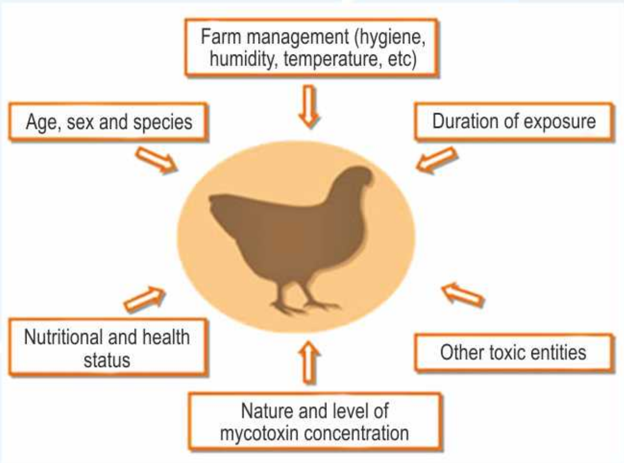


Table 1:

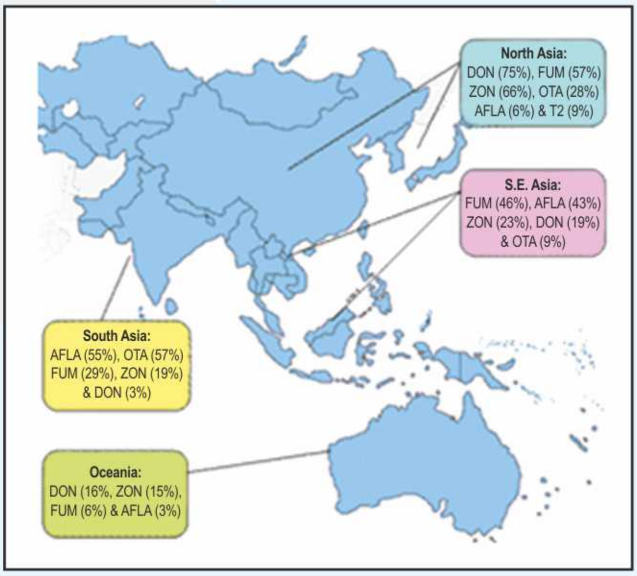
Moulds	*Fusarium spp.*	*Aspergillus spp.*	*Penicillium spp.*
Mycotoxins	Deoxynivalenol, Zearalenone, T-2 Toxin, Fumonisin, Moniliformin, Diacetoxyscirpenol, Fusaric acid etc.	Aflatoxin, Ochratoxin, Sterigmatocystin, Cyclopiazonoic acid etc.	Ochratoxin, PR Toxin, Citrinin, Cyclopiazonic acid etc.

Table 2

Climatic Conditions	*Crop Practices*	*Other Factors*
Temperature	Cropping pattern	Mechanical Harvesting
Humidity	Crop Variety	Improper Storage
Frost	Soil Characteristics	Moisture in the grain before storage
Air	Delayed Harvest	Insect infectations
Rain at Harvest	Post Harvest handling of Grain	Broken seed coat

production are related to weather extremes (causing plant stress or excess hydration of stored feedstuffs), inadequate storage practices, low feedstuff quality, and faulty feeding conditions. In general, environmental conditions – heat, water, and insect damage – cause plant-stress and predispose plants in the field to mycotoxin contamination.

Molds grow over a temperature range of 10-40° C (50-104° F), a pH range of 4 to 8, and above 10 percentage moisture level in feed (Table 2).



Prevalence Of Mycotoxin in Different Geographical Regions Within Asia-Pacific

North Asia:

Occurrence of mycotoxins in samples received from North Asia was 6%, 55% 75% 58% and 28% for Aflatoxins, Zearalenone (ZON), Deoxynivalenol (DON), Fumonisin (FUN) B1 and OchratoxinA (OTA) respectively. T-2 toxin was found in less than 1% of the total sample population. For aflatoxin, the highest level detected was 494 ppb in a corn sample from China. DON was prevalent in this region (75%) and the highest level detected was in a DDGS sample from China with 10,374 µg/kg. ZON and Fumonisin, on the other hand, were found in 55% and 58% of the samples respectively. Highest level of ZON found was 14,105 µg/kg in a corn gluten meal sample from China. For fumonisin B1, it was 14,714 µg/kg in a corn sample also from China. Ochratoxin A was detected in 28% of the samples; 82 µg/kg in a soybean meal sample from China was the maximum level analyzed.

South-East Asia:

The prevalence of mycotoxin contamination in South-East Asia is in the order as follows; Fumonisin (45%), Aflatoxin (40%), Zearalenone (23%), Deoxynivalenol (19%) and Ochratoxin (9%). T-2 toxin was not detected in the current analysis.

South Asia:

Although the number of samples analyzed from this region was comparatively smaller (total 31), prevalence of aflatoxin (55%) was evident. Occurrence of ZON and Fumonisin were observed at 19% and 29% respectively. OTA though was found in 57% of the samples, the levels detected were not high (maximum of 10 µg/kg). DON was found in less than 1% of the samples and T-2 toxin was not detected in any of the samples analyzed.

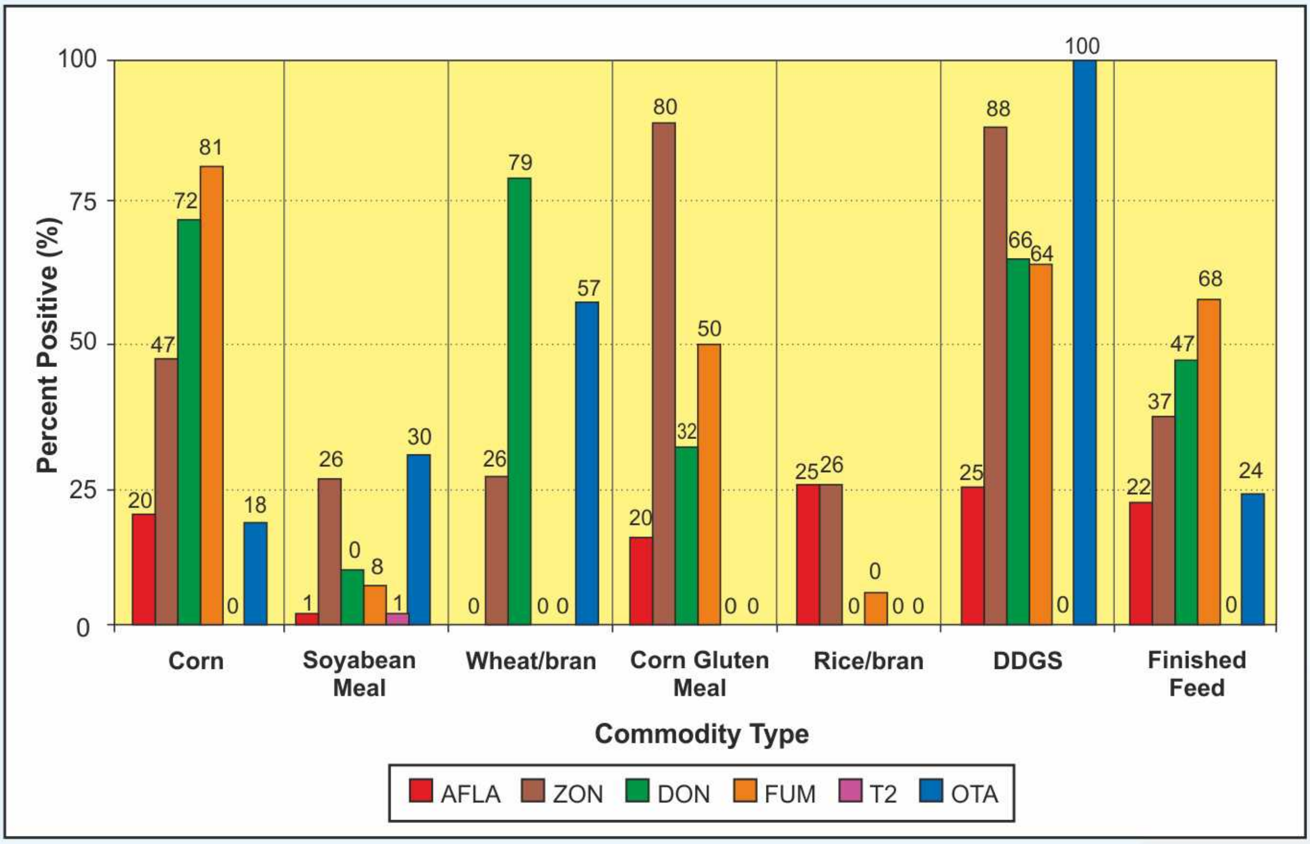
Oceania:

More than half of the samples analyzed were straws/ hay. The prevalence of mycotoxins from this region was DON (16%), ZON (15%), Fumonisin (8%) and Aflatoxins (3%). OTA and T-2 toxin were not detected in these samples.

FDA guidelines for acceptable Aflatoxin level in corn based on intended use (www.fda.gov)

Intended use	Aflatoxin level (ppb)
Milk (Dairy Feed)	None detected
Corn of unknown destination	<20
Corn for young animals	<20
Corn for dairy cattle	<20
Corn for breeding beef, cattle, swine, and mature poultry	<100
Corn for finishing swine	<200
Corn for finishing cattle	<300

Prevalence Of Mycotoxins In Different Commodities
(Ref. Limien Tan (Romer Labs Singapore Pte.)



Infections in Grains

Plants may be contaminated by mycotoxins in two ways.

- As fungi growing as pathogens on plants in the field
- Secondly, there are fungi growing on stored harvested plants

In this context, it has to be considered that not all of these fungi are able to produce mycotoxins or produce them in different amounts depending on the substrate on which they are growing. However, high incidence rates of contamination of cereal grains and animal feed have been reported worldwide. Cereal grains constitute the major portion of animal feeds. The breakup of different mycotoxins in respective feed ingredient is shown below which allows for better mycotoxin monitoring in the individual raw material.

Fungal Infections in Animals

Poultry birds and other animals are affected by fungal infections in four different ways:

- Feed ingredients in the field before harvest
- Feed ingredients during storage after harvest
- Mixed feeds in bulk bins and on feeding equipment
- Direct contamination

The fungi developed by first three ways exert deleterious effects in three inter-related forms. Production of toxic metabolites (mycotoxins), modification of nutrient composition of feed ingredients and modification of the nutrient utilization by the animals.

Mycotoxins in Different Animals

Mycotoxins in Poultry

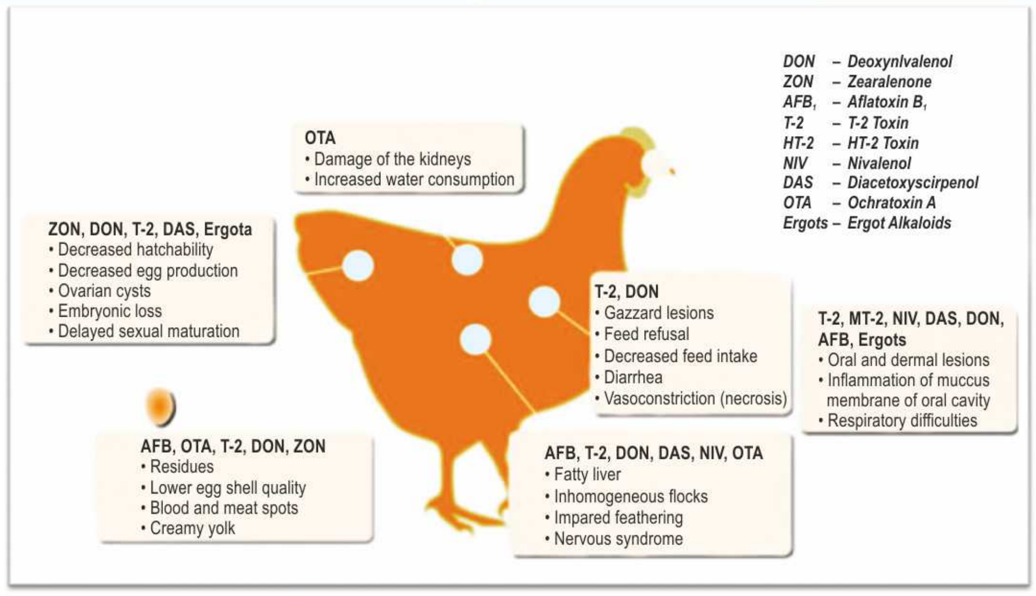
1. Aflatoxins

Layer consuming diets containing aflatoxin 5 ppm for 4 weeks, may show a reduction in egg production from the eighth day, reaching a decrease in production of around 35%, one week after removal of the mycotoxin diet (ROSA et al., 2001).

In outbreaks of aflatoxicosis, one of the most important features is the poor absorption of feed that is manifested by the presence of poorly digested feed particles in the feces of birds. It is associated with steatorrhea (Fat in droppings) or increased excretion of lipids. This can be severe with an increase of up to ten times of fat content in feces. In broilers, the steatorrhea is accompanied by a decrease in total and specific activities of pancreatic lipase, the main digestive enzyme from fat and a decrease of bile salts, necessary both for digestion and for absorption of fat, leading to hepatic steatosis (fatty liver). Mucosal pallor and legs is also observed in chickens and hens receiving feed contaminated with aflatoxins. This seems to be a deficient pigmentation result of reduced absorption, transport and decreased tissue deposition of carotenoids in the diet, and this type of aflatoxicosis is identified as "pale bird syndrome".

2. Trichothecenes

The main group of trichothecene mycotoxins includes the T-2 toxin, deoxynivalenol (DON or vomitoxin) and diacetoxyscirpenol (DAS).



Chronic contamination involving T- 2 toxin or DAS induce reduction in feed intake and weight gain, oral lesions, tissue necrosis lymphoid, haematopoietic and oral mucosa, with possible nervous disorders (abnormal position of the wings, decreased reflexes), abnormal feathering and thinning of the eggshells. Particularly in layers, oral lesions occur in approximately 50% of the flocks where these birds are fed a diet containing 2 ppm of T- 2 toxin.

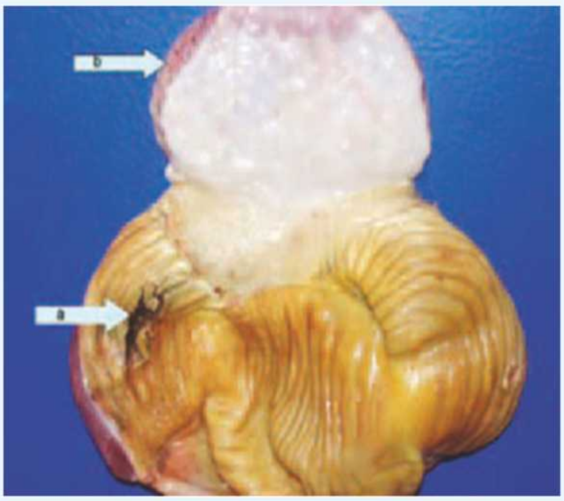
This toxin also induces the formation of peroxides from lipids, with the consequent decrease in the concentration of vitamin E in poultry. Mycotoxins T-2 and DAS induce oral lesions in broiler chickens when present at levels from 1 ppm in the diet. The birds have reduced food consumption, growth retardation, changes in blood parameters and neurotoxicity.

Trichothecenes generally do not induce an increase in mortality in birds other than chickens, requiring levels of several hundred parts per million (ppm) to result in significant mortality. Similarly, in outbreaks of mycotoxicosis attributed to T- 2 toxin that affected domestic ducks, geese, horses and pigs, mortality was observed only in geese, which suggests a greater sensitivity of these birds.

3. Fumonisins

In birds contaminated by fumonisins, clinical signs generally include reduce weight gain, mortality, diarrhea, ascites, edema and renal congestion, ulceration in the oral mucosa in turkeys, an increase in relative liver weight, proventriculus and gizzard.

Some studies indicate that toxic levels of fumonisins are above 80 ppm. Other researchers have performed experiments with extremely high doses of fumonisin (61 to 546 ppm) and found adverse effects of this toxin on performance of broilers.



The Caustic Effect of Some Fusariotoxins Is The Cause Of The Commonly Detected Erosions And Ulcers In Gizzard Cuticulum (Arrow a). Note The Thickened Wall Of The Proventriculus (Arrow b).

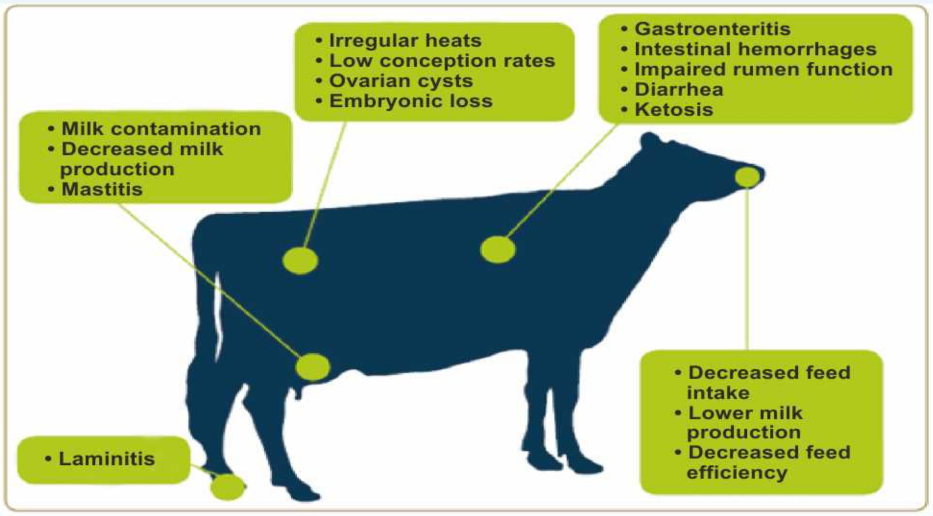
4. Ochratoxin

The minimal dietary growth inhibitory dose of Ochratoxin for young broiler chicks is 2 µg/g. Visible examination of the chicks showed no clinical symptoms due to ochratoxin, but the study of affected broiler chickens has shown severe hydration and emaciation, proventricular haemorrhages, and visceral gout with white urate deposits throughout the body cavity and internal organs. The main target organ is the kidney, with the liver also affected to a lesser degree. The kidneys of affected birds become enlarged, and the livers exhibit a tan coloured appearance, have an increased glycogen content, but the liver fat does not increase. Inhibition of liver phosphorylase and decreased mitochondrial respiration are thought to be responsible for the increased liver glycogen. This results in a decrease in egg production, food consumption and serum protein concentrations, and an increase in prothrombin times.

Aflatoxicosis and ochratoxicosis result in a rubbery condition of the bones apparently related to increased tibial diameters and perhaps poor mineralization of bone tissue in young broiler chicks.

5. Cyclopiazonic acid

The main clinical signs include decrease in weight gain, vomiting, and neurologic signs (opisthotonus, hyperesthesia, and seizure) and is usually fatal. Lesions include degeneration and necrosis of the liver, hemorrhagic lesions in the myocardium, proventriculus, gizzard and spleen (Kuilman-Wahls, 2002). Among the injuries



mentioned above, the most significant is the presence of erosions in the gizzard of birds contaminated.

Mycotoxins in Dairy Cattle

1. Aflatoxin

Aflatoxin contaminated feed not only reduces animal performance and overall health, but it also creates risks of residues in milk. Aflatoxin is secreted into milk in the form of aflatoxin M1 with residues approximately equal to 1 to 2 percent (1.7 percent average) of the dietary level. This ratio is not influenced greatly by milk production level since high yielding cows consume more feed and have a slightly higher transmission rate. Due to risks of milk residues, dietary aflatoxin should be kept below 25 ppb.

This level is conservative due to:

(1) Non-uniform distribution of aflatoxin in grain and feed

(2) Uncertainties in sampling and analysis

(3) The potential for having more than one source of aflatoxin in the diet. Replacement animals may tolerate 50 to 100 ppb aflatoxin.

2. DON (Vomitoxin)

DON is associated with reduced feed intake, lower milk production, elevated milk somatic cell counts, and reduced reproductive efficiency. Milk production loss appears to occur when diets contain more than 300 ppb DON. Although controlled research has shown no cause and effect relationship between DON levels and reduced milk production, field observations have shown that reductions in milk output of 11 kg per cow were seen when DON was 500 ppb or more. This suggests that DON may serve as a marker for feed that was exposed to a situation conducive to mold growth and mycotoxin formation. Dietary levels of 300 to 500 ppb DON in dairy feeds indicate mycotoxin problems and warrant attention.

3. Zearalenone

It causes estrogenic responses in dairy cattle, and large doses of this toxin are associated with abortions. Other responses of dairy animals to zearalenone may include reduced feed intake, decreased milk production, vaginitis, vaginal secretions, poor reproductive performance, and mammary gland enlargement in virgin heifers. Establishment of a tolerable level of zearalenone for dairy cattle is difficult, and is at best only a guess based on a meager amount of data and field observations. As with DON, zearalenone may serve as a marker for toxic feed. It is suggested that zearalenone should not exceed 250 ppb in the total diet.

4. T-2 toxin

It has been associated with feed refusal, production losses, gastroenteritis, intestinal hemorrhages, and death. T-2 has also been associated with reduced immune response in calves. Data with dairy cattle are not sufficient to establish a tolerable level of T-2 in the diet. Therefore, a practical recommendation may be to avoid T-2 in excess of 100 ppb in the total diet for growing or lactating dairy animals.

5. Fumonisin

It is another commonly isolated mycotoxin. However, fumonisin has only recently been isolated and only enough data exists to know that levels in excess of 20,000 ppb are potentially toxic to ruminants.

Beef Cattle

Aflatoxin and other mycotoxins can have considerable effects on beef cattle although the problems are usually less critical than for swine and poultry. Consumption of feeds highly contaminated with aflatoxin may reduce growth rate and increase the amount of feed required per pound of gain. Calves are generally more sensitive to feed contamination than adult cattle. In affected calves, some cases have revealed severe rectal straining and a prolapsed rectum. Lactating cows show a significant reduction in milk yield. Research has shown that high levels of aflatoxin can also cause liver damage in adult cattle. Feeding a high level of aflatoxin may also depress immune function, resulting in disease outbreaks.

Mycotoxins in Pig/ Swine

1. Aflatoxins

Symptoms may include feed refusal, reduced growth, paleness and jaundice. The latter is an indication of liver damage which may occur at levels above 100 ppb in the feed. Feed refusal may be observed at levels as low as 20

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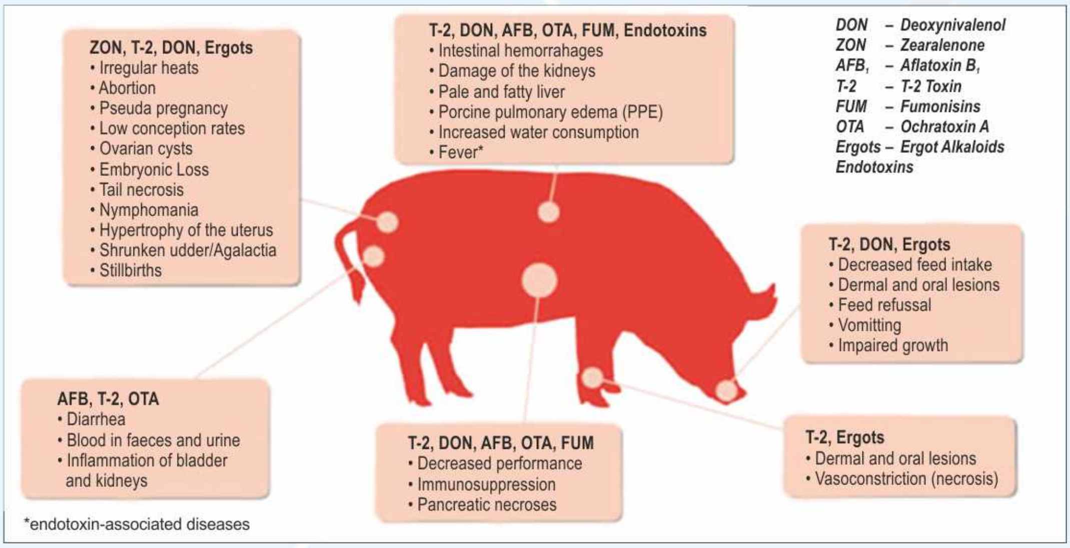
Possible mechanism for the impaired vaccine efficacy in piglets receiving fumonisin B1



ppb, depending on the variety of aflotoxin.

Possible mechanism for the impaired vaccine efficacy in piglets receiving fumonisin B1

2. Fumonisins

Symptoms of fumonisin contamination may be observed at levels above 10 ppm and include pulmonary edema, liver damage and reduced antibody titre.

3. Trichothecenes

Pigs are more sensitive to these mycotoxins than other farm animals, with levels as low as 300 to 500 ppb resulting in feed refusal, decreased weight gain and increased vulnerability to infectious diseases. The trichothecens include deoxynivalenol (DON), also known as vomitoxin because of its strong vomiting effect. At levels over 1 ppm, severe weight loss and vomiting occur. Weaning pigs are especially susceptible. Recent research suggests that, male pigs may be more tolerant than females. Effects on reproduction are unknown, but it is best to avoid feeding of breeding animals with infected grain.

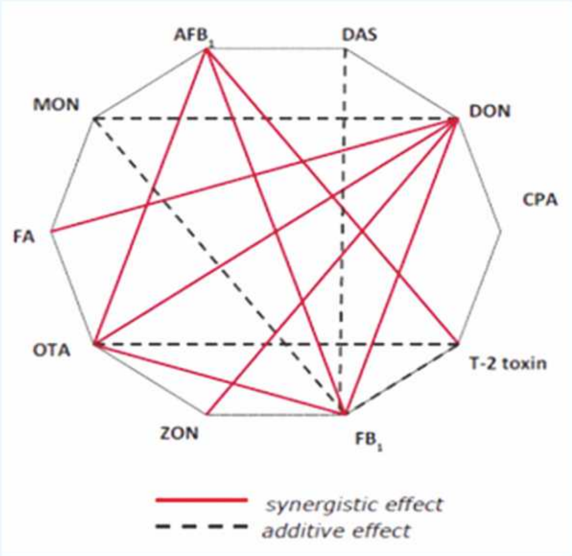
4. Ochratoxins

Ochratoxin A produces depressed appetite and reduced growth rate. At concentrations greater than 5 to 10 ppm, a number of conditions may arise such as impaired kidney function, necrosis of lymph nodes and fatty liver changes.

5. Zearalenone

Zearalenone is probably the mycotoxin most detrimental to swine with serious effects on the breeding stock. Pre pubertal gilts are most sensitive. Toxicity results in the reddening and swelling of the vulva, increased size of mammary tissue, straining with subsequent rectal and vaginal pro lapse, as well as pseudo pregnancy and false heat. The piglets of affected sows may experience depressed piglet growth in utero, early embryonic mortality and may be born with splayed legs. Fertility problems surface at 100 to 200 ppb. Zearalenone also produces swelling of the prepuce in boar

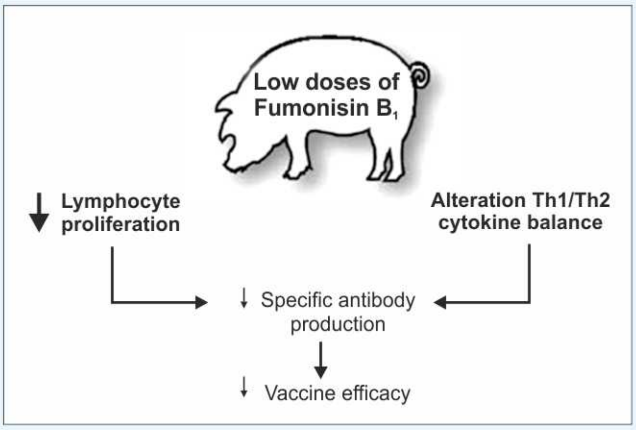
Synergistic and additive effects of mycotoxins in pigs

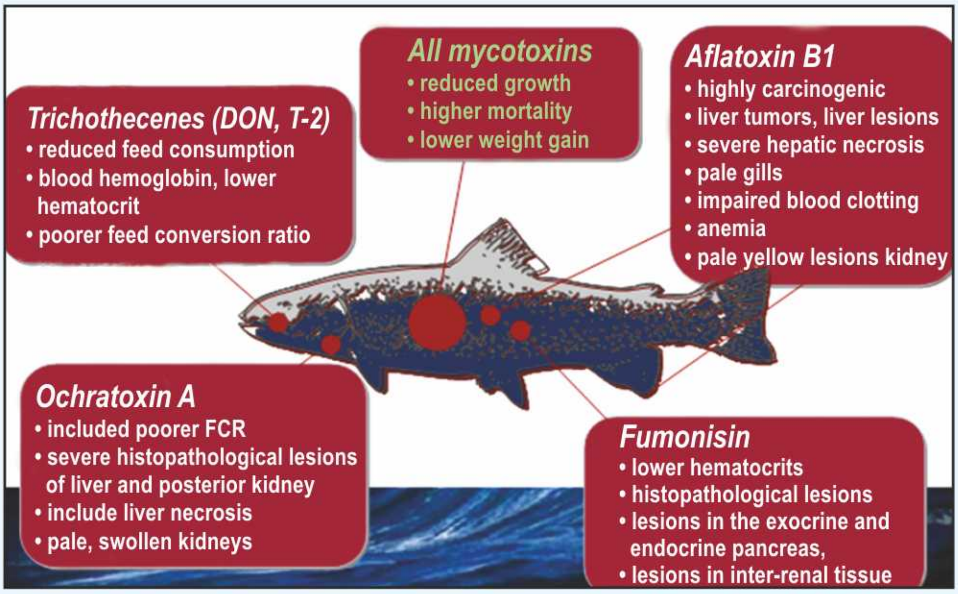


Mycotoxins in Aqua

1. Aflatoxins

Aflatoxin was the first of the mycotoxins to be investigated in aquaculture. As in other animal species, aflatoxin exerts carcinogenic effects in fish. Wolf and Jackson (1963) traced hepatomes in rainbow trout exposed to increased





concentrations of dietary aflatoxin B1; and exposure to low levels of aflatoxin was observed to cause hepatocellular carcinomas. In a 10-week feeding period, tilapia fingerlings were reported to tolerate 50 ppb aflatoxin B1 with little or no effect on performance. However, fingerlings showed a reduction in performance when feeding time was increased.

A reduction in performance was observed within 10 weeks when the mycotoxin dose was raised to 100 ppb (El-Banna et al., 1992). Since aflatoxin can impair immune function (Ottinger and Kaattari, 2000), exposure increases fish susceptibility to disease. A healthy fish is less likely to succumb to secondary infections and has a greater tolerance for the toxin. Reduced immune function has been reported in Indian major carp (Labeo rohita) after exposure to aflatoxin B1 in the feed at doses as low as 1.25 mg/kg body weight (Sahoo and Mukherjee, 2001). Sahoo and Mukherjee (2002) later reported that addition of high levels of α-tocopherol in feed (1000 mg/kg) significantly improved immune response in fish exposed to aflatoxin-contaminated feed.

2. Ochratoxin A

Higher inclusion levels (4-8 ppm) significantly reduced feed conversion efficiency and haematocrit values. Interestingly, in contrast to observations in mammalian and avian species, the toxin did not lead to any necrotic changes in the renal tubules. Necrosis was only reported in hepato-pancreatic tissue at toxin concentrations of 1000 ppb and above. In catfish, ochratoxin A has been shown to reduce weight gain when fed at 1000 ppb for 8 weeks (Manning et al., 2003b), although 500 ppb did not affect weight gain.

Shalaby (2004) noted a reduction in erythrocyte count, haemoglobin concentration and haematocrit value in response to ochratoxin (at 400 and 600 µg/kg of feed) in tilapia, possibly due to destruction of mature red blood cells and inhibition of new erythrocyte production. This effect is similar to that observed in African catfish (Clariasgariepinus) exposed to ochratoxin (Mousa and Khattab, 2003).

3. Trichothecenes and Zearalenone

Trichothecenes have been intensively researched, and are known to affect aquatic species. Chemically, they are structurally similar to compounds such as deoxynivalenone (DON), T2-toxin and diacetoxyscirpenol (DAS). In mammalian and avian species, trichothecenes have been shown to reduce feed intake and performance and impair immune function. In rainbow trout, Woodward et al. (1983) reported that diets containing levels of 1.0 to 12.9 ppm DON caused progressively greater reductions in 4-week live weight gain in juveniles. The depression in weight gain ranged from 12 to 92% compared with the control, and resulted from adverse effects on both feed intake and feed conversion. Complete feed refusal was observed when dietary DON concentrations reached >20 ppm.

Zearalenone has estrogen-like activity that has detrimental effects on the fertility of mammals, although it is probably of less importance in aquaculture. Arukwe et al. (1999) did report that zearalenone could affect reproductive success and the development of fish eggs, and α- zearalenone, one

Effect of different dietary concentrations of DON on performance of shrimp in a 16-week trial

	0 ppm	0.2 ppm	0.5 ppm	1.0 ppm
Final weight, g	11.22^{a}	10.63^{ab}	10.43^{b}	9.67^{c}
FCR	3.25	3.15	3.66	3.79
Survival, %	86.4	90.9	81.8	83.3
DON level in shrimp	ND	ND	ND	ND




Impact of Toxins - Shrimps	
Aflatoxin	Reduced growth at 100 ppb
DON	Reduced growth at 200 ppb
T-2	Reduced growth at 100 ppb
ZON	Reduced growth at 100 ppb
Impact of Toxins - Catfish	
Aflatoxin	Reduced growth 50 ppb (Pangasus catfish)
Aflatoxin	Reduced growth 40 ppb (Thai catfish)
Fum	Reduced growth fingerlings (Channel catfish)
T-2	0.652 ppm reductior in weight gain (Channel catfish)
Impact of Toxins - Tilapia	
Aflatoxin	Reduced growth at 100 ppb
DON	No date available
Moniliformin	Minor effect
Impact of Toxins - Trout	
Aflatoxin	Carcinogenic at 5 ppb
DON	Reduced weight gain at 1 ppm

of its metabolites, has been shown to reduce the number and quality of sperm in carp (Sándor and Ványi, 1990).

Toxic levels reported in both trout and shrimp seem comparable to concentrations reported in swine, where concentrations of 1 ppm or more are considered problematic, although in young piglets, lower concentrations have been shown to reduce feed intake (Spring and Strickler, 2004).

4. Fumonisin

Presence of Fumonsin mycotoxins suppresses growth of tilapia and catfish fingerlings and fish show liver & Kidney lesions. Potential risk of contamination remains despite in plant management of mycotoxin contamination.

Maximum tolerable concentrations (ppb, micrograms/Kg) of some mycotoxins in the complete feeding stuffs and in different aqua species is given in the chart.

Mycotoxins in Horses

Although the effects of mycotoxins on horses are not well documented in scientific literature, in field situations apparent mycotoxin problems appear to be significant. Mycotoxins have been implicated in a variety of health problems including colic, neurological disorders, paralysis, hypersensitivity, and brain lesions. The cumulative effect of feeding low levels of mycotoxins may also contribute to a gradual deterioration of organ functions. This in turn affects growth rate, feed efficiency, fertility, respiration rate, the ability to perform work, and life span. Due to the lack of conclusive scientific research concerning the levels of various mycotoxins tolerated by the horse, emphasis should be placed on feeding mycotoxin free grain and forage to all horses.

Horses are herbivores with a simple stomach (non-ruminant). The large intestine has an active microbial digestive ability to allow digestion of forages. However, in the horse the small intestine, which is the major site of absorption, occurs before the fermentative digestion. As a result, horses are more susceptible to mycotoxins than ruminants in which absorption occurs after fermentative digestion.

Productive or working horses have a high energy requirement and require a higher concentrate intake, and thus would be most susceptible to problems with mycotoxins contaminated grains. Working horses would include growing horses less than two years of age, brood mares in late gestation and early lactation, and horses at moderate or intense work levels.

Other horses, that are only lightly worked, would be more likely to be exposed to mycotoxin contaminated hays or forages. Since moldy forages are generally less palatable than normal forage, horses fed moldy forages typically refuse feed before ingesting enough feed to cause severe intestinal tract damage. Mild colic is typically noted in such cases. Grain mycotoxins are readily absorbed and should be considered to be potentially lethal for horses.

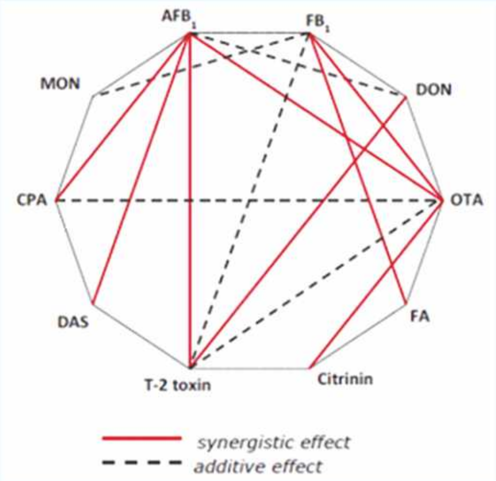
What are the antagonistic effects of mycotoxicosis?

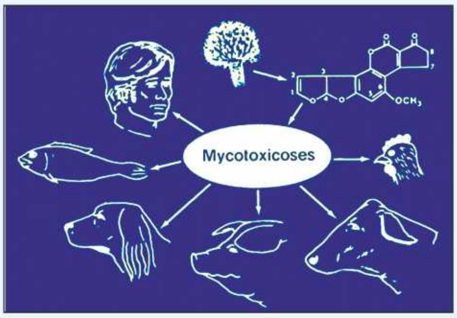
Genetic progress has produced breeds and hybrids in animals capable of ever-higher production performance. Due to their increased metabolic rate these breeds are less tolerant to mycotoxin contamination in feed. Mycotoxicosis is difficult to diagnose as symptoms are very common and affect morethan one organ. Although large doses of mycotoxins are needed to kill a broiler, contaminated feed can greatly reduce broiler performance resulting in serious economic losses for broiler producers. The synergy between different types of mycotoxins has deleterious effects on the birds. The effect of two or more mycotoxins fed to a bird simultaneously is often greater than the effect of either mycotoxin individually. Secondly it is also seen that it is not the case with all mycotoxins. Kubena, 1997 reported that there was synergy between Fumonisin and T-2 but not between Fumonisin and DON.

Synergistic And Additive Effects Of Mycotoxins In Poultry

Mycotoxins exert their effects through three primary mechanisms:

1. A reduction in the amount of nutrients available for use by the animal. This occurs in a multi factorial process. First, an alteration in nutrient content of feed may occur during the molding process. Mold growth can reduce the content of nutrients such as vitamins and amino acids like lysine in feedstuffs (Kao and Robinson, 1972). The energy value of feeds is usually reduced by mold growth. Second, some mycotoxins reduce feed intake which lowers nutrient intake. Third, a mycotoxin produced irritation to the digestive tract can reduce nutrient absorption, and fourth, certain mycotoxins interfere with normal nutrient metabolism such as the inhibition of protein synthesis by T-2 toxin.
2. Effects on the endocrine and exocrine systems. An example is the effect of zearalenone on reproductive performance due to its estrogenic effects. Zearalenone's estrogenic effect results from the affinity of zearalenone and its derivatives to bind with the animal's estrogen receptors (Klang et al., 1978).





3. Suppression of the immune system. The effects of mycotoxins on immunity have been reviewed (Sharma, 1993). Trichothecenes such as DON and T-2 toxin reduce immunity by inhibiting protein synthesis and thus cell proliferation. Some mycotoxins are cytotoxic tolymphocytes in vitro. Corticosteriods produced in response to stress also reduces immune function.

Mycotoxins can increase incidence of disease and reduce production efficiency. In the field, animals experiencing mycotoxicosis may exhibit a few or many symptoms including: digestive disorders, reduced feed consumption, unthriftiness, rough hair coat or abnormal feathering, under nourished appearance, subnormal production, impaired reproduction, and/or a mixed infectious disease profile.

The primary mechanisms through which mycotoxins affect animals are:

1. Reduction of feed intake
2. Reduced nutrition (reduced nutrient content of the feed, reduced nutrient absorption and altered nutrient metabolism)
3. Immuno-suppression
4. Mutagenicity
5. Teratogenicity
6. Cellular death

How to Manage Mycotoxins?

The mycotoxins problem in poultry has been well defined. Researchers are using their knowledge of the chemistry and biology of mycotoxins to construct prevention and neutralization mechanisms. The threat of mycotoxins is minimized by operating at two levels:

a) Prevention of mycotoxins
b) Treatment of mycotoxins

Prevention of mycotoxins start at the crop or plant levels, where the crop is harvested and grains are stored for the further usage in feed production.

Following methods may be adopted to minimize mycotoxins after harvest:

1. Harvest at the maturity and as soon as the moisture content allows minimum grain damage: This can be done when the moisture for Maize is 23-25 % ;Sorghum (Jowar)-12-17%; Soybeans 11-15 % etc. Adjust the harvesting equipments for minimum seed or kernel damage and maximum cleaning.

2. Dry all grain to at least 15 %moisture as rapidly as possible ,not to exceed a 24-48 hour period after harvest. Safe, long term storage can be achieved at uniform moisture level of 13 % or somewhat below. Slow drying (accomplished by low heat or natural air drying) is being used increasingly, but the grain can contain no more than 20-21% moisture in full bin drying.

3. Another possibility is high temperature (60-65° C.) drying until the grain reaches 20-21% moisture followed by low heat drying to 13% moisture. Cool the grain after drying and maintain dry storage conditions. When possible continue cooling until the grain temperature reaches 2°C to 5°C.

4. Thoroughly clean the grain and all bins before storage to remove dirt, dust, and other foreign matter, crop debris, chaff, and cracked or broken seeds and kernels. Caution has to be taken as mold infected kernels are fragile and easily broken. Broken or damaged kernels are more likely to be mycotoxin contaminated. The use of seed or grain, cleaning equipment can significantly reduce the mycotoxins content (particularly aflatoxin) of a grain lot. Storage has to be done in sealed air tight structures. Continue periodic aeration and probing for hot spots at intervals of 1 to 4 weeks throughout the storage period. The analytical tests now in use can detect levels of aflatoxin lower than those of toxicological significances in raw materials and finished food products and feeds. Use of propionic acid or a mixture with ammonium is obutyrate, on high moisture grain during storage is recommended. Although this acid will not remove any Aflatoxins already present in the grain, it will prevent the growth offungi if properly applied. Grains treated with propionic acid can be used only for livestock and poultry feeds. Where feasible, choose varieties of grain that are resistant to insects, diseases, and mechanical damage. Any damage to the grain provides a route of entry for Aspergillus flavors and other toxin forming fungi. Once the fungus or fungi has invaded the plant, then the appropriate environmental conditions will lead to toxin formation.

Preventive Measures

Growers and feeders can utilize several practices to minimize mycotoxin contamination. Some of these are as follows:

- Use recommended production practices. Everything should be done to maximize yields. It appears that Aflatoxins are not as much of a problem in corn where the yields are high as they are when yields are low. Therefore, only recommended practices should be followed.

- Plant early. Research has demonstrated that corn pollinated during periods of adequate moisture and moderate temperatures has less risk of high aflatoxin contamination. Such conditions are more likely to occur when the corn is planted early. Early planted corn is also generally exposed to lower insect populations.

- Dry and store crop properly A. flavus cannot grow in corn with a moisture content less than 12 to 13%. Therefore, if the corn is dried below this level, no additional growth of the fungus or production of aflatoxin will occur if proper storage practices are followed.

- Keep storage and feeding facilities clean. The fungus can survive in residues left in storage and feeding facilities and can rapidly produce mycotoxins under such conditions.

The poultry farmer usually relies on grain producers and traders to get quality raw ingredients for poultry feed. All the above parameters are not in his hand to ensure quality of his raw material. The different measures, which can be used by the farmer/feed producer, are discussed below. However, increasing protein levels is a costly approach to mycotoxins control.

Methionine Supplementation: Veltmann et al observed that increasing the dietary total sulphur amino acids to levels in excess of NRC, protected chicks from the growth depressing effects of aflatoxin, possibly through an increased rate of detoxification by Glutathione, a sulfur amino acid metabolite.

Dietary Lipids: Lipids exert their effects in part by interfering with absorption of the aflatoxin. Diets containing higher levels of linolenic acid supported better-feed conversion and lower mortality in chicks fed diets with aflatoxin.

Vitamin Supplementation: Supplemental vitamins did not differ in their response to dietary aflatoxin as compared to birds fed diet with four times the NRC Recommended levels.

Antioxidants: Antioxidants differ in their ability to alleviate aflatoxicosis. BHT was able to relieve the adverse effect of aflatoxicosis, when added at levels of 8-30 times the normal usage level but in no case were the adverse effects of aflatoxicosis were completely overcome.

Pelleting: Pelleted poultry feed with or without a propionic acid based mold inhibit or indicated that pelleting and propionic acid interacted to reduce the mold count in a model system. It appears that the effectiveness of propionic acid as a mould inhibitor can be greatly increased by the pelleting process.

Use of Mold Inhibitors

A number of products like low molecular weight organic acids and their salts (for eg. Propionic acids) are widely used as mold inhibitors. It is important to note that these are fungi stats and not fungicides; that is, they only inhibit the further growth of molds and do not actually kill them. Mold inhibitors will not keep mold growth in check indefinitely.

Non- Nutritive adsorbents

One of the most promising aspects of controls of mycotoxins is the use of various adsorbents to reduce their adverse effects. Sorbents act by reducing the bioavailability of mycotoxins by adsorption on their surface. Indeed, if a stable sorbent mycotoxins complex is formed, the absorption of mycotoxins in the gastrointestinal tract can be reduced, decreasing both toxic effects for the animal and carry over in animal products for human consumption.

With this aim numerous sorbents from different sources have been tested, such as hydrated sodium calcium alumiono silicate (HSCAS), zeolites, bentonites, clays and activated carbons. HSCAS, a phyllosilicate derived from natural zeolite, is perhaps the most extensively investigated sorbent. It is ineffective in binding dangerous mycotoxins other than Aflatoxin B1. Its protective properties are very low towards Ochratoxin and Zearalenone and nil toward Trichothecenes. Zeolites are hydrated alumiono silicate of alkali and alkaline earth cations characterized by infinite three-dimensional structure. The pore size distribution of synthetic Zeolites, as opposed to natural ones, varies very little, being generally concentrated within a narrow diameter range. If the size of the pores is compatible with those of the mycotoxins molecules, adsorption can occur. On the contrary, adsorption can be low or nil due to the absence of intermediate sized pores. Bentonites are sorbents with layered (lamellar) crystalline microstructure and variable composition. Their adsorption properties mainly depend on the interchangeable cations. Other clays such as kaolin, sepiolite and montmorillonite have a variable ability to reduce toxic effects of Aflatoxin B1 as reviewed by Ramos et al. However, their efficacy is limited to Aflatoxin B1 and is lower than that of HSCAS and bentonite. Activated Carbons (AC) are another important group of sorbents. They are a family of carbonaceous substances manufactured by activation processes aimed at developing a highly porous structure. Overall evidence of the high ability of AC in binding mycotoxins in vitro has been seen. The highest abilities have been observed in the adsorption of AFB1 and OTA (Ochratoxin A), whereas the lowest in the adsorption of DON (Deoxynivalenol). AC has been demonstrated to adsorb efficiently FB1 simultaneously with AFB1. When compared to HSCAS, AC showed much higher adsorption abilities toward all the tested mycotoxins.

Activated carbon was the best adsorbent, binding 100% ZEA (pH 3 and 7.3) at 0.1, 0.25, 0.5, and 1% dose levels. Bentonite, talc, and calcium sulfate were less efficient than activated carbon but still could bind ZEA to some extent. On the other hand, sandstone was inactive in the experimental conditions employed. Our results indicate that activated carbon could be a good candidate for detoxification of ZEA present in foods. (Bueno Dante J.; di Marco Liliana; Oliver Guillermo; Bardón Alicia)

Activated Carbon

Some salient features of Activated carbon include;

- Active over a wide variety of Mycotoxins including Aflatoxins, DON, T-2 toxin and even Zearalenone
- Excellent adsorbent of bacterial endotoxins, pesticides, insecticides and other harmful agents
- MB Index of 250 mg/g indicates the mesoporosity which adsorbs mycotoxins effectively

Conclusion

Minimizing the adverse effects of mycotoxins in animal diets is not a simple task that can be accomplished with one intervention strategy. It begins with the purchase of quality grains (characterized by low moisture content and minimal broken kernels), followed by proper handling of grains during mixing and delivery. Use of mycotoxin binding agents seems to be a very promising approach to the detoxification of mycotoxins. The positive outcome from the initial investigations of AFB1 binding to HSCAS and bentonite fostered additional interest in this approach and further studies were conducted on other sorbents and mycotoxin. Activated carbon is a promising sorbent which has shown higher adsorption abilities towards all tested mycotoxins in in-vitro test. The adsorption properties of activated carbon are strictly dependent on the source material and physico-chemical parameters such as surface area and pore size distribution. Chemical treatments can strongly modify activated carbon surface characteristics. Because of the ubiquitous nature of mycotoxins in nature, the problem of minimizing mycotoxicosis will not be easy to solve and will require constant attention to detail throughout the entire process of grain harvest, shipping, storage, feed manufacturing and animal production. No one segment of the animal production chain can totally protect or prevent the problems associated with Mycotoxicosis. A holistic approach is needed to minimize the adverse effects, enhance overall animal production and produce a safe food for human consumption.

Table 1: Factors influencing Mycotoxin contamination in Feed stuffs

High temperature	High Humidity	Mechanical Harvesting
Drought	Weather condition at harvest	Cropping practices
Frost	Soil Characteristics	Plant variety
Delayed Harvest	Improper storage	

Table 2: Symptoms of Mycotoxicosis in Poultry

Severe depression in body weight gain and diarrhoea	Lesions on the margins of the beak and esophageal Mucosa
Fatty liver hemorrhagic syndrome	Enlarged liver, kidney and spleen
Increased requirement for Methionine, antioxidants and other nutrients	Anemia/reduced hematocrit
Depressed egg production and weak shells	Lower Hatchability
Poor feathering and skin pigmentation	Impaired immunity
Increased incidence of tibial dysplasia	Regression of bursa of Fabricius
Focal necrotic lesions on the liver	

Table 3: Major Mycotoxins, fungal species, Feed ingredients affected and the toxic effects

Mycotoxins	Fungal Species	Feed ingredients affected	Possible toxic effects on animals
Aflatoxins (B1, B2, G1 and G2)	*A. Flavus and A. Parasiticus*	Cereals Grains, Groundnuts, soybeans	Hepatotoxin: carcinogenic; reduced growth rate; hemorrhagic enteritis; suppression of Natural immunity to infection; decreased Production of meat, milk and eggs
Ochratoxins	*A. Ochraceous* *P. viridicatum*	Cereal Grains	Toxic to kidneys and liver, Poor feed conversion, reduced growth rate. General Unthriftiness; reduced immunity to infection
Patulin	*P. Urticae*	Cereal Grains P. expansion P. claviforme and A. clavatus	Hemorrhages of lung and brain; edema; toxic to kidneys; possible carcinogenic
Citrinin	*P. Citrinum*	Cereals especially in Rice and Groundnut	Kidney damage. Haemorrhages, Lymphoid necrosis. Immunosuppression in poultry
Fusarium Toxins a. Zearalenone	*F. Graminearum* *F. Tricinctum*	Cereal grains	Ingertility, stunting and even death
b. Vomitoxin, Dexoynivalenol or DON	*F. Graminearum*	Cereal grains	Reduction in weight gain
c. Trichothecenes (T-2, HT-2, Diacetoxyscripenol Or DAS)	*F. Tinctum*, some strains of *F. Graminearum* *F. Equuiseti* *F. lateritium* *F. poae* and *F. sporotrichoides*	Cereal grains	Severe inflammation of gastrointestinal tract and possible hemorrhage; oedema; vomiting and diarrhoea; infertility; degeneration of bone marrow; death; reduced weight gain, slow growth, sterility.
Fumonisin B1, B2	P. Monoliforme	Maize	Leuco/encephalomalacia

Table 4: Effect of some of common mycotoxines on pullet and layer performance

Mycotoxin	Toxin Level in feed	Symptoms
Aflatoxin	50 ppb	Increased embryonic Mortality
	1 ppm	Decreased body weight and egg weight
	2 ppm	Decreased egg Production
	2.7 ppm	Decreased hatchability
T-2 Toxin	20 ppb	Oral Lesions
	2 ppm	Decreased hatchability and Vomiting
	3 ppm	Gizzard Lesions, dermatitis sub epidermal haemorrhages
	4 ppm	Decreased weight gain in pullets, poor feathering
	5 ppm	Feed Refusal in Pullets
	8 ppm	Feed Refusal in adult birds. Decrease production and thin shells. Decreased chick weight.
Ochratoxin	1 ppm	Delayed Maturity, decreased production, Hatchability and progeny performance.
	2 ppm	Increased Mortality
	2.5 ppm	Decreased weight gain, poor feed efficiency.
	4 ppm	Feed refusal and decreased egg weight, Cessation of production
Zearalenone	50 ppm	Decreased oviduct and Liver weight
	250 ppm	Decreased shell quality
Citrinin	220 ppm	Decrease weight gain
	400 ppm	Decreased feed efficiency
Deoxynivalenol (DON)	200 ppb	Feed refusal
	350 ppb	Fatty Liver
	16 ppm	Decreased Growth
Cyclopiazonic Acid	50 ppm	Decreased Body weight. Proventriculus and Mucosa

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