

## Aflatoxin control and prevention strategies in maize for Sub-Saharan Africa

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### Abstract

Mycotoxins are secondary fungal metabolites that contaminate agricultural commodities and can cause sickness or death in humans and animals. Risk of mycotoxin contamination of food and feed in Africa is increased due to environmental, agronomic and socio-economic factors. Environmental conditions especially high humidity and temperature favour fungal proliferation. Farming practices in Africa sustain fungal and toxin contamination of food and feed. The socio-economic and food security status of the majority of inhabitants of sub-Saharan Africa leaves them little option in choosing good quality products.

Several technologies have been tested in Africa to reduce mycotoxin risk. Field management practices that increase yields may also prevent aflatoxin. They include use of resistant varieties, timely planting, fertilizer application, weed control, insect control and avoiding drought and nutritional stress. Other options to control the toxin causing fungi *A. flavus* contamination in the field are use of atoxigenic fungi to competitively displace toxigenic fungi, and timely harvest. Post-harvest interventions that reduce mycotoxins are rapid and proper drying, sorting, cleaning, drying, smoking, post harvest insect control, and the use of botanicals or synthetic pesticides as storage protectant. Another approach is to reduce the frequent consumption of 'high risk' foods (especially maize and groundnut) by consuming a more varied diet, and diversifying into less risky staples like sorghum and millet. Chemo-preventive measures that can reduce mycotoxin effect include daily consumption of chlorophyllin or oltipraz and by incorporating hydrated sodium calcium aluminosilicates into the diet. Detoxification of aflatoxins is often achieved physically (sorting, physical segregation, flotation etc.), chemically (with calcium hydroxide, ammonia) and microbiologically by incorporating pro-biotics or lactic acid bacteria into the diet. There is need for efficient monitoring and surveillance with cost-effective sampling and analytical methods. Sustaining public education and awareness can help to reduce aflatoxin contamination.

Keywords: Aflatoxin, Sub-Saharan Africa, Control measures

### 1. Introduction

Aflatoxins are secondary metabolites primarily produced by the fungi *Aspergillus flavus* Link, *A. parasiticus* Speare and to a lesser extent *A. nomius* Kurtzman et al. (CAST, 2003). Optimal conditions for fungal development are 36 to 38°C, with a high humidity of above 85% (Diener et al., 1987). Suitable conditions for the growth of the fungi and toxin production occur in most areas of Africa and aflatoxin contamination of food is a widespread problem across the continent, which has been reviewed by several authors (Sibanda et al., 1997; Shephard, 2003; Bankole and Adebajo, 2003; Bankole et al., 2006; Wagacha and Muthomi, 2008). African communities and populations are exposed to aflatoxins before birth and throughout their lives with serious impact on their health (Williams et al., 2003).

Aflatoxins are the most potent natural carcinogenic substance and they have been linked with a higher prevalence of hepatocellular cancer in Africa (Strosnider et al., 2006). There is a very high risk of Hepatitis B and Hepatitis C carriers to develop liver cancer when they are exposed to aflatoxin (Williams et al., 2003). There have been recent outbreaks of acute aflatoxicosis in Kenya (Probst et al., 2007), but chronic exposure to aflatoxins has much wider health effects than these rare acute poisonings (Williams et al., 2004). Aflatoxins have been linked to immune suppression by Turner et al. (2005) and Jiang et al. (2005). Children in areas of high aflatoxin exposure have been found to have stunted growth (Gong et al., 2004). Aflatoxin contamination has been linked to micronutrient deficiencies in animals (Williams et al., 2004), but Gong et al. (2004) reported that there was no relationship between aflatoxin-albumin, the biomarker of aflatoxin exposure, and micronutrients.

Aflatoxin contamination in several foodstuffs in Africa has been a recurrent problem (Shephard, 2003). In many parts, maize has become the preferred cereal for food, feed and industrial use, displacing traditional cereals such as sorghum and millets. However, it was significantly more heavily colonized by aflatoxin-producing *Aspergillus* spp. than either sorghum or millet (Bandyopadhyay et al., 2007). This review paper outlines some of the potential solutions to controlling toxins in Africa that are being developed by researchers either within or from outside Africa. Some of the potential solutions to controlling mycotoxins in Africa that are being developed by researchers are presented. These strategies can be broadly divided into: stopping the infection process (host plant resistance, biocontrol); control of environmental factors (temperature, rainfall, relative humidity, evapotranspiration, soil type) including efforts to build predictive models; crop management strategies (good agricultural practices (GAP), pre- and post-harvest management); post-harvest strategies (harvesting, drying, storage, use of plant extracts and preservatives) and decontamination (sorting, processing).

## 2. First strategy: stopping the infection process

### 2.1. Breeding for resistance

Several screening tools have been developed and used to facilitate corn breeding for developing germplasm resistant to fungal growth and/or aflatoxin contamination (Brown et al., 2003). Sources of resistance to *Aspergillus* infection and aflatoxin contamination in corn have been identified, but commercial hybrids have not been developed. This is largely due to the difficulty in finding elite lines that maintain high yields and maintain resistance within multiple environments (Clemons and White, 2004). Brown et al. (2001) tested aflatoxin resistance in thirty-six maize inbred lines selected in West and Central Africa for moderate to high resistance to maize ear rot for their resistance to aflatoxins, more than half the inbred accumulated aflatoxins at levels as low as or lower than the resistant U.S. lines. In 2008, six tropical maize germplasm lines with resistance to aflatoxin were registered by the same research group (Menkir et al., 2008) and their distribution to national programs will start soon for the development of locally adapted hybrids.

Many new strategies that enhance host plant resistance against aflatoxin involving biotechnologies are being explored and are reviewed by Brown et al. (2003). These approaches involve the design and production of maize plants that reduce the incidence of fungal infection, restrict the growth of toxigenic fungi or prevent toxin accumulation. In the long term the identification of compounds that block aflatoxin biosynthesis would significantly enhance mycotoxin control.

### 2.2. Biological control

Another potential means for toxin control is the biocontrol of fungal growth in the field. Numerous organisms have been tested for biological control of aflatoxin contamination including bacteria, yeasts, and nontoxigenic (atoxicogenic) strains of the causal organisms (Yan et al., 2008) of which only atoxicogenic strains have reached the commercial stage. Biological control of aflatoxin production in crops in the US has been approved by Environmental Protection Agency and a commercial product based on atoxicogenic *Aspergillus flavus* strains is being marketed (Afla-Guard®). In Africa, two isolates of *A. flavus* have been identified as atoxicogenic strains to competitively exclude toxigenic fungi in the maize fields. These strains have been shown to reduce aflatoxin concentrations in both laboratory and field trials, reducing toxin contamination by 70 to 99% (Atehnkeng et al., 2008b). A mixture of four atoxicogenic strains of *A. flavus* of Nigerian origin has gained provisional registration (AflaSafe) to determine efficacy in on-farm tests and candidate strains have been selected for Kenya and Senegal.

## 3. Second strategy: control of environmental factors

To design strategies for the prevention or reduction of aflatoxins, a thorough understanding of the factors that influence the infection of the plant with the aflatoxin causing fungi and the conditions that induce their formation is required. Environmental factors that favor *A. flavus* infection in the field include high soil and/or air temperature, high relative humidity, high rates of evapotranspiration, reduced water availability, drought stress, nitrogen stress, crowding of plants and conditions that aid the dispersal of conidia during silking (CAST, 2003). Some of these factors have been included in a model to predict toxin contamination in peanut systems in Mali. Weather and satellite based variables that could be used to indicate aflatoxin presence in peanut were identified (Boken et al., 2008).

Significant correlations exist between Agroecozones (AEZ) and aflatoxin levels, with wet and humid climates and drier regions after longer storage periods increasing aflatoxin risk (Hell et al., 2000). Agroecozones are geographic areas that share similar biophysical characteristics for crop production, such as soil, landscape, and climate. Kaaya et al. (2006) observed that aflatoxin levels in Ugandan maize samples were higher in more humid areas compared to the drier areas and similar results were obtained in maize samples from Nigeria (Atehnkeng et al., 2008a); these trends could be used to elaborate predictive models. Modelling of interactions between host plant and environment during the season can enable quantification of preharvest aflatoxin risk and its potential management (Boken et al., 2008; Chauhan et al. 2008). Predictive growth models for fungal and mycotoxin developments are available and have been reviewed by Garcia et al. (2009).

Factors that influence the incidence of fungal infection and subsequent toxin development include invertebrate vectors, grain damage, oxygen and carbon dioxide levels, inoculum load, substrate composition, fungal infection levels, prevalence of toxigenic strains and microbiological interactions. Insects vector fungi and cause damage that allows the fungi to gain access, increasing the chances of aflatoxin contamination, especially when loose-husked maize hybrids are used (Dowd, 2003). High incidence of the insect borer *Mussidia nigrivenella* Ragonot, was positively correlated with aflatoxin contamination of maize in Benin (Setamou et al., 1998). Storage pests, in particular *Cathartus quadricollis* Guerin and *Sitophilus zeamais* Motschulsky, play an important role in the contamination of foods with fungi, especially those that produce toxins (Hell et al., 2003; Lamboni and Hell, 2009).

#### 4. Third strategy: crop management strategies

Controlling or reducing infection and regulating the factors that increase the risk of contamination in the field for maize will go a long way in controlling aflatoxins. Management practices that reduce the incidence of mycotoxin contamination in the field include timely planting, optimal plant densities, proper plant nutrition, avoiding drought stress, controlling other plant pathogens, weeds and insect pests and proper harvesting (Bruns, 2003). In Africa, crops are cultivated under rain fed condition, with low levels of fertilizer and practically no pesticide application. These management practices promote *A. flavus* infection in fertility stressed plant. Any action taken to interrupt the probability of silk and kernel infection will reduce aflatoxin contamination (Diener et al., 1987).

Pre-harvest measures that are efficient in reducing aflatoxin levels are the same as those that will enhance yields. Crop rotation and management of crop residues also are important in controlling *A. flavus* infection in the field.

Tillage practices, crop rotation, fertilizer application, weed control, late season rainfall, irrigation, wind and pest vectors all can affect the source and level of fungal inoculum, maintaining the disease cycle in maize (Diener et al., 1987).

##### 4.1. Timely harvesting

Extended field drying of maize could result in serious grain losses during storage (Borgemeister et al., 1998; Kaaya et al., 2006), and as such harvesting immediately after physiological maturity is recommended to combat aflatoxin problems. Kaaya et al. (2006) observed that aflatoxin levels increased by about 4 times by the third week and more than 7 times when maize harvest was delayed for 4 wk. However, after early harvesting products have to be dried to safe levels to stop fungal growth. Leaving the harvested crop in the field prior to storage promotes fungal infection and insect infestation, this is common practice in Africa often due to labour constraints, and the need to let the crop dry completely prior to harvest (Udoh et al., 2000).

##### 4.2. Rapid drying

Moisture and temperature influence the growth of toxigenic fungi in stored commodities. Aflatoxin contamination can increase 10 fold in a 3-d period, when field harvested maize is stored with high moisture content (Hell et al., 2008). The general recommendation is that harvested commodities should be dried as quickly as possible to safe moisture levels of 10 – 13 % for cereals. Achieving this through simple sun-drying under the high humidity conditions of many parts of Africa is difficult. Even, when drying is done in the dry season, it is not completed before loading grains into stores like observed by Mestre et al. (2004) and products can be easily contaminated with aflatoxins. There are several

technologies to increase the efficacy of grain drying and reduce the risk of toxin contamination even under low-input conditions; these are the use of drying platforms, drying outside the field, drying on mats (Hell et al., 2008). Farmers should be able to determine the actual moisture content of their products. A simple moisture meter has been developed by IRRI using standard electronic components and can be produced locally:

([http://www.irri.org/irrc/streams/farmer friendly moisture meter.asp](http://www.irri.org/irrc/streams/farmer_friendly_moisture_meter.asp)).

There are technological solutions that could aid in reducing grain moisture rapidly, which are reviewed by Lutfy et al. (2008). However, these dryers are not used by African farmers because large capital investments are needed to acquire them. However, Gummert et al. (2009) described the very positive effect dryers had on maintaining rice quality and reducing mycotoxin risk in Southeast Asia.

#### *5. Fourth strategy: post harvest crop management practices*

Aflatoxin contamination of foods increases with storage period (Kaaya and Kyamuhangire, 2006). It is compounded in Africa through excessive heat, high humidity, lack of aeration in the stores, and insect and rodent damage resulting in the proliferation and spread of fungal spores. Thus strategies to minimize quantitative and qualitative post harvest losses have been developed (Hell et al., 2008). These improved postharvest technologies have been used successfully to reduce the blood aflatoxin-adducts level in populations in Guinea, where exposure was more than halved 5 mo after harvest in individuals from the intervention villages (Turner et al., 2005).

Traditional storage methods in Africa can be divided into two types, namely temporary storage that is mainly used to dry the crop and permanent storage that takes place in the field or on the farm. The latter includes containers made from plant materials (woods, bamboo, thatch) or mud placed on raised platforms and covered with thatch or metal roofing sheet. The stores are constructed to prevent insect and rodent infestation and to prevent moisture from getting into the grains. It is difficult to promote new storage technologies, such as the use of metal or cement bins, to small-scale farmers due to their high cost. Many farmers nowadays store their grains in bags, especially polypropylene which are not airtight, but there is evidence that this method facilitates fungal contamination and aflatoxin development (Hell et al., 2000; Udoh et al., 2000). Presently there are efforts to market improved hermetic storage bags in Africa, based on triple bagging developed for cowpea (Murdock et al., 1997) which has been or is being tested for other commodities (Ben et al., 2009).

#### *5.1. Disinfestation methods*

Smoking is an efficient method of reducing moisture content and protecting maize against infestation by fungi. The efficacy of smoking in protecting against insect infestation was found to be high. About 4 to 12% of farmers in the various ecological zones in Nigeria used smoke to preserve their grains, and this practice was found to be correlated with lower aflatoxin levels in farmers' stores (Udoh et al., 2000). Other compounds used for seed fumigation like ethylene oxide and methyl bromide were found to significantly reduce the incidence of fungi including toxigenic species on stored groundnuts and melon seeds (Bankole et al., 1996). Among the chemical compounds tested in feeds, propionic acid, sodium propionate, benzoic acid and ammonia were the best anti-fungal compounds, followed by urea and citric acid (Gowda et al., 2004). Farmers use local plant products for controlling insect infestation, past studies have looked at the use of these substances for the control of fungi mostly proving their efficacy in-vitro (Hsieh et al., 2001), but these products have not proven their efficiency in farmers stores. There is need to review the efficacy of the multiple products used by farmers and tested by researcher to get a complete picture about their potential in reducing toxin contamination. Use of pesticides to control mycotoxins and their efficacy, have been reviewed by D'Mello et al. (1998), but their use by farmers in Africa is not always well practiced and deaths due to pesticide use have been reported. Extension workers should educate farmers on the importance of using recommended chemicals for specific crops at appropriate concentrations and within a safe delay before consumption.

#### *5.2. Physical separation and hygiene*

Aflatoxin is unevenly distributed in a seed lot and may be concentrated in a very small percentage of the product (Whitaker, 2003). Sorting out of physically damaged and infected grains (known from colorations, odd shapes and size) from the intact commodity can result in 40-80% reduction in aflatoxins

levels (Park, 2002; Fandohan et al., 2005; Afolabi et al., 2006). The advantage of this method is that it reduces toxin concentrations to safe levels without the production of toxin degradation products or any reduction in the nutritional value of the food. This could be done manually or by using electronic sorters. Clearing the remains of previous harvests and destroying infested crop residues are basic sanitary measures that are also effective against storage deterioration. Cleaning of stores before loading in the new harvests was correlated to reduced aflatoxin levels (Hell et al., 2008). Separating heavily damaged ears i.e. those having greater than 10% ear damage also reduces aflatoxin levels in maize (Setamou et al., 1998). Wild hosts, which constitute a major source of infestation for storage pests, should be removed from the vicinity of stores (Hell et al., 2008).

### *5.3. Reduction through food processing procedures*

Sorting can remove a major part of aflatoxin contaminated units, but levels of mycotoxins in contaminated commodities may also be reduced through food processing procedures that may involve processes such as sorting, washing, wet and dry milling, grain cleaning, dehulling, roasting, baking, frying, nixtamalization and extrusion cooking. These methods and their impact on mycotoxin reduction have been reviewed by Fandohan et al. (2008). The effect of extrusion cooking on mycotoxins in cereals was reviewed by Castells et al. (2005). Dehulling maize grain can reduce aflatoxin contamination by 92% (Siwela et al., 2005). The effect of nixtamalization in reducing aflatoxin contamination (Park, 2002) has lately been questioned with Méndez-Albores et al. (2004) reporting that nixtamalization is reversible.

Fermentation can increase the safety of some food products contaminated with mycotoxins. However, the available reports are contradictory, with some showing very efficient reductions in mycotoxins associated with fermentation, whereas others find lesser or no effects. Fandohan et al. (2005) found that processing maize into makume (a solid state fermented maize based product) resulted in 93% reduction of aflatoxin, while reduction levels were 40% for 'owo' which is a non-fermented dry milled maize porridge. The authors identified sorting, winnowing, washing, crushing combined with dehulling of maize grains as the critical mycotoxin reducing steps in the production chain, while fermentation and cooking appeared to have insignificant effect. There are diverse processing methods for the highly predisposed commodities (maize and groundnuts) in different parts of Africa and investigations of the effect of these processing techniques will identify those methods that expose consumers to less aflatoxins.

### *5.4. Other strategies*

The other strategies to reduce the risk of aflatoxin ingestion in Africa are dietary change, chemoprevention, detoxification and vaccination against hepatitis B would significantly reduce liver cancer risk (Strosnider et al., 2006).

### *6. Conclusions - perspectives of aflatoxin research in Africa*

It is clear that aflatoxin contamination in agricultural crops is widespread in Africa, but food insecurity together with drought are a major obstacle to improvements in food safety. Increased pressure on limited food resources and undernutrition exacerbates the mycotoxin problem by increasing the likelihood of human consumption of contaminated foods and by rendering the population more susceptible to the consequent adverse health effects. Even though considerable research efforts have been made to control toxin contamination, there are several factors that lead to high aflatoxin risk in Africa:

- 1) Lack of political commitment to mycotoxin research,
- 2) Shortage of trained personnel especially for mycotoxin monitoring,
- 3) Limited awareness on risks at all levels and insufficient knowledge on options to reduce aflatoxin contamination in production to consumption chain.

The perspectives of aflatoxin research in Africa can be foreseen as follows:

- i. Getting policy makers in the sub-region to recognise that the stimulation of the postharvest sector is an important avenue to increase food production and ensure food safety for the protection of the health of their citizens.
- ii. Educating stakeholders on the danger of commercializing and consuming mouldy foods.
- iii. Training personnel at all levels (scientists, technicians, extension agents) in sampling protocol and modern methods of mycotoxin analysis.
- iv. Conducting food baskets surveys for aflatoxin contamination using uniform sampling protocols and modern analytical methods to obtain sound and reliable data on aflatoxin incidence in different food crops, which could then be used to define control strategies.
- v. There should be a co-ordinated and collaborative effort on aflatoxin research in Africa to minimize repetitions so that resources can be focused on identified priority areas, including documenting the impact of aflatoxin on health and economies in Africa.
- vi. Investigation should focus on the effect of different pre- and post-harvest crop management systems on aflatoxin contamination in different agro-ecologies in Africa and the effect of different traditional food processing methods on aflatoxin production so that technologies that result in a significant reduction in aflatoxin levels could be promoted.

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