POLICY BRIEF

UNIVERSITY OF VETERINARY AND ANIMAL SCIENCES LAHORE

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Detection of aflatoxins in different food items using Enzyme Linked Immunosorbent Assay (ELISA)

AFLATOXINS: FOOD SAFETY AND SECURITY ISSUES FOR HUMANS AND LIVESTOCK

POLICY GUIDELINES FOR RESOLVING THE ISSUES

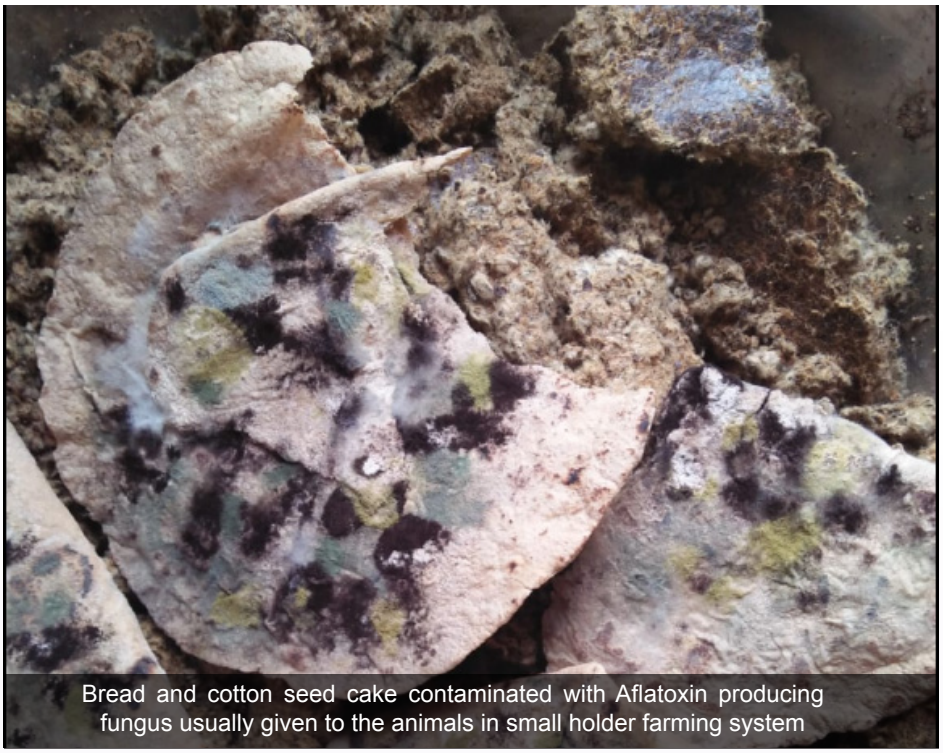
Policy Message

- A global issue, more prevalent in hot and humid regions like Pakistan.
- Every year 420,000 people die globally by consuming contaminated food.
- Global research shows that about 150,000 cases of liver cancer annually and 35% of child stunting is attributed to aflatoxins alone.
- Not properly placed on food security and safety agenda in Pakistan's health and livestock policy.
- As there is limited data available on aflatoxin contamination in various food and feed items, it is hard to estimate the extent of economic and health losses accompanied by consuming aflatoxin contaminated food and feed.
- Climate change is causing increase in heat and humidity in Pakistan with consequential rise in the prevalence of aflatoxin and a number of products used for human food and animal feed are at risk of contamination by aflatoxin
- Aflatoxin contamination leads to health and economic losses.
- Generally aflatoxins lead to chronic illnesses such as stunting, wasting and poor growth in children, various types of cancers, liver toxicity, and immunosuppression, however, they may cause acute health problems.
- Limited awareness among producers, consumers as well as medical professionals, unavailability of cost effective diagnostic tests, limited implementation of aflatoxin control regulations in Punjab and inadequate policy framework for aflatoxins control in other provinces are the main hurdles to tackle the problem.
- Pakistan Standards and Quality Control Authority (PSQCA) has developed maximum tolerable levels of aflatoxins for limited products, which also needs to be revised to meet the international standards

Aflatoxins are a family of toxins produced by fungi, mainly by *Aspergillus flavus* and *Aspergillus parasiticus* which grow on soil, decaying vegetation, hay, silage and stored food and feed. Aflatoxins have four different types, i.e. B1, B2, G1 and G2, and among these B1 is the most potent carcinogen as classified by the International Agency for Research on Cancer. These toxins or their metabolites which are present in animal products cannot be destroyed with cooking or boiling methods. Therefore, consumption of contaminated grains, meat, milk, eggs and fish leads to toxicity, thereby putting the human and animal population at risk and adversely affecting the economy of the country. Pakistan being hot and humid is more prone to aflatoxin contamination in food and feed; however, limited awareness and inadequate policy measures and their implementation are the major obstacles to the effective control of aflatoxicosis. This policy brief will discuss the prevalence of aflatoxin in food and feed chain, associated risk factors and adverse effects on health, needed regulatory measures, and control and management strategies to ensure food and feed safety.

Featured case study

Kenya had faced problem of aflatoxin contamination for many decades and went through great losses of human and livestock during the outbreaks of aflatoxin contamination. 2004 outbreak, in which 125 fatalities of humans occurred through consumption of contaminated maize, drew the attention of authorities and international communities, which in turn started initiative at public and private sectors. In order to develop a quality system approach for managing aflatoxins, Aflatoxin Task Force and National Food and Safety Coordinating Committee (NFSCC) were developed. The task force conducts surveillance and investigates outbreaks and NFSCC is responsible to ensure safety standards, inspection, analysis of samples, making and implementation of laws, education and communication, serves as a knowledge and information management platform accessible to all stakeholders. Public and private sector took initiatives such as training of relevant staff and farmers for aflatoxin management, development of a ware house system committed to drying and storage of grain at a fee, extensive awareness campaigns, providing moisture meter and aflatoxin testing equipment to farmers, use of nixtamalization as post-harvest intervention, evaluation of rapid diagnostic test and establishment of mycotoxin research facility. Furthermore, purchasing of grains after aflatoxin testing was promoted and the Safe Dairy Project was launched. After taking these initiatives incident cases of aflatoxicosis were significantly reduced.



Bread and cotton seed cake contaminated with Aflatoxin producing fungus usually given to the animals in small holder farming system

Aflatoxin prevalence in the food and feed supply chains

High temperatures and humidity favor the growth of molds that produce aflatoxins. Children are more prone to the toxicity than adults. In children and young stock, it is associated with stunted growth, delayed development, poor absorption of nutrients, immune system suppression, liver damage, and liver cancer. According to the Food and Drug Administration Authority (FDA), permissible levels for aflatoxin presence in food/feed is 20 to 300 ppb, with mono-gastric animals more susceptible than ruminants. In the pre-harvest period, the presence of aflatoxin-producing fungi (and then the production of the toxin) could be influenced and potentiated by different factors such as the plant genetics. During the growing and harvesting stages, toxin production and its evolution is predisposed by agricultural practices, including the wrong use of fungicides and pesticides, the use of open-pollinated varieties, weather conditions and climate during planting and growing and, finally, insect damage. Several studies from Pakistan have reported various levels of aflatoxin contamination in different food and feed commodities. Aflatoxin contamination in milk and milk products have been studied by many researchers in different regions of Pakistan and the authors concluded that this problem is present for years but could not acquire attention of the policy makers which may be due to inadequate research showing its adverse impact on health.

Factors contributing to the increased prevalence of aflatoxicosis in humans and livestock

Pakistan being located in subtropical zone has favorable conditions for the growth of molds. In addition, pre-harvest conditions of plant stress due to insect infestation and periods of drought, soil conditions like high organic content and soil moisture, use of inappropriate chemical agents and poor storage conditions are the important factors that produce a favorable environment for the growth of molds. Furthermore, many of the farmers add wasted pieces of breads contaminated heavily with molds as well as aflatoxins to animal feed which does not only affect the animal health but also contaminate the meat and milk of those animals. Above all, inadequate knowledge among general public especially the food/feed producers, people involved in supply chain and consumers about the aflatoxin contamination is one of the most important factor contributing towards the higher prevalence of aflatoxicosis in humans as well as animals. And those who have some knowledge about food contamination think that all the contaminants are destroyed when they cook food which is not true for aflatoxins. However, in the past, this issue could not gain enough attention from responsible authorities resulted in inadequate facilities in the laboratories – equipment/consumables as well as skilled laboratory workers – for the detection of aflatoxins. Aflatoxin testing is limited to the export quality products and the same is seldom applied for local consumers.

Health and economic losses associated with aflatoxin toxicity

Human health

Consumption of contaminated chicken meat, eggs, milk and milk products and aquaculture by humans causes acute or chronic toxicity. At high levels of exposure, aflatoxin causes acute onset of abdominal pain, vomiting, pulmonary edema and liver necrosis or even it can be fatal. Continued ingestion of aflatoxins contaminated food is now considered as the leading cause of liver cancer. Chronic exposure has led to increasing liver and kidney diseases, decreasing immunity and reproductive function and poor growth in children. If a pregnant mother is taking aflatoxin contaminated diet, this can adversely affect the growth of fetus as aflatoxins can cross the placental barrier. Similarly, a newborne can get exposed to aflatoxin M1 through breast milk resulting in stunting, wasting and/or underweight.

Animal health

In ruminants, aflatoxins have adverse ef-

fects not only on liver and kidney function, but they also cause decrease in feed intake and milk production, poor reproductive performance and growth impairment in young and lactating animals. In poultry, aflatoxins can produce increased mortality, adversely affect all important production parameters, impair uptake of essential nutrients, cause tissue damage and drop in egg production. Similar effects have been observed in fisheries. To the lesser extent, hunters usually use poor quality grains that are mostly contaminated with aflatoxins and may have adverse effects on wildlife and migratory birds.

Economic losses

The aflatoxin producing molds invade crops in the field and grow on stored food/feed ingredients which leads to direct economic losses to agriculture, livestock and fisheries sector. Aflatoxicosis is associated with reduced productivity, increased disease incidence, decreased healthy life years and increased diagnostic and healthcare costs for humans as well as animals. The productivity and nutritive value of aflatoxin contaminated crops also drop after contamination. Due to aflatoxin contamination many of the food products are not allowed to export to the developed countries and this directly affects the economy of the country.

Policy measures

In Pakistan, food safety standards are regulated by federal as well as provincial government. Federal government applies Food & Drug Administration (FDA-USA) and CAC standards on imported food products and is responsible for the Federal territory. PSQCA serves as central collaborating body for all national, international, regional organizations and institutions such as ISO, CAC and WTO while Provincial Food Authorities such as Punjab Food Authority regulate standards of food articles, their manufacturing, storage, distribution, sale and import in respective provinces. Despite of the active working of both organizations in the country, progression towards the aflatoxin control is still slow and there is a desperate need for setting maximum tolerable limit for aflatoxins in all food and feed commodities on national level. Also, there is a need to develop and implement laws for proper disposal of food items rejected for exports due to aflatoxin contamination. Moreover , there is a dearth of available literature and updated information regarding policy measures on aflatoxin contamination in Pakistan.

Methods to detect aflatoxins

Several methods are available for the detection of aflatoxins but two of them are more reliable and commonly used, i.e. High Performance Liquid Chromatography (HPLC) and Enzyme Linked Immunosorbent Assay (ELISA). Although, HPLC method provides a complete quantification of the toxin in question, this technique is expensive (approx. cost PKR 4,000/-per sample test) while ELISA analyzes sample qualitatively and semi quantitatively but is comparatively cost effective (PKR 2000/-per sample test). Blood samples can be tested for the presence of AFB-albumin adduct (which remains in blood for weeks to months) while urine samples (which can only be detected within 24 hours of exposure) can be used to check for the presence of AFM1.

Detoxification of aflatoxin contaminated food

Several methods have been developed for detoxification of aflatoxin contaminated food. Physical methods may include Thermal inactivation, UV irradiation, gamma irradiation, adsorption and solvent extraction. Chemical methods may include ammoniation, acid treatment, ozonation, chlorine treatment and oxidizing agents. Biological methods may include bacterial and yeast degradation, microbial enzymatic degradation and fermentation.

Effect of storage conditions on food contamination

Growth of mold in various food items is mainly affected by climatic factors – temperature and humidity, and moisture levels in the food item. The weather conditions in Pakistan are highly favorable for the growth of mold, therefore, lack of precautionary measures while storing various food items can lead to growth of mold and hence contamination with aflatoxins. On a large scale, grains can be stored in crop storage bags; a three layered bag that effectively reduce chances of oxygen entrance from the environment and escape of carbon dioxide from the bag which prevent mold growth and insect infestation. Metal or cement bins and treatment with ozone and gamma irradiation are also effective preventive measures. Household products should be stored in air tight containers to avoid air exposure of food. Containers should be used after proper cleaning and government should take measures to discourage the prolong storage of food items at household level.

Major hurdles to reduce aflatoxin contamination

One of the major problem is limited awareness about aflatoxin contamination of food and feed and its impact on health and economic losses. The main reason for very low level of awareness among general public and medical practitioners is that the aflatoxins, mostly, do not cause acute problems and therefore could not get attention of the food regulatory and health departments. Secondly, there is a deficiency in surveillance system and testing facilities due to high costs involved in the detection of aflatoxins. Thirdly, there is a scarcity of research based systematic work showing the adverse impacts of aflatoxin consumption on health leading to not getting enough attention by the higher authorities. Although childhood stunting and liver problems are very common in Pakistan for many years and aflatoxins presence in different food commodities have been reported in many studies, yet researchers have not tried to find out the association between these two. Lastly, absence of law enforcement by the responsible authorities against aflatoxin contamination has exacerbated this problem.

Policy implications

1—Aflatoxin Task Force should be made to develop a comprehensive integrated approach for the prevention and control of aflatoxin from field to table in One Health fashion involving the agriculture, livestock, health, trade, and environmental sectors.
2—Harmonized standards and regulatory measures should be applicable uniformly across the country. Hence, a federal level agency should be responsible to set standards while provincial bodies such as food

Table: Comparison of maximum permissible levels of aflatoxins in various food commodities as set by different organizations

Food	PSQCA	EFSA	FDA	CAC
Packaged liquid milk	0.5 ppb	0.05 ppb	0.5 ppb	0.5 ppb
Milk powder	10 ppb	0.05 ppb	0.5 ppb	0.5 ppb
Food grains & roasted nuts (all)	20 ppb	2-12 ppb	20 ppb	15 ppb
Animal feed	50 ppb	20 ppb	20 ppb	20 ppb

ppb = parts per billion
PSQCA=Pakistan Standards and Quality Control Authority; EFSA=European Food Safety Authority; FDA=Food and Drug Authority; CAC=Codex Alimentarius Commission

authorities of respective provinces should only ensure enforcement and compliance of the set standards so that inter provincial uniformity with regards to food production, processing, distribution and consumption comply with uniform standards.

3—Aflatoxin control policy should be included in national nutrition action plan while national nutrition survey should include aflatoxicosis detection.

4—Develop intensive surveillance programs through regulated inspection services for generating data on population exposure and aflatoxin's adverse effects.

5—Aflatoxin contamination is a neglected issue, therefore, extensive awareness campaigns should be launched among general public – producers as well as consumers – and health professionals about possible sources of aflatoxin contamination and its adverse effects on health.

6—Need of capacity building of field and laboratory staff for inspection, sampling and testing for detection of aflatoxins.

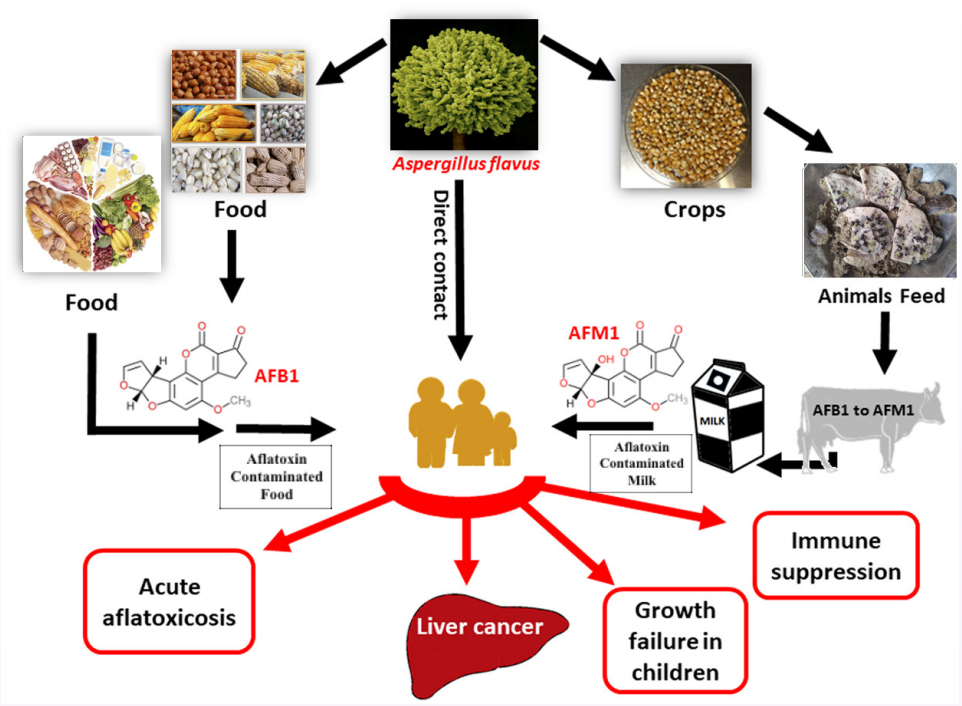
7—Develop policies and procedures for pre-harvest, post-harvest and storage stages of crop management such as strict monitoring of cataloging operations, application of functional bin systems and improved store management practices.

8—Detoxification methods should be available to the producers in order to reduce losses of commodities.

9—Special emphasis should be given to develop indigenous technologies for cost effective detoxification methods through provision of technology development funds etc.

10—Packaged food products should be encouraged and sale of loose milk should be baned. Moreover, aflatoxin level should be mentioned in the product label.

11—The regulatory bodies should update their websites regularly and ensure the availability of information and policy guidelines to general public.



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Further Reading

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Explained Terminologies:

Aflatoxicosis: Poisoning caused by the consumption of aflatoxin.
Atoxigenic: Which does not produce toxins.
Chronic: condition which persists for a long period.
Incidence: The number of new cases of a particular disease in a specified population during a specified time period.
Nixtamalization: A process for the preparation of maize in which the corn is soaked and cooked in an alkaline solution washed, and then hulled. This process is known to remove up to 97–100% of aflatoxins from mycotoxin-contaminated corn
Stunted: Prevention from growing or developing properly.
Tolerable limit: The highest level of nutrient intake that is likely to pose no risk of adverse health effects for almost all individuals in the general population.

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