FA95

UF UNIVERSITY of FLORIDA
IFAS Extension

Molds in Fish Feeds and Aflatoxicosis[1]

Juli-Anne Royes Russo and Roy P.E. Yanong[2]

Aflatoxicosis and Aflatoxins

Aflatoxicosis is a disease that can affect many species of fish, and results when feed contaminated with aflatoxins is eaten by the fish (Ashley, 1970). Aflatoxins are chemicals produced by some species of naturally occurring fungi (*Aspergillus flavus* and *Aspergillus parasiticus*) commonly known as molds. Aflatoxins are common contaminants of oilseed crops such as cottonseed, peanut meal, and corn. Wheat, sunflower, soybean, fish meal, and nutritionally complete feeds can also be contaminated with aflatoxins.

Four major aflatoxins (AFB1, AFB2, AFG1 and AFG2) are direct contaminants of grains and finished feeds. Factors that increase the production of aflatoxins in feeds include environmental temperatures above 27°C (80°F), humidity levels greater than 62%, and moisture levels in the feed above 14%. The extent of contamination will vary with geographic location, feed storage practices and processing methods. Improper storage is one of the most important factors favoring the growth of aflatoxin-producing molds, and it is a major element that can be controlled by the fish producer.

Implications of Aflatoxins in Fish Feeds

Aflatoxin B1 (AFB1) is one of the most potent, naturally occurring, cancer-causing agents in animals. The first documented incidences of aflatoxicosis affecting fish health occurred in the 1960s in trout hatcheries. Domesticated rainbow trout (*Oncorhynchus mykiss*) that were fed a pelleted feed prepared with cottonseed meal contaminated with aflatoxins, developed liver tumors (Ashley, 1970). As many as 85% of the fish died in these hatcheries. Although cottonseed meal is no longer used as a major ingredient in feed formulations, poor storage of other feed ingredients and nutritionally complete feeds can lead to contamination with aflatoxins.

Aflatoxicosis is now rare in the rainbow trout industry due to strict regulations enforced by the U.S. Food and Drug Administration (FDA) for aflatoxin

1. This document is Fact Sheet FA-95, one of a series from the Department of Fisheries and Aquatic Sciences, Florida Cooperative Extension Service, Institute of Food and Agricultural Sciences, University of Florida. First published: March 2002. Major revision: August 2006. Please visit the EDIS Web site at http://edis.ifas.ufl.edu.

2. Juli-Anne Royes Russo, Post Doctoral Research Associate, Fish Nutrition, Hubbs-SeaWorld Research Institute, San Diego, CA 92109; and Roy P.E. Yanong, Associate Professor, Tropical Aquaculture Laboratory - Ruskin, FL 33570-3434, Department of Fisheries and Aquatic Sciences; Florida Cooperative Extension Service, Institute of Food and Agricultural Sciences, University of Florida.

screening in oilseeds, corn and other feed ingredients. However, interest in the toxic effects on cultured warm-water fishes, such as tilapia (*Oreochromis* sp.) and channel catfish (*Ictalurus punctatus*), has increased as diets for these species are now being formulated to contain more plant and less animal ingredients. This increases the potential for development of aflatoxicosis in these species because, as noted earlier, plant ingredients have a higher potential than animal ingredients for contamination with aflatoxins.

In tropical and subtropical conditions, this potential is further increased due to storage under humid and hot conditions. International trade in affected commodities and exposure to aflatoxins are worldwide concerns and the economic impact due to animal losses can be enormous.

The extent of disease, caused by consumption of aflatoxins, depends upon the age and species of the fish. Fry are more susceptible to aflatoxicosis than adults and some species of fish are more sensitive to aflatoxins than others.

Studies on the Nile tilapia (*Oreochromis niloticus*) showed reduced growth rates when tilapia were fed diets containing 1.8 milligrams (mg) of AFB1 per one kilogram (kg) of feed for 75 days. In addition, tissue abnormality or lesions in the livers of these tilapia showed the beginnings of cancer development. Another study (Tuan et al. 2002) tested effects of varying concentrations of AFB1 on 2.7-gram Nile tilapia. Fish fed diets that contained 2.5, 10, or 100 mg AFB1 per kg of feed for 8 weeks had reduced weight gain and reduced red blood cell counts. Fish fed the 10 mg AFB1 per kg feed had abnormal livers. Those fed 100 mg AFB1 per kg feed had weight loss and significant damage to the liver, and 60 % of these fish died by the end of the experiment. Other studies have shown that tolerance levels for tilapia can vary with the production system. In green water and flow-through systems, the presence of aflatoxins at 25 to 30 parts per billion (ppb) in the water decreased growth without any noticeable signs of mortality. However, in cage culture, concentrations of aflatoxins above 5 ppb in the water caused an increase in mortality rates.

Rainbow trout are extremely sensitive to AFB1, while channel catfish are much less responsive. Rainbow trout fed diets containing AFB1 at 0.0004 mg per kg feed (0.4 ppb) for 15 months had a 14% chance of developing tumors. Feeding rainbow trout a diet containing AFB1 at 0.02 mg per kg feed (20 ppb) for 8 months resulted in 58% occurrence of liver tumors, and continued feeding for 12 months resulted in 83% incidence of tumors. Channel catfish, fed a diet containing purified AFB1 at 10 mg per kg feed (10,000 ppb) for 10 weeks, exhibited decreased growth rates and moderate internal lesions (Jantrarotai and Lovell 1990).

Signs of Aflatoxicosis in Fish

Initial findings associated with aflatoxicosis include pale gills, impaired blood clotting, anemia, poor growth rates or lack of weight gain. Prolonged feeding of low concentrations of AFB1 causes liver tumors, which appear as pale yellow lesions and which can spread to the kidney. Increases in mortality (higher numbers of dead fish) may also be observed.

Aflatoxins can cause disease indirectly through their effects on essential nutrients in the diet. For example, fat soluble antioxidants, such as vitamin A, and water soluble antioxidants and vitamins, such as vitamin C (necessary for immune function) and thiamin (necessary for metabolic and nervous function), in feeds can be destroyed by these toxins. Hence, it is not surprising that aflatoxins depress the immune system, making fish more susceptible to bacterial, viral or parasitic diseases. These subtle effects often go unnoticed and profits are lost due to decreased efficiency in production, such as slow growth, reduced weights of the finished product, an increase in the amount of feed needed to reach market weight, and increased medical costs.

Management and Control

Purchase of feeds that have been recently prepared and properly stored is recommended. Debris must be removed from feed ingredients and grains should be stored in clean bins or buildings. Where possible, complete fish feeds should be stored in an air-conditioned building for temperature and humidity control. Otherwise, feed should be stored

off the ground, on pallets and at least one foot away from any walls (to avoid condensation) in a cool, dry area and for no longer than three months. If feed is held in bins outside, storage for longer than two weeks is not recommended.

When feeds are stored for long periods or under poor conditions, fish health problems may arise, not only from molds, but also from loss of vitamins and rancidity of oils in the feed. Control of rodents and insects is also important in maintaining nutrient quality and aflatoxin-free feeds.

Feeds that have the manufacturer's date stamped on the bags will prevent the purchasing of old feed. It is also a good idea to be familiar with when the feed was bought, and how the feed was being stored by the feed supplier prior to purchasing feed.

Feeds stored for a long time and probably contaminated with molds appear stale, are discolored, lump together and smell musty. Stale foods are often saturated with moisture and appear to 'sweat'. Any containers that are used to store food (bins, automatic feeders) should be cleaned thoroughly on a regular basis to prevent mold growth on their surfaces (which may be hidden by newly placed fresh feed).

Regular testing for aflatoxins is a good idea. Simple on-farm inspection can be done visually (look for the presence of blue/grey mold on feed) or with a black light which may cause a bright greenish/yellow fluorescence if *A. flavus* is present. While the black light method is a rapid procedure, it is only a potential indicator of the presence of *A. flavus*, and it may not work in all cases.

Commercial kits for detecting concentrations of aflatoxins in feeds, such as Veratox® for aflatoxins and Afla BTM, are highly recommended if aflatoxicosis is suspected. These kits are now readily available from companies such as Neogen Corporation and VICAM. They are easy to run, require less than 20 minutes for analysis, are inexpensive and can be used on the farm. More information on these products can be found on the internet at the Neogen Corporation and VICAM Web sites listed below under Other Resources.

Traditional methods of laboratory analysis to detect exact concentrations of aflatoxins include extensive chemical extraction processes, followed by thin layer chromatography (TLC) or high-pressured liquid chromatography (HPLC) techniques. However, these methods are often lengthy, costly, and complicated.

The use of feed contaminated by aflatoxins carries risks and producers should consider the consequences described above. Remember that young fish are more susceptible than adults. If it is suspected that the feed may be contaminated by mold, it is best to separate or discard contaminated feed and avoid feeding it to fry, breeders or replacement fish.

A possible remedy for feeds that do contain small amounts of mold is to add a toxin inactivator to the stored feeds. Nutriad International (see Web address listed under Other Resources) is one of many companies that specializes in feed additives.

A toxin inactivator such as Toxy-nil® controls mold development by binding with aflatoxins in the feed, making them inactive and preventing further growth of the mold in feeds. If all the feed is heavily contaminated, new feed should be purchased. Increased awareness and constant monitoring for aflatoxins in the feed will reduce the risk of aflatoxicity in fish.

Regulatory Control of Aflatoxin Levels in Fish Feeds

Due to their frequent occurrence, especially in the southeastern USA, agricultural commodities are routinely monitored for the presence of aflatoxins. The FDA enforces strict regulations for aflatoxin screening in oilseeds, corn and other feed ingredients. There are no official tolerances or safe levels for aflatoxins in fish feeds. The FDA, however, allows a maximum of 20 ppb in feeds or feed ingredients for other domestic animals.

Summary

Aflatoxins are poisons produced by naturally occurring molds. These molds can grow in grains and prepared feeds intended for fish production when

storage conditions are suboptimal: temperatures of 27°C (80°F) or warmer, and moisture at levels greater than 14%. These conditions are frequently seen in tropical and subtropical aquaculture.

To prevent aflatoxicosis, follow manufacturer's recommendations regarding shelf life and try to determine the feed manufacture date. Be aware of your supplier's storage practices. Avoid using feeds that appear discolored, lump together, and smell musty. Clean feed storage bins and automatic feeders regularly.

Aflatoxins lower production efficiency of cultured fish by reducing growth rates, impairing immunity, and, in some cases, causing mortality. Storing feed properly (in a cool, dry area on pallets and at least one foot away from any walls) can prevent unnecessary economic losses.

Other Resources

http://www.aquafeed.com

http://www.chinod.com/toxinbinder.html

http://www.neogen.com/FoodSafety/FS_NT_Index.html (follow Aflatoxin links)

http://www.vicam.com/products/afla-b.html

Ashley, L.M. 1970.Pathology of fish fed aflatoxins and other antimetabolites *In:* A symposium on diseases of fishes and shellfishes. American Fisheries Society Special Publication 5, 366-379.

Jantrarotai, W. and R.T. Lovell.1990. Subchronic toxicity of dietary aflatoxin B1 to channel catfish. Journal of Aquatic Animal Health 2: 248-254.

Tuan, N.A., J.M. Grizzle, R.T. Lovell, B.B. Manning, and G.E. Rottinghaus. 2002. Growth and hepatic lesions of Nile tilapia (*Oreochromis niloticus*) fed diets containing aflatoxin B1. Aquaculture 212: 311-319.