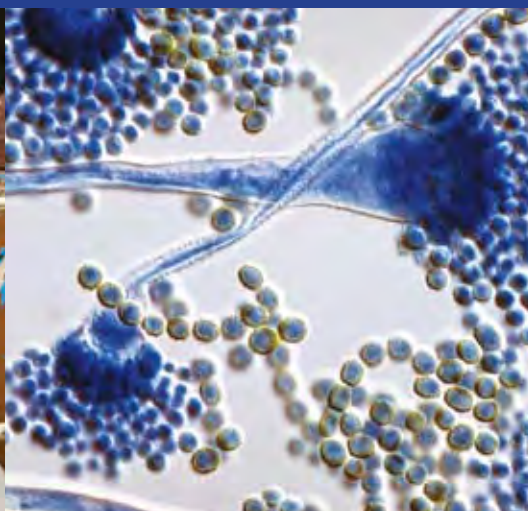


# AFLATOXIN: A SYNTHESIS OF THE RESEARCH IN HEALTH, AGRICULTURE AND TRADE





# TABLE OF CONTENTS

<b>Foreword</b>	5
<b>Acronyms</b>	6
<b>1. Introduction</b>	7
A. Background	7
B. Purpose of the Literature Review	7
C. Overview of the Literature Review	7
<b>2. Health</b>	9
Summary	9
A. Introduction	10
B. Health Consequences of Aflatoxin	11
B.1 Acute and Chronic Aflatoxicosis	12
B.2 Liver Cancer	13
B.3 Liver Cirrhosis	15
B.4 Immune System and Links with Stunting	15
B.5 Links to HIV and TB	18
C. Impact of Aflatoxin in Africa	19
C.1 Impact in East Africa	19
C.2 Impact in Southern Africa	22
C.3 Impact in West and Central Africa	23
D. Health Sector Capacity to Respond	25
D.1 Diagnosis and Treatment	25
D.2 Prevention and Control	25
D.3 Epidemiological Surveillance Systems	27
D.4 Policies and Strategies	28
D.5 Using Evidence from Research	29
E. Endnotes	30
F. References	37
<b>3. Agriculture</b>	43
Summary	43
A. Introduction	43
B. Prevention Through Pre-harvest Handling	44
B.1 Biocontrols	44
B.2 Competitive Displacement	45
B.3 Farming Techniques	46
B.4 Plant Breeding for Resistance	47
C. Prevention Through Post-harvest Handling	49
C.1 Processing	49
C.2 Storage Strategies	50
C.3 Implementation Package	53
D. Animal-Feeding Practices	54
D.1 Contamination Levels in By-products	54
D.2 Effects on Livestock	55
E. Handling Contaminated Commodities	57
E.1 Treatment	57
E.2 Alternative Uses	58
F. Endnotes	59
G. References	63

<b>4. Trade</b>	67
Summary	67
A. Introduction	68
B. Genesis of Aflatoxin Standards	68
B.1 Multinational Standard Setting	68
B.2 European Harmonization	69
C. Aflatoxin Standards for Humans and Animals	70
C.1 Human Consumption	70
C.2 Animal Feed	70
D. Testing Procedures	71
E. Trade Barriers	72
E.1 Impact of Mycotoxins on Commodity Trade	72
E.2 Nontariff Barriers to Trade	73
F. Trade Flows	74
F.1 Imports and Exports	74
F.2 U.N. World Food Programme Purchases	74
G. Endnotes	75
H. References	77
<b>Glossary</b>	79



## FOREWORD

The availability of a safe and nutritious food supply comprises one of the cornerstones of food security across Africa. To support this, The Partnership for Aflatoxin Control in Africa (PACA) will launch a comprehensive program to formulate policies, identify solutions, and support the implementation of programs to address health, agriculture, and trade issues related to aflatoxin contamination in the staple food supply. At this time, aflatoxin contamination is often a problem of unknown dimensions on farms, and in warehouses, processing facilities, and food products. What is known, however, is that the pervasive and chronic consumption of aflatoxin-contaminated foods and feeds throughout Africa continues to pose a significant threat to both human and animal health. Economic losses to producers and traders due to high levels of aflatoxin contamination in grains and legumes are also significant. The purpose of this paper is to provide a summary of published data on aflatoxin, with a focus on Africa and the East Africa region. This analysis lays the groundwork for informed policy and program development and identifies knowledge gaps for future research and planning efforts. The paper provides a shared understanding through which USAID and PACA will engage regional organizations, governments, donors, nongovernmental organization, and producers, traders, processors, and consumers in a constructive dialogue based on factual evidence. We thank the members of PACA and the donor community for their commitment to address this complex and challenging problem.

### *Feed the Future*

The Office of Regional Economic Integration  
USAID East Africa Regional Mission  
Nairobi, Kenya

## ACRONYMS

AGOA	African Growth and Opportunity Act
AIDS	Acquired Immune Deficiency Syndrome
AU	African Union
AUC	African Union Commission
BW	Body Weight
C	Celsius
CAADP	Comprehensive Africa Agriculture Development Programme
CD4	Cluster of Differentiation 4 (also known as T-cells)
CDC	Centers for Disease Control and Prevention
DNA	Deoxyribonucleic acid
ELISA	Enzyme-linked immunosorbent assay
EU	European Union
FEHD	Food and Environment Hygiene Department
FAO	Food and Agricultural Organization
HIV	Human Immunodeficiency Virus
IARC	International Agency for Research on Cancer
KBS	Kenya Bureau of Standards
Kg	Kilogram
MRC	Medical Research Council
MT	Metric Tonnage
NEPAD	New Partnership for Africa's Development
Ng	Nanogram
PACA	Partnership for Aflatoxin Control in Africa
SPS	Sanitary and Phytosanitary
TB	Tuberculosis
USAID	United States Agency for International Development
WFP	World Food Programme
WHO	World Health Organization

# 1

*“There is need for targeted monitoring and surveillance systems for aflatoxins in at-risk countries.”*

## INTRODUCTION

### BACKGROUND

Aflatoxin is a highly carcinogenic toxin produced by the fungus *Aspergillus flavus* (*A. flavus*). This fungus, as well as the toxins it produces, is commonly found in soils and on plant matter, including grains or cereals, peanuts, seeds, and other legumes. Aflatoxin poisoning in the East African region has become an epidemic, particularly in arid and semi-arid areas. Chronic aflatoxin exposure can have a negative impact on health and has been associated with liver cancer, growth retardation and stunting in children, and suppression of the immune system. It has also recently been linked with HIV and tuberculosis (TB). At high levels of concentration, aflatoxin exposure can cause hemorrhaging, edema, and even immediate death. In countries such as Kenya, documented cases of widespread aflatoxin poisoning are fast becoming a common occurrence, particularly in rural areas. Although research and limited interventions have been ongoing in countries such as The Gambia since the early 1940s, comparable efforts are lagging in many other areas of Africa. Further research into innovative solutions is necessary to address the often overlooked global issues of aflatoxin contamination and exposure.

### PURPOSE OF THE LITERATURE REVIEW

This literature review highlights the effects of aflatoxin contamination both globally and within various regions of Africa, with particular emphasis on Eastern Africa. This review, covering the key sectors of health, agriculture, and trade, serves as a resource for the Partnership for Aflatoxin Control in Africa (PACA) and provides a framework for addressing issues of aflatoxin in East Africa. Researchers at Danya International, Inc., with funding from the USAID East Africa Regional Mission, conducted this review, which is intended to inform the development of ongoing and future cross-sectoral aflatoxin initiatives, as well as the new 5-year East Africa *Feed the Future* strategy.

### OVERVIEW OF THE LITERATURE REVIEW

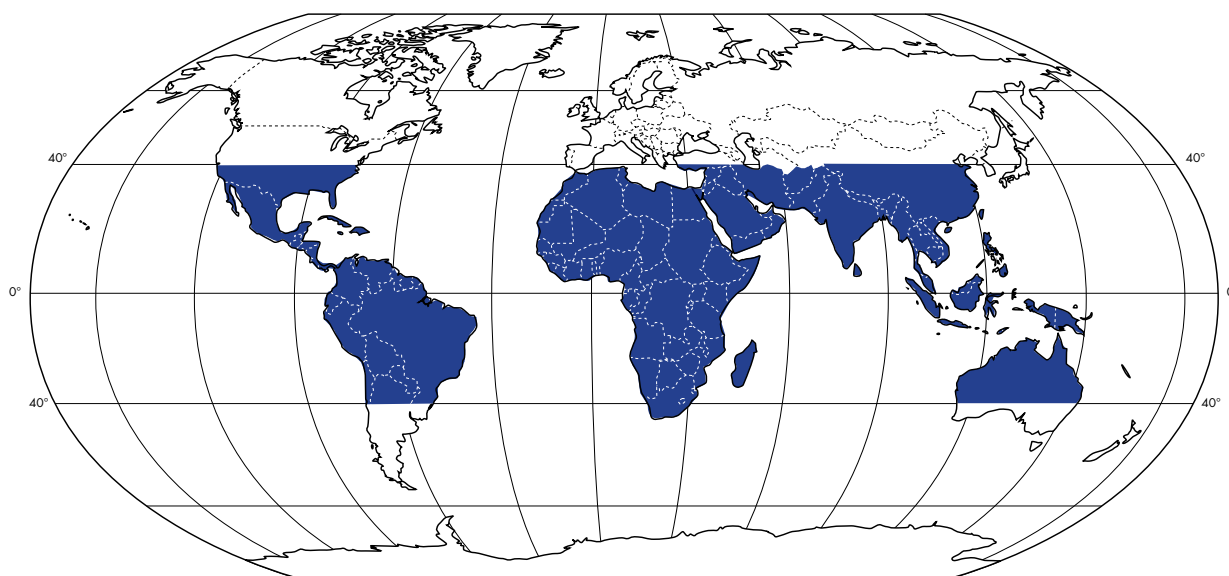
Since a multi-sectoral approach is required to prevent and control aflatoxin, this literature review, covering more than 100 major scientific research articles published since the year 2000, surveys the topic of aflatoxin contamination as it affects the health, agriculture, and trade sectors. Each section summarizes key issues, as well as comprehensively reviewing the latest published research.

The review, includes research and findings from more than 100 published articles from the last 10 years in health and physical sciences, microbiology, agricultural sciences, trade, and macroeconomics. Key issues have been identified and summarized. Relevant visual aids, such as tables, charts, and maps, have been included in the text. Key scientific terms have been defined and included in the glossary at the end of this review.

**Health.** Section 2 presents research findings on varying levels of aflatoxin exposure and the impact of that exposure on human growth and the immune system. The relationship of consumption patterns of at-risk foods to chronic serum aflatoxin levels within the body is also described. Incidence of acute and prevalence of chronic aflatoxin exposure throughout Africa are covered, with a particular emphasis on geographical epicenters. **Figure 1** highlights

global areas that are at risk of contamination. Interventions for diagnosis and treatment are also reviewed in this section, highlighting the need for targeted monitoring and surveillance systems in at-risk countries and regions.

Figure 1. Areas and Populations at Risk of Chronic Exposure to Uncontrolled Aflatoxin Contamination



Source: Williams et al., 2008

**Agriculture.** Section 3 outlines risks to the farming industry posed by both high and low levels of aflatoxin contamination. It reviews methods for pre- and post-harvest handling, including biocontrols, improved storage methods, and promising detoxification interventions. Statistics on the effects of aflatoxin exposure on livestock, poultry, and animal by-products are presented along with interventions to limit contamination in products for human consumption. Alternative uses for contaminated food products are also presented in this section.

**Trade.** Section 4 presents the impact that aflatoxin exposure, contamination, and regulations have on the trade of several agricultural products which include maize, nuts, and coffee beans. In addition, standards, guidelines, and regulations on aflatoxin limits across food categories, countries, and regions are reviewed. Research findings are presented on export trade flows as well as the impact of sanitary and phytosanitary (SPS) standards on trade. Stringent SPS standards of importing countries limiting aflatoxin levels have had significant impact on East African imports.



# 2

*“Aflatoxin, and the molds and fungi that produce it, is not visible in contaminated foods.”*

*“Humans are exposed to aflatoxin mainly through consumption of contaminated agricultural products or animal products such as meat, eggs, poultry, and milk.”*

## HEALTH

### Summary

Aflatoxin is a class 1 carcinogen naturally produced by fungi of the *Aspergillus* family, particularly *Aspergillus flavus* (*A. flavus*). The most toxic form, aflatoxin B<sub>1</sub>, is the most potent microbial carcinogen and is directly correlated to adverse health effects such as liver cancer and cirrhosis. Humans are exposed to aflatoxin by eating contaminated foods. Aflatoxin contamination is widespread throughout Africa as well as several other countries in Asia. Aflatoxin has been detected in grains, specifically maize, millet and sorghum, as well as peanuts, and animal products such as meat, eggs, poultry, and milk. Aflatoxin is also present in cassava and cotton seeds.

Exposure to aflatoxin leads to several health-related conditions including acute and chronic aflatoxicosis, aflatoxin-related immune suppression, liver cancer, liver cirrhosis, and nutrition-related problems such as stunted growth in children.<sup>1</sup> Exposure to aflatoxin may also compound pre-existing health concerns. Individuals infected with the hepatitis B virus who have been exposed to aflatoxin have 30 times the risk of getting liver cancer than people who are hepatitis B-negative.<sup>2</sup> Globally it is estimated that aflatoxin exposure contributes to between 4.6 percent and 28.2 percent of all liver cancer cases, most of which occur in Sub-Saharan Africa, Southeast Asia, and China, the regions with the highest aflatoxin exposure. Mitigation of the effects of aflatoxicosis on human liver disease can be achieved through three main interventions: clinical, dietary, and agricultural.

A broad range of signs and symptoms can be used to diagnose aflatoxicosis based on the level of exposure. The signs and symptoms of this condition include vomiting, abdominal pain and hemorrhaging, pulmonary edema, acute liver damage, loss of digestive tract function, convulsions, cerebral edema, and coma. Hepatitis B vaccinations, education through awareness campaigns, and chemoprevention measures such as competitive displacement, plant extract application, and methyleugenol spray (see Section 3) have proven to be effective interventions in controlling and preventing the adverse health effects of aflatoxin exposure.<sup>3</sup>

Aflatoxin constitutes a serious health concern to the entire food chain, necessitating a multidisciplinary approach to analysis, action, and solution. To maximize resources, a targeted monitoring and surveillance system for high-risk areas and their populations should collect and analyze appropriate specimens (usually food, urine, and serum).<sup>4</sup> According to Hell and Mutegi,<sup>5</sup> aflatoxin research in Africa is necessary to get policymakers in the Sub-Saharan region to recognize that the increased implementation of pre- and post-harvest interventions is important for increasing food security and ensuring food safety to protect the short and long term health of the population.

*“Exposure to aflatoxins leads to several health-related conditions.”*

## INTRODUCTION

Aflatoxin, a potent, naturally occurring microbial carcinogen, is produced primarily by *A. flavus* and *Aspergillus parasiticus* (*A. parasiticus*) and constitutes a group of approximately 20 related types of fungus. **Figure 2** shows yellow mold, caused by *A. flavus* and *A. parasiticus*, which commonly produces aflatoxin. Although the presence of other molds on foods can denote contamination, aflatoxin, and the molds and fungi that produce it, is not visible in contaminated foods.

There are four primary naturally produced aflatoxin strains known as B<sub>1</sub>, B<sub>2</sub>, G<sub>1</sub>, and G<sub>2</sub>. Two additional strains, M<sub>1</sub> and M<sub>2</sub> are the metabolic products of contaminated food or feed and are found in milk and other dairy products. The most toxic strain, aflatoxin B<sub>1</sub>, has been directly linked to adverse health effects such as liver cancer.

*Figure 2: Yellow Mold Caused by A. flavus and A. parasiticus*

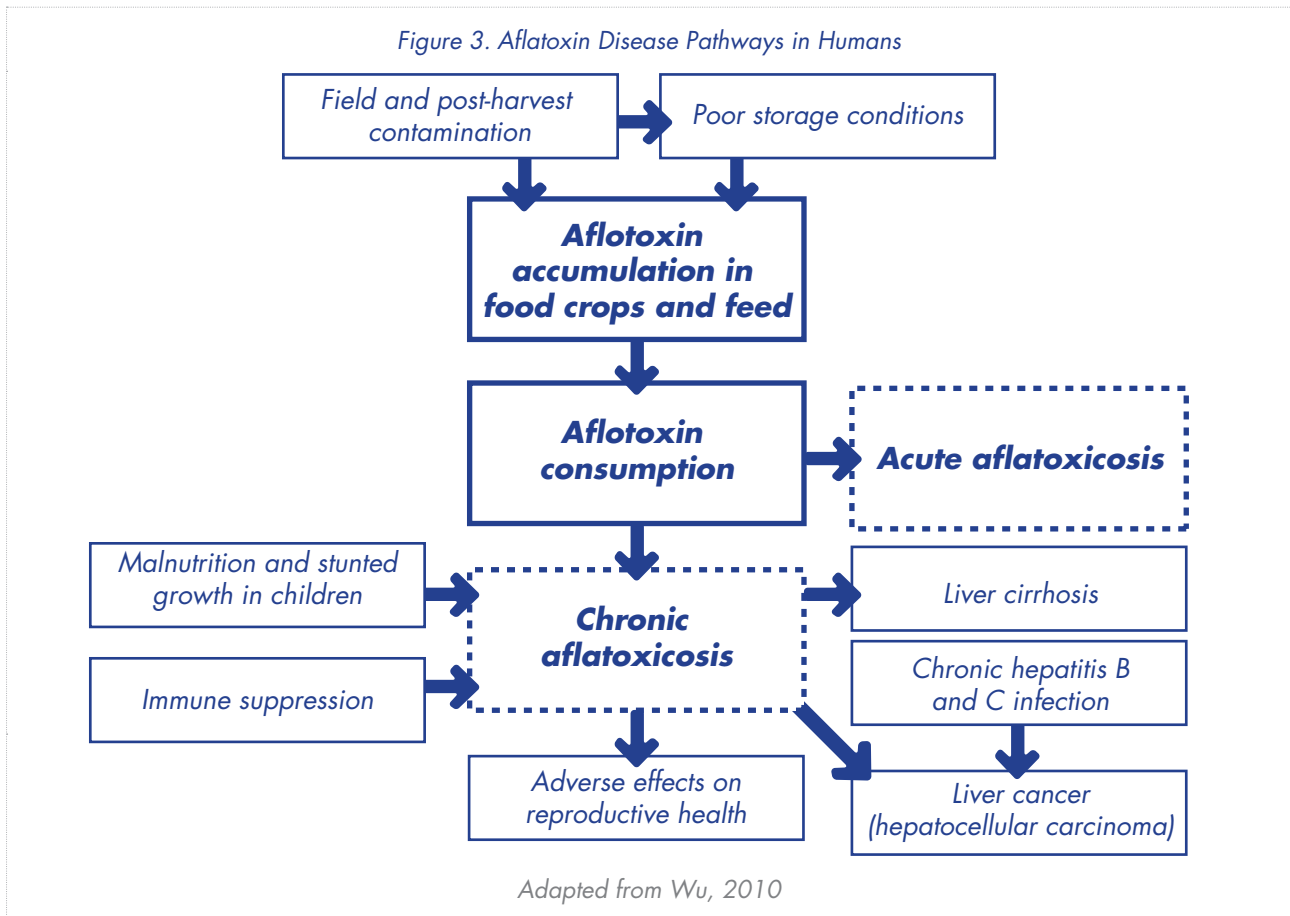


*Source: APSnet, Mycotoxins in Crops: A Threat to Human and Domestic Animal Health, 2011*

Humans are exposed to aflatoxin primarily through the consumption of contaminated agricultural or animal products. Other modes of exposure include the inhalation of toxins through occupational exposure.<sup>6</sup> Human exposure to aflatoxin has a negative impact on health. Exposure can lead to acute or chronic aflatoxicosis, based on the duration and amount of exposure, and can compound existing health issues or the risk of disease transmission.

## HEALTH CONSEQUENCES OF AFLATOXIN

Exposure to aflatoxin can lead to several health-related conditions including acute and chronic aflatoxicosis, aflatoxin-related immune suppression, liver cancer, liver cirrhosis, as well as nutrition-related problems in children such as stunted growth.<sup>7</sup> In many areas, due to widespread high-level consumption, aflatoxin contamination through food and feed is unavoidable due to the absence of alternative food and feed resources. When ingested, aflatoxin binds to liver proteins. The metabolic products may persist for 2 to 3 months or longer and can be detected through blood tests.<sup>8</sup> **Figure 3** demonstrates consequences of aflatoxin exposure on health.



Aflatoxin exposure can be measured in two ways: (1) an analysis of prepared foods or (2) through biological markers of exposure from blood or urine samples that are obtained and analyzed for the presence of aflatoxin derivatives. Possibilities to minimize biological exposure include (1) chemoprotection through the use of drugs and dietary supplements that detoxify aflatoxin and (2) enterosorptive food additives that bind to the toxin and render the aflatoxin biologically unavailable to the body.<sup>9</sup>

*“Chronic aflatoxicosis occurs more commonly than acute aflatoxicosis.”*

*“It is estimated that more than 5 billion people in developing countries worldwide are exposed to aflatoxin and are at risk of chronic exposure through contaminated food.”*

*“Acute aflatoxicosis is associated with extremely high doses of aflatoxin.”*

## **B.1 ACUTE AND CHRONIC AFLATOXICOSIS**

Aflatoxicosis is a disease caused by aflatoxin poisoning. The disease can be acute, meaning it is caused by the short-term exposure to high levels of aflatoxin, or chronic, meaning that it has been caused by long-term exposure to low to moderate levels of aflatoxin. Symptoms differ between the acute and chronic forms of the disease and have been outlined in this section.

### **Acute Aflatoxicosis**

Acute aflatoxicosis, associated with extremely high doses of aflatoxin, is characterized by hemorrhaging, acute liver damage, edema, and high mortality rates in humans. Acute aflatoxicosis is associated with sporadic outbreaks of the consumption of highly contaminated foods. Early symptoms of acute high-level exposure to aflatoxin include diminished appetite, malaise, and low fever; later symptoms, which include vomiting, abdominal pain, and hepatitis, can signal potentially fatal liver failure.<sup>10</sup> Acute aflatoxicosis in animals was first documented in 1960, after more than 100,000 turkeys died following an outbreak in the United Kingdom.<sup>11</sup> Kenya has experienced several recurrences of acute aflatoxicosis in humans and has recorded hundreds of deaths in the last 4 decades.<sup>12</sup>

### **Chronic Aflatoxicosis**

Chronic aflatoxicosis is associated with long-term exposure to low to moderate levels of aflatoxin in the food supply. It is estimated that more than 5 billion people in developing countries worldwide are at risk of chronic aflatoxin exposure through contaminated foods.<sup>13</sup> Chronic low-level exposure to aflatoxin, particularly aflatoxin B<sub>1</sub>, is associated with an increased risk of developing hepatocellular carcinoma, or liver cancer, as well as impaired immune function and malnutrition and stunted growth in children. Aflatoxin B<sub>1</sub> is the most potent liver carcinogen and is found in greater concentrations than any other naturally occurring aflatoxin.<sup>14</sup> According to the World Health Organization (WHO), hepatocellular carcinoma is the third leading cause of cancer deaths globally.<sup>15</sup> Approximately 83 percent of cancer fatalities in East Asia and Sub-Saharan Africa are due to liver cancer.<sup>16</sup>

Hepatocellular carcinoma, as a result of chronic aflatoxin exposure, presents most often in persons with a chronic hepatitis B virus and/or chronic hepatitis C virus infections.<sup>17</sup> This indicates that exposure to aflatoxin and hepatitis B infection, key risk factors for liver cancer, are particularly prevalent in developing nations in which people subsist largely on grains.<sup>18</sup>

Chronic aflatoxicosis also increases the risk of developing impaired immune function and malnutrition, a concern already prevalent in populations consuming high levels of cereals.<sup>19</sup> Cancer risk assessments and acute toxicity studies across species show that adult humans are relatively tolerant of aflatoxin; however, data reviewed in earlier sections indicate that there is evidence that aflatoxin exposure affects early development, as well as some aspects of human immunity and nutritional processes.<sup>20</sup>

Maize (*Zea mays* L.) and peanuts (*Arachis hypogaea* L.) form the staple food of many African and Asian diets. As these two crops are highly susceptible to aflatoxin infection, the incidence of aflatoxin exposure is closely related to the subsistence diet of populations in developing countries.<sup>21</sup> From 2001 to 2003, developing countries produced 46 percent of the global maize crop.<sup>22</sup> Poor harvesting and storage practices and weak regulations of mycotoxin contamination in developing countries exacerbates rates of aflatoxin exposure.<sup>23</sup>

*“Aflatoxin B1 is the most potent liver carcinogen known to man and it causes liver cancer.”*

According to a 1978 survey conducted in Ghana, 50 to 80 percent of peanuts were found to contain levels of aflatoxin in excess of recommended levels. In a more recent study, Gambian populations who subsist on a diet of peanuts and maize had some of the highest recorded levels of chronic exposure to aflatoxin.<sup>24</sup>

## **B.2 LIVER CANCER**

Aflatoxin B<sub>1</sub> is the most toxic of the aflatoxins and the strongest naturally occurring chemical liver carcinogen known. Aflatoxin, metabolized by enzymes in the liver, binds to proteins and causes acute toxicity (aflatoxicosis). Aflatoxin exposure causes acute liver damage and liver cirrhosis, as well as development of tumors or other genetic effects.<sup>25</sup> Liver cancer has increased in incidence and parallels chronic hepatitis B and hepatitis C infections. Studies have shown that persons with hepatitis B infection who live with chronic aflatoxin exposure have a risk of contracting liver cancer that is 30 times greater than people who are hepatitis B-negative.<sup>26</sup> **Figure 4** shows the higher incidence rates, per 100,000 people in 2008, of liver cancer in males and females from Kenya and Mali compared to North America and Europe.

*Figure 4. Liver Cancer Incidence Rates in Males and Females (per 100,000)*

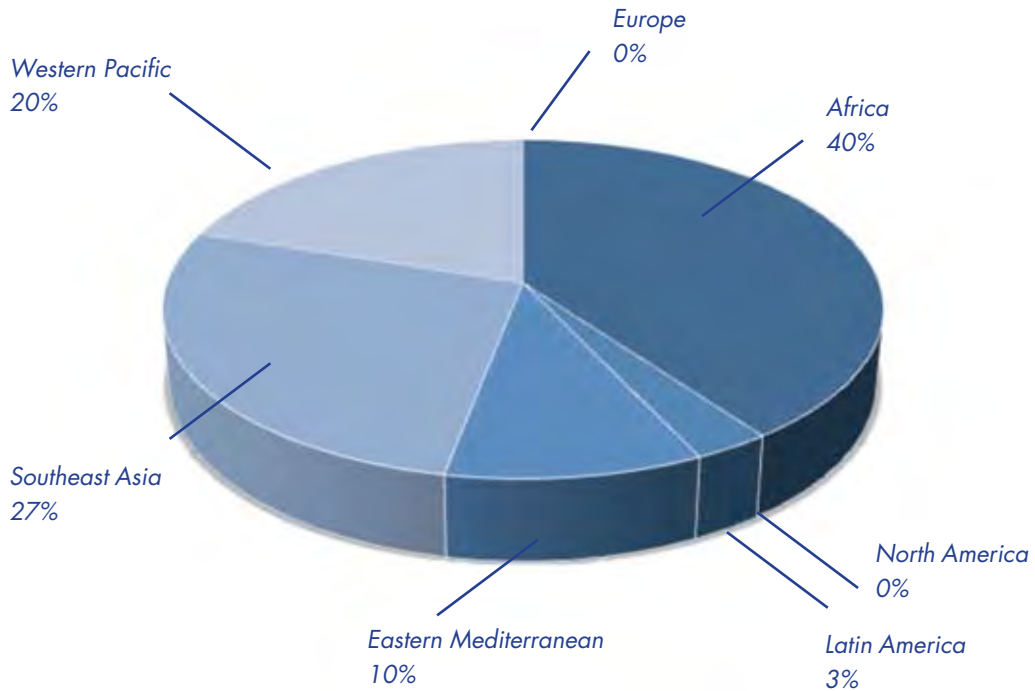
<b>Nation</b>	<b>Males</b>	<b>Females</b>
Kenya	8.5	4.9
Mali	19.4	8.8
North America	6.8	2.2
Europe	6.5	2.2

*Source: IARC GLOBOCAN, 2008*

Aflatoxins have also been known to have a related effect in inducing liver cancer in persons with hepatitis C.<sup>27</sup> Sub-Saharan African and Asian populations that have endemically high hepatitis B and hepatitis C rates are, therefore, likely to have a significantly increased disease burden for liver cancer.<sup>28</sup> Despite the relationship between hepatitis infection and liver cancer, the causal pathways of liver cancer are influenced by a variety of environmental and host factors that researchers do not yet fully understand. Further studies are needed to understand the mechanisms of aflatoxin exposure in conjunction with hepatitis B and hepatitis C.<sup>29</sup> In addition, other aflatoxin interactions are likely contributors to the disease burden, but still remain to be identified.<sup>30</sup>

Globally it is estimated that aflatoxin contributes to between 4.6 and 28.2 percent of liver cancer cases. Each year, 550,000–600,000 new liver cancer cases are recorded worldwide, and of these, approximately 25,200–155,000 are attributable to aflatoxin exposure. In 2008, liver cancer was the third leading cause of cancer-related deaths worldwide.<sup>31</sup> The global liver cancer burden is primarily borne by Sub-Saharan Africa, Southeast Asia, and Western Pacific nations, as shown in **Figure 5**.

Figure 5. Distribution of Hepatocellular Carcinoma Attributable to Aflatoxin



Source: Yu & Liu, 2010

The global disease burden of aflatoxin is influenced greatly by the geographic and temporal incidence patterns of liver cancer. **Figure 6** below depicts the correlation between high liver cancer rates and high risk of chronic exposure to aflatoxin.

Figure 6. Correlation Between High Liver Cancer Rates and High Risk of Chronic Exposure to Aflatoxin Contamination



Source: GLOBOCAN 2002 database and Williams et al., 2004



*“Globally it is estimated that aflatoxin contributes to between 4.6 and 28.2 percent of liver cancer cases.”*

*“...aflatoxin impairs growth and contributes to immune suppression...”*

Lack of reliable data is a key challenge to quantifying the magnitude of the economic and health consequences associated with the consumption of aflatoxin-contaminated food in developing countries. It is difficult to fully attribute the impact of aflatoxin-contamination on liver disease in developing countries, as liver disease can be masked by acute aflatoxicosis and can be misdiagnosed.<sup>32</sup>

There are three primary interventions to mitigate the effects of aflatoxicosis on human liver disease: clinical, dietary, and agricultural. One effective clinical intervention is administering the hepatitis B vaccination, particularly to children.<sup>33</sup> Complete elimination of aflatoxin is unlikely, and as a result, only proper management can mitigate the health effects globally.<sup>34</sup>

### **B.3 LIVER CIRRHOSIS**

The link between aflatoxin and liver cirrhosis is not as well documented as that with liver cancer. Some studies have suggested that the link between aflatoxin and liver cirrhosis is weak,<sup>35</sup> whereas other studies have indicated that there is sufficient evidence to associate aflatoxin with cirrhosis. A study on aflatoxin exposure and the cause of liver cirrhosis in The Gambia found that chronic hepatitis B infection and aflatoxin exposure—either separately or in synergy—were the agents most likely responsible for most cirrhosis cases in that West African population.<sup>36</sup>

### **B.4 IMMUNE SYSTEM AND LINKS WITH STUNTING**

Research shows that aflatoxin impairs growth and contributes to immune suppression in animals; however, immune suppression in humans has only recently been seriously investigated, mostly in children. A study in Benin and Togo found that stunted and/or underweight children had an average of 30 to 40 percent higher levels of aflatoxin-albumen levels in the blood than children with a normal body weight.<sup>37</sup> A study carried out in The Gambia by Turner et al. demonstrated that elevated aflatoxin-albumin levels were associated with stunting and underweight among children ages 6 to 9 years, the oldest cohort yet to show that linkage. Children in developing countries appear to be naturally exposed to aflatoxin through their diet at levels that compromise the immune system in other species. Immune functions associated with increased susceptibility to bacterial and parasitic infections have also been attributed to aflatoxin exposure.<sup>38</sup>

In general, the proportion of childhood growth stunting is directly correlated with the proportion of the population living below the national poverty line, and is inversely correlated with gross domestic product per capita.<sup>39</sup> As is the case with liver cancer, childhood stunting is prominent in regions such as Southeast Asia and Sub-Saharan Africa, where aflatoxin exposure through consuming contaminated food is common. **Figure 7** describes the correlation between socioeconomic characteristics, aflatoxin exposure, and the prevalence of stunting in 12 countries.

Figure 7. Aflatoxin Exposure, and Health and Economic Characteristics of Selected Nations

Country	% population living below national poverty line	GDP per capita, 2010 USD	Aflatoxin exposure, ng/kgBW/day	% stunted children*
France	6.2	34,250	0.3 – 1.3	NA
Spain	19.8	29,649	0.3 – 1.3	NA
USA	12	47,702	0.26	4
Argentina	30	15,030	0 – 4	8
Thailand	13	8,479	53 – 73	16
China	5	7,240	17 – 37	22
The Gambia	58	1,479	4 – 115	28
Philippines	37	3,604	44 – 54	34
Kenya	52	1,783	3.5 – 133	36
Nigeria	34	2,357	139 – 227	43
Tanzania	36	1,484	0.02 – 50	44
India	29	3,176	4 – 100	48

Note: GDP = gross domestic product; NA = not available or unreported; \*For the percentage of stunted children, this figure uses data for “children under the age of 5 years that are underweight”

Sources: Khlangwiset, 2011, *Indexmundi* [www.indexmundi.com/g/r.aspx?r=69](http://www.indexmundi.com/g/r.aspx?r=69)

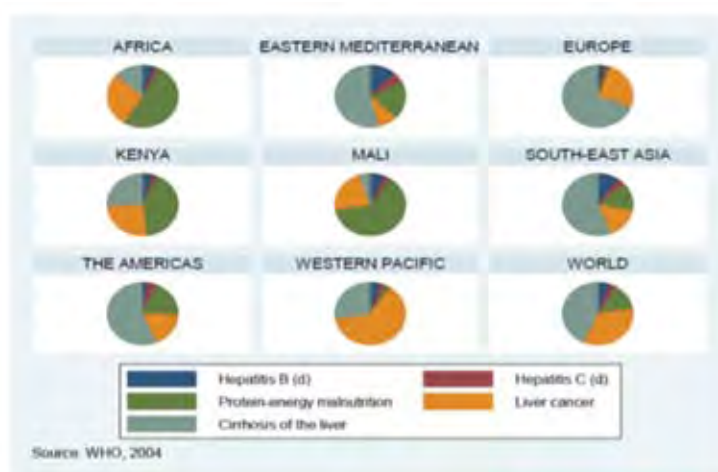
Aflatoxin has also been linked to kwashiorkor, a disease caused by protein-energy malnutrition. Kwashiorkor has some characteristics associated with the pathological effects caused by aflatoxin exposure in animals, but the link between aflatoxin exposure and kwashiorkor is not clear.<sup>40</sup> Despite these preliminary findings, the mechanisms by which aflatoxin affects growth are currently unknown and require further clarification.<sup>41</sup> The mortality rates from liver disease, which are high in Africa, especially in relation to protein-energy malnutrition, are shown in **Figure 8**.<sup>42</sup> The worldwide incidence of liver disease, including hepatitis B and C, protein-energy malnutrition, liver cirrhosis, and liver cancer, is shown in **Figure 9**.

Figure 8. Mortality Rates from Liver Disease per 100,000 Worldwide by Region and in Kenya and Mali

	Hepatitis B	Hepatitis C	Liver Cancer	Cirrhosis of Liver	Protein-Energy Malnutrition
<b>World</b>	1.63	0.84	9.47	11.99	3.89
<b>Africa</b>	1.64	0.72	8.19	3.85	15.09
<b>Americas</b>	0.57	0.92	4.16	12.87	4.46
<b>Europe</b>	0.83	0.52	7.33	20.94	0.54
<b>Southeast Asia</b>	2.21	0.82	3.50	12.57	3.30
<b>Western Pacific</b>	1.60	0.88	21.68	9.53	0.82
<b>Kenya</b>	0.50	0.22	2.58	2.47	4.02
<b>Mali</b>	4.99	2.24	16.91	3.82	47.93

Source: WHO, 2004

Figure 9. Incidence of Liver Disease per 100,000 Worldwide by Region and in Kenya and Mali



*“...a possible link between the HIV epidemic and aflatoxin poisoning...”*

*“...frequency of HIV transmission is positively associated with maize consumption in Africa.”*

## **B.5 LINKS TO HIV AND TB**

It has been suggested that the immunosuppression and nutritional effects of chronic aflatoxin exposure may be linked to the high prevalence of HIV in Southern Africa. This possible link, however, is not conclusive, as research targeting the cancer-causing effects of aflatoxin has generally overshadowed research focusing on nutrition and immunity.<sup>43</sup> Aflatoxin exposure has been shown to cause immune suppression, particularly in cell-mediated responses.<sup>44</sup>

The correlation between aflatoxin-albumin levels and CD4 counts in HIV positive individuals has recently been studied. CD4 interacts with cells that act as the gateway for HIV infection. CD4 proteins that have been weakened by aflatoxin exposure may correlate positively with HIV infection.<sup>45</sup> In addition, for the first time, new research has linked high aflatoxin levels with an increased risk of developing tuberculosis (TB) in HIV positive individuals. TB transmission associated with aflatoxin exposure raises a new health concern among HIV positive individuals, in addition to concerns related to increased susceptibility to liver disease.<sup>46</sup>

Persons who are exposed to aflatoxin and are HIV positive have decreased plasma vitamin A and vitamin E in the blood, although there was no interaction detected between aflatoxin and HIV infection.<sup>47</sup> Nevertheless, other mechanisms have been proposed to explain the link between HIV and aflatoxin exposure. Williams et al. hypothesized that HIV infection is likely to increase aflatoxin exposure by two possible routes: (1) HIV infection decreases the levels of antioxidant nutrients that promote the detoxification of aflatoxin, or (2) the high degree of co-infection of HIV-infected people with hepatitis B also increases the biological exposure to aflatoxin. Although no specific studies on humans have yet been conducted, the evidence suggests a decrease in animal immune systems as a result of aflatoxin exposure.<sup>48</sup>

A more recent study by Williams concluded that the frequency of HIV transmission is positively associated with maize consumption in Africa. However, the relationship between cancer and nutrition suggests that contamination by fumonisin, another prevalent agricultural mycotoxin, rather than aflatoxin may be the most likely factor in maize that promotes HIV infection. Research suggests that improvements in the quality of maize may avoid up to 1,000,000 transmissions of HIV annually.<sup>49</sup>

*“... cases of aflatoxin poisoning in the East African region have since become a revolving epidemic...”*

## IMPACT OF AFLATOXIN IN AFRICA

Aflatoxin contamination is widespread in Africa and has been studied in several countries throughout Africa. Aflatoxin has been detected in many staple food crops such as maize, sorghum, teff, wheat, and milk. Although East African communities are of particular importance to the objectives of this review, the researchers have presented data on several regions throughout Africa. Research has been sectioned by country to maintain the integrity of reported research findings.

### C.1 IMPACT IN EAST AFRICA

In East Africa, there are numerous reports of fungal invasion and mycotoxic contamination of food crops. The recurrent cases of aflatoxin poisoning in the East African region have become a revolving epidemic, particularly in the arid and semi-arid areas of the region. This epidemic has been attributed primarily to crop planting and post-harvest practices. In Ethiopia, aflatoxin B<sub>1</sub> was detected in four crops: barley, sorghum, teff, and wheat. Retail stores and open markets had the highest risk of crop contamination. Studies in select countries in the East and Central African regions provide evidence of cases where aflatoxin poisoning has been observed.

#### Kenya

Aflatoxin poisoning continues to cause widespread disease and death in the rural areas of Kenya's Eastern and Central provinces. Rates of aflatoxin exposure and hepatitis B prevalence in rural populations are higher than rates in urban populations, even within the high-burden developing countries. This disparity may be explained by differences between an urban diet that is more diverse than rural populations' staple-based diet consisting of maize, peanuts, and other foods prone to aflatoxin contamination.<sup>50</sup>

Kenya has experienced several aflatoxicosis outbreaks during the last 25 years, most of which have occurred in the Makueni and Kitui districts in Eastern Province.<sup>51</sup> Both districts are prone to food shortages due to poor and unreliable rainfall and high temperatures. Aflatoxin contamination of maize is of particular global concern because maize is a widely cultivated staple food in many countries. In Kenya alone, more than 40 percent of rural and urban diets consist of maize and maize products.<sup>52</sup>

The first reported outbreak of aflatoxicosis in Kenya was in 1978; other outbreaks occurred in 1981, 2001, 2004, 2005, 2006, 2007, and 2008 that resulted in sickness, death, and the destruction of contaminated maize stocks.<sup>53</sup> The largest outbreak of 317 cases, including 125 deaths, reported in the world during the last 20 years occurred in Kenya from January to June 2004.<sup>54</sup> Analyses of brain and blood serum samples revealed that maize kernels from case households had higher concentrations of aflatoxin than did kernels from control households. Maize from the affected region contained as much as 4,400 ppb of aflatoxin B<sub>1</sub>, which is 440 times greater than the 10 ppb tolerance level set by the Kenya Bureau of Standards.<sup>55</sup>

A representative survey of maize products from agricultural markets and outlets in the Makueni, Kitui, Thika, and Machakos districts was conducted to assess the extent and magnitude of aflatoxin. Preliminary results indicated widespread, high-level aflatoxin contamination. A total of 182 (53.2%) of 342 samples had greater than the U.S. Department of Agriculture's (USDA) and WHO's acceptable level of 20 parts per billion (ppb) of aflatoxin. Moreover, a

*“Aflatoxin contamination of maize is of particular global concern because maize is a widely cultivated staple food in many countries.”*

*“Swift replacement of the aflatoxin-contaminated maize with noncontaminated maize proved to be a critical intervention.”*

substantial percentage of samples from each district, including Makueni (12.1%), Kitui (9.6%), Thika (3.9%), and Machakos (2.9%), had aflatoxin levels that were greater than 1,000 ppb. The government of Kenya provided replacement food in the most heavily affected districts, which included the Makueni district (population: 771,545) and the Kitui district (population: 515,422). The residents of affected districts were advised to avoid consumption of maize or other foods suspected to be moldy or appearing discolored. In addition, food inspections by public health authorities were conducted, and suspect food was seized, destroyed, and replaced. Surveillance for possible aflatoxin poisoning in humans was extended to other parts of Kenya by the Ministry of Health, and screening of potentially contaminated maize was increased.<sup>56</sup>

The 2004 Kenyan outbreak followed a poor maize harvest that had been damaged and consequently made susceptible to mold by drought. To guard against theft of the meager harvest, people stored the maize in their houses, which were warmer and moister than the granaries where the crop was usually stored. Health officials ruled out viral liver diseases when, suspecting acute aflatoxin poisoning, they examined maize samples and found aflatoxin B<sub>1</sub> concentrations as high as 4,400 ppb, which is 220 times the Kenyan limit for food. Swift replacement of the aflatoxin-contaminated maize with noncontaminated maize proved to be a critical intervention; however, as of July 2004, a limited number of new cases continued to be detected. As this outbreak demonstrates, aflatoxin poisoning will continue to be a public health concern until safe and scientifically appropriate post-harvest handling and storage methods for maize are adopted by the local population. In addition, enhanced surveillance for human aflatoxin poisoning and testing of commercially sold maize for aflatoxin levels will lead to long-term improvements in public health.<sup>57</sup> Prevention and detoxification methods have been outlined further in Section 3.

### **Uganda**

In Uganda, maize and peanuts are the two commodities most researched for aflatoxin contamination, and samples of both have turned up evidence of contamination. These two crops are major staple foods for the majority of people in the country.<sup>58</sup>

Aflatoxin research on food crops in Uganda started in the 1960s and continued into the early 1970s. The results of these studies indicated that a significant portion of the population was regularly exposed to aflatoxin contaminated foods. These studies also linked cases of liver cancer with high levels of aflatoxin in Ugandan foods. No aflatoxin research was reported in Uganda between 1971 and 1989, likely due to political insecurity during that time. From 1990 to the present, studies on produce stored at farms and markets have resumed. Little work has been done on the pre-harvest contamination of produce; additional research in designing management and control programs for the proper follow-up of aflatoxin contamination is necessary for recommendations on how to handle produce from the field through the post-harvest period.<sup>59</sup>



*“In Uganda approximately 1,000 cancer cases are registered annually.”*

*“...foodstuffs most vulnerable to aflatoxin in Sudan are peanuts and peanut products...”*

In Uganda approximately 1,000 cancer cases are registered annually.<sup>60</sup> At the Mbarara hospital in southwestern Uganda, only 40 cases are reported annually, yet many cases may go undiagnosed. Other than the hepatitis viruses that may lead to hepatocellular carcinoma, studies have determined that the aflatoxin levels of various suspected foods commonly consumed in the region may also be to blame for the high cancer rates.

### **Sudan**

The literature suggests that the foods most vulnerable to contamination with aflatoxin in Sudan are peanuts and peanut products. Sudan is a leading world producer of peanuts.<sup>62</sup> Younis and Malik<sup>63</sup> studied aflatoxin contamination in Sudanese peanuts and peanut products and found that the percentage of aflatoxin contamination was 2 percent, 64 percent, 14 percent, and 11 percent for kernels, butter, cake, and roasted peanuts, respectively. The researchers confirmed that aflatoxin B<sub>1</sub> was predominant in all samples, followed by G<sub>1</sub>, B<sub>2</sub>, and G<sub>2</sub>. The occurrence of liver cancer in Sudan could be substantially reduced by bringing the accepted levels for aflatoxin-contaminated food in line with internationally accepted levels.<sup>64</sup>

Sudanese peanut exports are governed by the strict standards of European and other countries with very low acceptable levels of aflatoxin. The high standards for the export market have resulted in the thorough sorting of peanut products to eliminate contaminated kernels. These contaminated kernels, however, may still find their way into the local market, particularly for oil processing factories.<sup>65</sup> The majority of oil produced in these factories may be contaminated with aflatoxin. Several vegetable oils, including peanut, cotton seed, sesame, and sunflower oil, are produced in Sudan and consumed by almost all segments of the population. Elzupir et al.<sup>66</sup> reported elevated aflatoxin levels in vegetable oils in the Khartoum state of Sudan. The magnitude of health risk resulting from the consumption of aflatoxin-contaminated oils is unknown.<sup>67</sup>

A study to determine the occurrence of mycotoxins in commodities, feeds, and feed ingredients sourced directly at animal farms and feed production sites found that Sudanese samples showed a high prevalence of aflatoxin, with 54 percent of the samples testing positive.<sup>68</sup> This illustrates the potential economic consequences of contamination to a global export such as peanuts.

“A high incidence of mycotoxin contamination in maize porridge suggests aflatoxins may play a role in kwashiorkor.”

## C.2 IMPACT IN SOUTHERN AFRICA

Several studies on aflatoxin have been conducted in Botswana, Malawi, and South Africa. Relevant findings have been highlighted in the following section.

### Botswana

In Botswana, Mphande et al.<sup>69</sup> reported the presence of aflatoxins as well as other contaminants in maize meal, with half of the samples containing aflatoxin at concentrations greater than 20 ppb. Fumonisin, another carcinogenic mycotoxin, has also been found in foods and feed in Botswana, even though aflatoxins were the most common toxins detected in the samples.<sup>70</sup>

### Malawi

Levels of up to 1,020 ppb of aflatoxin were reported in Malawian grains.<sup>71</sup> Malted maize and millet are used to make local brews that are widely consumed in Malawi and in Kenya. Although brews were not analyzed in this study, researchers determined in previous studies that toxins present in grains are not affected by normal cooking temperatures. The authors also reported a number of deaths in Kenya, which have been attributed to the consumption of local brews.

### South Africa

Researchers in South Africa have noted a high incidence of mycotoxin contamination in maize porridge.<sup>75</sup> Patulin, a mycotoxin produced by *Aspergillus Penicillium* and which damages the immune system in animals,<sup>72</sup> is generally associated with moldy fruits and vegetables and has been found in high levels in cider apples in South Africa,<sup>73</sup> particularly in areas where the temperatures are higher than 120 degrees Celsius.<sup>74</sup> Patulin, like aflatoxin, is another harmful mycotoxin produced by molds and fungi in the *Aspergillus* family.

The number of children suffering from kwashiorkor at hospitals in Durban has risen since 1992. These cases of kwashiorkor, marasmus, and underweight that were reported during this period correlated with findings of impaired liver function. As discussed earlier, researchers have suggested that aflatoxin may play a role in the pathogenesis of kwashiorkor.<sup>76</sup>

*“Lack of funded research activity by indigenous specialists in the mycotoxicology field deters meaningful breakthroughs for African scientists.”*

### **C.3 IMPACT IN WEST AND CENTRAL AFRICA**

Aflatoxin and fumonisin contamination are prevalent in crops in Sierra Leone and Ghana. Villages in Burkina Faso have experienced fumonisin contamination on maize, with levels as high as 29,000 ppb.<sup>77</sup> Researchers confirmed that 100 percent of the 72 samples from several local markets tested positive for fumonisin. Researchers also raised alarms about the consequences of chronic fumonisin exposure for the human population. Ghana, Nigeria, Senegal, Togo, and Burkina Faso have recorded aflatoxin contamination on sorghum, maize, cotton seeds, peanuts and peanut products, yams, and cassava at varying levels with contamination levels generally exceeding the EU and the USDA standards.<sup>78</sup>

#### **Nigeria**

As early as 1961, scientists at the National Stored Products Research Institute and the Institute of Agricultural Research, with the assistance of the Tropical Products Research Institute of London, demonstrated the susceptibility of peanuts to aflatoxin contamination in Nigeria. The prevalence of the toxic *Aspergillus* species on maize kernels from three agro-ecological zones in the northern part of Nigeria has also been well established.<sup>79</sup> The Nigeria Mycotoxin Awareness and Study Network has set up a system to create a mycotoxin map of the country that is expected to aid further studies and management.

Lack of funded research activity by indigenous specialists in the mycotoxicology field is a detriment to research opportunities in Nigeria, just as the continuing Western-dependent sourcing of procedures, materials, and personnel may hamper meaningful breakthroughs for African scientists. Direct national and regional initiatives are very important in the management of mycotoxins in food and feed.<sup>80</sup>

#### **Benin and Togo**

In West Africa, many people are not only malnourished, but are chronically exposed to high levels of mycotoxins. A study to determine the level of aflatoxin exposure among young children from Benin and Togo suggests a link with food consumption, socioeconomic status, agro-ecological zones of resilience, and culture-specific measures. Elevated aflatoxin levels were associated with child stunting, child mortality, immune suppression, and childhood neurological impairment.<sup>81</sup>

#### **Cameroon**

In Cameroon, researchers determined that cassava chips consumed by locals contained elevated aflatoxin levels, which may have occurred as a result of processing practices, conditions in storage facilities, and storage duration.<sup>82</sup> Although early studies first reported low levels of aflatoxin and other mycotoxins in maize in the humid forests and the Western Highlands of Cameroon, the constant presence of fumonisin from *Fusarium pallidoroseum* on stored maize calls for greater attention to toxigenic fungi, particularly those that can produce mycotoxins in the field.<sup>83</sup>

#### **The Gambia**

The Gambian experience of aflatoxin traces the presence of aflatoxin in the sera, maternal intravenous blood, breast milk, and umbilical cords of patients in the maternity wards.<sup>84</sup> In The Gambia, peanuts are a staple food and the primary cash crop, and their common consumption results in high and prolonged exposure to aflatoxins.

*“The Gambia Hepatitis Intervention Study clearly showed that the hepatitis B vaccination can be implemented in the national immunization programs of developing countries and that immunization is highly effective in preventing chronic hepatitis B infection and the likely onset of hepatocellular carcinoma.”*

The Gambia has a high liver cancer incidence,<sup>85</sup> endemic chronic hepatitis B infection, low but ever-present levels of hepatitis C infections, and near ubiquitous aflatoxin exposure. There is a long history of collaborative research between The Gambian Government Department of State for Health and international groups, starting with the establishment of the Medical Research Council-UK (MRC) field station in 1947. Early MRC research efforts focused on a variety of diseases, including hepatitis B infection. In 1986, the International Agency for Research on Cancer (IARC), in collaboration with the above partners, initiated The Gambia Hepatitis Intervention Study and the first program in Africa designed to assess the efficacy of hepatitis B vaccination in the prevention of chronic liver disease and liver cancer. A series of case-control studies were implemented to assess the role of aflatoxin exposure and hepatitis C infection, in addition to hepatitis B infection, in the case of liver cancer.<sup>86</sup>

The National Cancer Registry (NCR) data confirmed the high incidence of liver cancer in The Gambia with age-standardized incidence rates of 35.7 and 11.2 per 100,000 for males and females, respectively, the early onset of liver cancer with median age at presentation of 45 years, and male predominance, with an overall gender ratio of 3.4 males per female.<sup>87</sup> Researchers observed similar demographic patterns in liver cancer case-control studies conducted in The Gambia in 1981–1982, 1988–1989, and more recently in 1997–2001.<sup>88</sup>

In The Gambia, extensive research efforts have documented high liver cancer incidence resulting from childhood hepatitis B infections, lifetime dietary aflatoxin exposure, and chronic hepatitis C infections. In The Gambia, 57 percent of liver cancer cases are attributable to chronic hepatitis B infection. The Gambia Hepatitis Intervention Study clearly showed that the hepatitis B vaccination can be implemented in the national immunization programs of developing countries and that immunization is highly effective in preventing chronic hepatitis B infection and the likely onset of hepatocellular carcinoma.<sup>89</sup>

### **Ghana**

Studies in Ghana that collected samples from major processing sites in Accra reported aflatoxin levels that ranged from 2 ppb to 662 ppb,<sup>90</sup> quantities that far exceed the WHO and the USDA regulatory limit of 20 ppb.

*“Aflatoxin contamination is a serious health concern rooted throughout the entire food chain, thus necessitating a multidisciplinary approach to analysis, action, and solution.”*

## HEALTH SECTOR CAPACITY TO RESPOND

Aflatoxin contamination is a serious health concern rooted throughout the entire food chain, thus necessitating a multidisciplinary approach to analysis, action, and solution. In 2005, an Expert Group Meeting on Aflatoxin and Health held in Brazzaville, Republic of Congo, recommended that African countries take concrete measures to address this issue that negatively impacts livelihoods and lives, particularly those of poor people, who have limited freedom in food choices.<sup>91</sup>

### D.1 DIAGNOSIS AND TREATMENT

A broad range of signs and symptoms can be used to diagnose aflatoxicosis, depending upon the level of exposure.<sup>92</sup> Signs and symptoms include vomiting, abdominal pain and hemorrhaging, pulmonary edema, acute liver damage including fatty change, loss of digestive tract function, convulsions, cerebral edema, and coma. Other symptoms include yellow eyes, vomiting, abdominal swelling, water in the abdomen, leg swelling, general weakness, and drowsiness.<sup>93</sup> The onset of symptoms is relatively slow-acting, occurring about 8 hours after exposure. In cases of ingestion, feeding large quantities of an adsorbent, such as clay additives like NovaSil, may be used.<sup>94</sup>

### D.2 PREVENTION AND CONTROL

Hepatitis B vaccinations, education through awareness campaigns, and chemoprevention measures such as competitive displacement, plant extract applications, or methyleugenol sprays (see Section 3) have been shown to be effective in preventing and controlling the adverse health effects of aflatoxin.

#### Hepatitis Vaccinations

Hepatocellular carcinoma, liver cancer, is the fifth most common cancer in the world, with 80 percent of cases occurring in developing countries. The major risk factors for this cancer have been identified as chronic viral infections, such as hepatitis B and hepatitis C, and dietary exposure to aflatoxin. Given estimates that approximately 70 percent of liver cancer in developing countries is attributable to hepatitis B, a safe and effective vaccination to prevent chronic hepatitis B infection could prevent more than 250,000 cases per year.<sup>95</sup> Hepatitis B vaccination has not been formally considered as an aflatoxin control intervention, as the vaccine itself has no impact on actual aflatoxin levels in diets. However, it reduces the synergistic impact of hepatitis B and aflatoxin in inducing liver cancer.<sup>96</sup>

Studies have put the reduction in liver cancer cases attributable to hepatitis B vaccination from 30 to 50 percent. Perz<sup>97</sup> estimated that 50 to 80 percent of global liver cancer cases are attributable to hepatitis B and postulated that the corresponding reduction of hepatocellular carcinoma risk due to hepatitis B vaccination ranges from 45 to 50 percent. According to estimates by Kuniholm et al., lowering the risk of chronic hepatitis B infection through vaccination could reduce the risk of aflatoxin-induced liver cancer by as much as 30 times. Vaccination may also play some role in reducing aflatoxin-induced cirrhosis.<sup>98</sup> More recently, Khlangwiset estimated that the hepatitis B vaccine reduces total liver cancer by about 50 percent.<sup>99</sup>

With improved recognition of causal factors and understanding of the mechanics of the disease, interventions to reduce hepatocellular carcinoma incidence and morbidity can be designed and implemented. Vaccination against hepatitis B

in infancy is the most effective approach to prevent liver cancer, particularly in developing countries.<sup>100</sup> Concurrent reduction of exposure to aflatoxin B<sub>1</sub> may also prove an effective primary prevention measure. Vaccines for prevention of hepatitis C, however, are only just reaching early-phase clinical trials. Anti-viral treatments against hepatitis infection may also interrupt or delay the progression to hepatocellular carcinoma.<sup>101</sup>

A major challenge to incorporating the hepatitis B vaccination into a national immunization program is how to ensure the availability of the vaccine in countries with prevalent cases of aflatoxin poisoning. Key barriers to vaccination dissemination may include insufficient funding, lack of access to technology, and concerns about intellectual property rights. Development of a vaccine against hepatitis C is more problematic, due to the genetic heterogeneity of the virus. However, with 24 percent of liver cancer in developing countries attributable to hepatitis C (approximately 93,000 cases per year), such a vaccine would make a major contribution to cancer prevention.<sup>102</sup>

In Nigeria, aflatoxin and chronic hepatitis B infection account for approximately 8 to 27 percent and 59 to 62 percent of total liver cancers, respectively. Of the three aflatoxin control strategies tested in Nigeria (hepatitis B vaccine, biocontrols, and a calcium adsorbent called NovaSil clay), hepatitis B vaccination would reduce the greatest number of total liver cancer cases. Out of 43,000 total liver cancer cases, it was calculated that the hepatitis B vaccine, biocontrols, and NovaSil would reduce liver cancer by 49 percent, 5 to 19 percent, and 3 to 10 percent, respectively, as shown in **Figure 10**.<sup>103</sup>

Figure 10. Risk Reduction Estimates of Selected Control Interventions in Liver Cancer by Etiology

Items (per 100,00)	Interventions	Incidence of Liver Cancer		
		Aflatoxin-Related	HBV-Induced	Overall
<b>Baseline Risk</b>		65.73 – 220.58	475.42 – 499.59	805.78
<b>ARR</b>	Vaccine	50.76 – 170.39	450.44 – 475.81	395.05 – 424.47
	Biocontrol	37.17 – 151.47	31.27 – 127.41	36.98 – 151.82
	NovaSil	26.29 – 88.23	22.12 – 74.22	26.30 – 88.23
<b>RRR</b>	Vaccine	0.77	0.95	0.49 – 0.53
	Biocontrol	0.57 – 0.69	0.06 – 0.27	0.05 – 0.19
	NovaSil	0.40	0.04 – 0.16	0.03 – 0.11
<b>NNT</b>	Vaccine	587 – 1,970	210 – 222	236 – 253
	Biocontrol	660 – 2,690	785 – 3,198	659 – 2,704
	NovaSil	1,133 – 3,803	1,347 – 4,521	1,133 – 3,803

RRR = relative risk reduction; ARR = absolute risk reduction; NNT = number needed to treat; HBV = hepatitis B virus

### Awareness Campaigns

During the 2005 Kenya aflatoxin outbreak, individuals received information on maize processing and storage through an awareness campaign run by the Food and Agricultural Organization (FAO) of the United Nations and Kenya's Ministry of Health and Ministry of Agriculture. Those individuals



*“Awareness campaigns should use systems that are already in place for disseminating information...”*

*“As diseases in the developing world often go unreported, known outbreaks are likely to underestimate the problem...”*

receiving this information had lower serum aflatoxin levels than those who did not receive this information.<sup>104</sup> Awareness campaigns should use systems that are already in place for disseminating information to subsistence farmers. Awareness campaigns should distribute information to multiple organizations and use multiple means for spreading information to reach a broad range of people, given the diversity of cultures and the remoteness of villages.<sup>105</sup> Organizations providing this information would need to identify groups that are not receiving messages from current campaigns and appropriate methods for further outreach for those populations. The campaign disseminators would also need to determine why individuals or groups are unwilling or unable to adopt the recommendations.

### **Advances in Biomarker Technology**

Studies of how animals and humans metabolize aflatoxin have provided opportunities to develop chemoprevention approaches in human populations. Researchers examine the effects in the body and chart chemical effects stemming from aflatoxin exposure. The appearance of new chemical markers, or biomarkers, signal alterations in the body brought about by aflatoxin. Biomarkers can be used as outcome measures in these and other primary prevention studies. Biomarkers can also be charted in plants prone to aflatoxin exposure. The examination of biomarkers enables scientists to pinpoint areas where chemoprevention measures may be applied to crops, limiting the impact of aflatoxin contamination.<sup>106</sup> Chemoprevention measures such as clinical interventions to control aflatoxin contamination can be considered a “secondary” intervention as it cannot reduce aflatoxin levels in food, but can ameliorate aflatoxin-related illness by reducing the bioavailability of either aflatoxin or its reactive oxygen species that binds to DNA to initiate cancer.<sup>107</sup>

### **D.3 EPIDEMIOLOGICAL SURVEILLANCE SYSTEMS**

Recent exposure to aflatoxin is reflected in excreted urine, but only a small fraction of the dose is excreted in this way.<sup>108</sup> Previous outbreaks in Kenya have been identified by physicians who noticed an increase in cases of jaundice, despite the lack of any organized or official reporting system.<sup>109</sup> Although a national reporting system for jaundice would prove beneficial for developing countries, the baseline rate of jaundice and all of its possible causes are not known.<sup>110</sup> Given that aflatoxicosis confirmation tests using biologic markers are limited, an active and organized reporting system of possible aflatoxin cases may allow for earlier detection of potential outbreaks.

As diseases in the developing world often go unreported, known outbreaks are likely to underestimate the problem. Furthermore, the burden of disease attributable to chronic aflatoxin exposure such as liver cancer, impaired growth, immune suppression remains undefined.<sup>111</sup> These recurrent outbreaks emphasize the need to quantify and control aflatoxin exposure in developing countries and highlight the potential role of public health. An early warning system to monitor aflatoxin levels in food sources or individuals would prevent or reduce its adverse health effects. Monitoring aflatoxin levels in food or individuals to identify those at risk for disease is more difficult than monitoring incidence of aflatoxicosis; however, monitoring the rates of jaundice may identify susceptibility sooner and allow for a more timely intervention.<sup>112</sup>

In developed countries, adequate infrastructure exists for monitoring contaminant levels in foods, whereas poor, rural agricultural communities in developing countries tend not to have the choice of diverting contaminated foods away

*“An early warning system must include a response protocol to prevent further aflatoxin exposure and associated health outcomes once a contaminated food source has been identified.”*

from human consumption, and the overall food diversity tends to be low.<sup>113</sup> While at-risk foods are heavily regulated in North American and Europe, many developing countries lack regulations and consistent enforcement.<sup>114</sup> Several key interventions listed in Sections 3 and 4 includes biocontrols and testing methods which have previously been inaccessible to farmers and regulators in developing countries. A robust monitoring or surveillance system—such as that used in developing countries—would be difficult to establish and sustain.

To maximize resources, a targeted monitoring or surveillance system for high-risk areas or populations could be put into place that uses the most appropriate specimen (whether food, urine, or serum) that is appropriate for the country’s capacity to collect samples.<sup>115</sup> A combination of rapid field tests and laboratory confirmation tests that analyze aflatoxin in food or biologic samples would be ideal for an early warning system. An early warning system must include a response protocol to prevent further aflatoxin exposure and associated health outcomes once a contaminated food source has been identified. A protocol will be effective only if the infrastructure and funds to replace contaminated food exist and a method for identifying families in need has been determined. For an early warning system and response protocols to succeed, key members from various government agencies, the health care sector, and nongovernmental organizations need to be part the development and implementation of effective communication strategies and response systems.

#### **D.4 POLICIES AND STRATEGIES**

The African Union Ministers of Health came together in Johannesburg in 2007 to harmonize all existing health strategies by drawing an Africa Health Strategy to cover the period 2007–2015, which Regional Economic Communities (RECs), other regional entities, and member states could use to enrich their national strategies.<sup>116</sup> The overall strategy is meant to complement other specific and detailed strategies by adding value from the unique perspective of the African Union (AU) and provide a strategic direction to Africa’s efforts in creating better health for all. Unfortunately, the burden of aflatoxin contamination is not mentioned in the strategy.

In March 2011 at the 7th Comprehensive Africa Agriculture Development Programme (CAADP) Partnership Platform meeting in Yaoundé, Cameroon, African leadership requested that the African Union Commission (AUC) explore a “Partnership for Aflatoxin Control in Africa” (PACA) and link it to the CAADP process. The PACA will be an innovative, Africa-owned and led consortium to coordinate aflatoxin mitigation across the health, agriculture, and trade sectors of Africa, serving as a holistic model for a multisectoral solution. It will be embedded within existing African institutions and aligned with the CAADP process to leverage existing continent-wide harmonization efforts.

In June 2010 at the 10th annual African Growth and Opportunity Act (AGOA) forum in Lusaka, Zambia, the U.S. government (USG) announced that \$12 million of fiscal year 2011 (FY11) Feed the Future funding would be used to support the PACA priorities. The United States Department of Agriculture (USDA), the United States Agency for International Development (USAID) and the Centers for Disease Control and Prevention (CDC) were designated as the key agencies to lead USG aflatoxin mitigation programs across Africa. This funding is complemented by other donors and nongovernmental organizations.

*“...aflatoxin research in Africa is necessary to get policymakers in the Sub-Saharan region to recognize that the increased implementation of pre- and post-harvest aflatoxin control is an important avenue to increase food production and ensure food safety for the protection of the health of their citizens...”*

The PACA convened its first organizational planning meeting under the auspices of the AUC in Nairobi, Kenya in October 2011. Forty-one participants representing AUC member countries, regional economic communities (REC), trade organizations, donors, nongovernmental and farmers’ organizations, the private sector, and technical experts attended. The momentum put into motion as a result of this meeting marks a significant step toward addressing this formidable public health, agriculture, and trade issue.

Identifying public health strategies for the reduction of morbidity and mortality associated with the consumption of aflatoxin-contaminated food in the developing world means outlining an integrated plan that more effectively combines public health and agricultural approaches to the control of aflatoxin.<sup>117</sup> In the wealthy grain-producing countries of the world, sufficient economic resources exist to ensure that regulations to limit aflatoxin exposure in the food supply are implemented and the prices of corn and peanuts are often dictated by aflatoxin content, which contributes to much lower levels of exposure in wealthy countries.<sup>118</sup> Aflatoxin regulations in many least developed countries (LDCs) do little to protect public health, as there is limited enforcement of food safety regulations, especially among rural communities where food quality is rarely formally inspected.<sup>119</sup> Often there is no price differentiation in the market between contaminated and noncontaminated food and food products.

Policy development and implementation remains a critical issue in Africa, and this may be due to the lack of policy review and insufficient well-trained national expertise.<sup>120</sup> Systematic policy evaluations are also often not undertaken. National Environment and Health Action Plans (NEHAPs) are government documents that address environmental health problems in a comprehensive, holistic, and cross-sectoral way. NEHAPs are normally drawn up in cooperation with a wide range of partners, including professional and technical experts; national, regional, and local authorities; and nongovernmental organizations. However, these plans must be translated into action, so that they can be used as a platform for tackling aflatoxicosis incidences in Africa.

## **D.5 USING EVIDENCE FROM RESEARCH**

According to Hell and Mutegi,<sup>121</sup> aflatoxin research in Africa is necessary to get policymakers in the Sub-Saharan region to recognize that the increased implementation of pre- and post-harvest aflatoxin control is an important avenue to increase food production and ensure food safety for the protection of the health of their citizens. It would also be useful to educate stakeholders on the dangers of commercializing and consuming moldy foods. Research would also inform the development of infrastructure to accommodate surveillance as well as research on mycotoxins.

Coordinated and collaborative effort on aflatoxin research in Africa to minimize repetition would ensure that resources are focused on identified priority areas, including documenting the impact of aflatoxin on health and economies in Africa. Coordination and collaboration would also ensure the development of early warning mechanisms, especially in the highly prone areas, to avert the cases of acute poisoning that lead to fatalities.<sup>122</sup>

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# 3

*“Even low levels of aflatoxin may have long-term effects on livestock, poultry, and animal products.”*

## AGRICULTURE

### Summary

Aflatoxin contamination poses a serious risk to the farming industry with moderate to high levels of aflatoxin causing morbidity and mortality for both humans and livestock. Ongoing low levels of aflatoxin may cause long-term effects in livestock, poultry, and animal products such as meat, eggs, and dairy products. While the aflatoxin monitoring infrastructure in place in developed countries would be difficult to readily implement in the developing world, there are some relatively less expensive interventions that can be implemented pre- and post-harvest. Methods of pre-harvest handling to avoid aflatoxin contamination pre-harvest include the use of biocontrols, crop rotation, competitive displacement, and the use of different strains of maize and other crops. Use of interventions to reduce exposure to environmental stress can reduce aflatoxin contamination. Best practices for post-harvest handling include proper drying and processing, in addition to temperate, dry, pest-free storage.

Strategies to eliminate or limit the spread of aflatoxin contamination once crops have been harvested include food processing, storage strategies such as drying and improving conditions, and measures that are suitable and appropriately tailored for specific agro-ecological zones. Implementing a package or set of procedures to prevent aflatoxin contamination of crops is more effective than traditional post-harvest procedures and has reduced post-harvest exposure in the food chain by more than half.

Studies have shown that foods contaminated by aflatoxins can be detoxified through the use of inorganic salts and organic acids, ammoniation, and use of aflatoxin B<sub>1</sub> binding agents. Innovative research is currently ongoing on methods to detoxify contaminated crops using natural acids, salts and plant extracts. Ammoniation of crops also boosts deliverable protein in animal feed. Alternate uses of aflatoxin-contaminated crops include animal feed (if levels are sufficiently low), and ethanol production for biofuels. The pros and cons of these alternatives have been reviewed below.

### INTRODUCTION

Aflatoxin contamination poses a serious risk to the farming industry; moderate to high levels of aflatoxin can cause illness in humans and livestock. Even low levels of aflatoxin may have long-term effects on livestock, poultry, and animal products. Several methods of pre-harvest handling and best practices for post-harvest handling can reduce contamination. Currently, researchers are examining the effects of natural acids, salts, and plant extracts to detoxify contaminated crops. A more common method is ammoniation, or treating contaminated crops with ammonia vapor, which eliminates the aflatoxin-producing fungus, *A. flavus*.

*“In the prevention of aflatoxin, natural biocontrol methods can be applied pre-harvest or in the field as plants grow and mature.”*

## PREVENTION THROUGH PRE-HARVEST HANDLING

Methods of pre-harvest handling to avoid aflatoxin contamination include using biocontrols, instituting a process of competitive displacement using atoxigenic strains, employing different farming techniques, and developing breeds of stronger or more resilient strains of maize and other crops.

### B.1 BIOCONTROLS

Environmental pollution caused by excessive use of chemical pesticides paired with growing fear of the effects of chemicals on food destined for human consumption has led to increasing public pressure to remove pesticides from the agricultural market. Many regions of the world are beginning to regulate and even ban hazardous chemicals from use by farmers. In the last several years, pest management researchers have begun development of more natural methods in agriculture, chiefly in the development of biocontrols. *Biocontrols*, used in place of traditional chemical pesticides, are environmentally safe and derived from natural means and may include beneficial insects, plant extracts, or the introduction of other natural organisms. In the prevention of aflatoxin, biocontrol methods can be applied pre-harvest or in the field as plants grow and mature. Although methods of biocontrol may not be as effective as chemical counterparts, farmers and researchers continue to weigh the pros and cons of long and short term use of preventative measures.

#### Plant Extracts

Essential oils extracted from certain plants have proven to be a valid alternative to chemical pesticides and fungicides. Using plant extracts and cultivations of commonly used biocontrols, researchers examined the effects of applications of different amounts to stored quantities of rice. Compared to untreated rice, the application of certain plant extracts was able to reduce the level of aflatoxin B<sub>1</sub> by as much as 99 percent; the application of certain other biocontrols reduced aflatoxin B<sub>1</sub> levels by as much as 83 percent. These results demonstrate that treatment with either of these natural alternatives can effectively inhibit or reduce aflatoxin in crops.<sup>1</sup>

The effects of 41 types of essential oils on the growth of *A. flavus* were evaluated on maize grain in different conditions of water activity. The essential oils tested included anise, boldus, mountain thyme, clove, griseb, and poleo. The addition of essential oils showed an effect on the growth and accumulation of *A. flavus*, with clove, mountain thyme, and poleo having the greatest effect. Essential oils can be applied as a vapor, making application particularly convenient for use in closed storage.<sup>2</sup>

#### Methyleugenol Spray

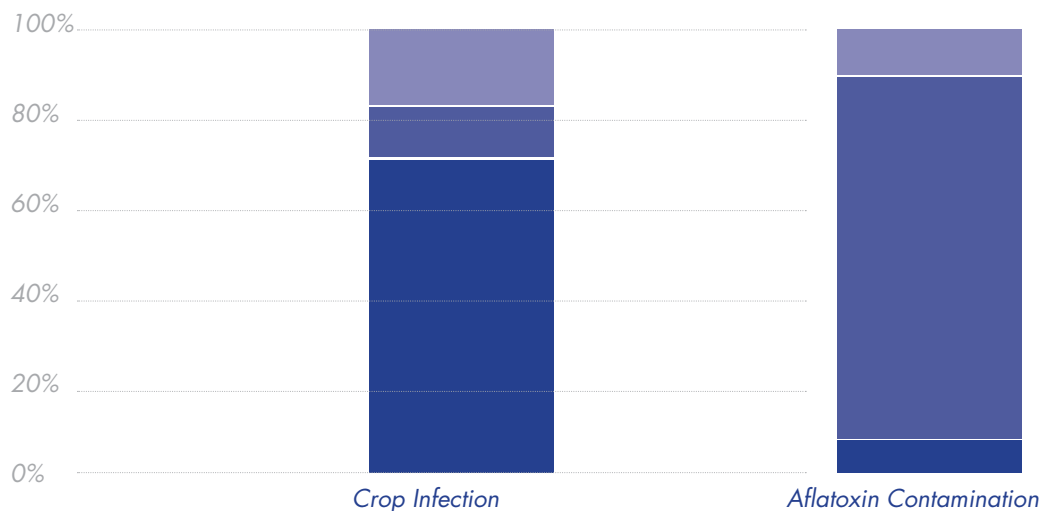
Methyleugenol is a naturally occurring substance present in essential oils and fruits. It is water soluble and is typically used as a flavoring in jellies, baked goods, nonalcoholic beverages, chewing gum, candy, puddings, relish, and ice cream at low concentrations. It is an organic alternative to manufactured chemical pesticides or fungicides, the use of which typically excludes peanuts for human consumption. In a study done in 2009, the use of methyleugenol proved to be a significant inhibitor of *A. flavus* growth on 56 percent of peanut agar meal. Methyleugenol also inhibited growth in the peanut pods and kernels. Although field testing should be conducted, the use of naturally occurring methyleugenol spray may be a valuable alternative to chemicals in preventing the growth of *A. flavus* on stored peanut crops.<sup>3</sup>

“Not every strain of *A. flavus* will produce aflatoxin.”

## B.2 COMPETITIVE DISPLACEMENT

There are several strains of *A. flavus*, however not every strain of *A. flavus* will produce aflatoxin. Some strains are atoxigenic, meaning that the fungus is not poisonous when consumed. When atoxigenic strains are applied to crops, they compete against toxic strains for resources. Generally, the application of atoxigenic strains results in a significant decrease in or elimination of the toxigenic strains that produce aflatoxin. Research has been done on peanuts<sup>4</sup> and corn (also referred to as *maize*)<sup>5</sup> with great results. Horn and Dorner<sup>6</sup> demonstrated reductions of 77 to 98 percent in aflatoxin contamination in peanuts with the application of atoxigenic strains. Abbas, Zablutowicz, Bruns, and Abel<sup>7</sup> determined that developing a mixture of atoxigenic strains (in their case CT3 and K49) had a greater impact than one strain alone. **Figure 11** shows the differences over all instances of aflatoxin contamination in two primary strains of *A. flavus*. These statistics demonstrate that the L strain of *A. flavus* may be a contender for competitive displacement situations.

Figure 11. Percentage of Crop Infection and Percentage of Aflatoxin Contamination Caused by Two Strains of *A. flavus*



Both L & S	18%	11%
S Strain	11%	81%
L Strain	71%	8%

Source: US Department of Agriculture (USDA) Agricultural Research Service (ARS), Etiology, 2010

*“The application of atoxigenic strains of *A. flavus* has proved to be highly effective in eliminating aflatoxin contamination in various crops.”*

*“Use of farming techniques, such as crop rotation and interventions to reduce exposure to environmental stress, can also reduce aflatoxin contamination.”*

Probst et al.<sup>8</sup> identified atoxigenic strains particular to districts in Kenya; 12 of these strains proved to be effective at reducing aflatoxin levels in maize by more than 80 percent, a level of efficiency comparable to a strain commercially distributed in the United States. These findings suggest that specific atoxigenic strains should be identified for use across the ecological zones of Africa. Identification of region-specific strains eliminates concerns regarding exposure rates of foreign strains of fungi. There is also the potential that useful strains that have been identified for a particular region may more effectively displace toxigenic strains to which they are more closely related.<sup>9</sup>

The application of atoxigenic strains of *A. flavus* has proved to be highly effective in eliminating aflatoxin contamination in various crops. However, certain methods of application have proven to be more effective than others. In a 2009 study, researchers examined several delivery methods: 1) inoculating soil with the atoxigenic strain, (2) spraying plants with cultured conidia—or spores—of the atoxigenic strain, and 3) spraying plants with a water-soluble product that included the atoxigenic strain. Researchers found that 50 percent of *A. flavus* sampled from inoculated soil was atoxigenic, 65 percent of the samples from plants treated with conidia were atoxigenic, and 97 percent of the samples collected from plants sprayed with formulated atoxigenic strains were atoxigenic. These results indicate that the most effective means of delivery of atoxigenic strains to competitively replace toxigenic strains of *A. flavus* is aerial spraying of formulated atoxigenic strains. However, as this option may not always be possible for smaller farms or communities, inoculating soil around plants can also be highly effective.<sup>10</sup> Competitive replacement is widely used for commercial farming operations across the southern United States with significant success.

### **B.3 FARMING TECHNIQUES**

Use of farming techniques, such as crop rotation and interventions to reduce exposure to environmental stress, can also reduce aflatoxin contamination.

#### **Crop Rotation**

Environmental factors that facilitate aflatoxin infection in the fields include soil and air temperature, relative humidity, water availability, drought stress, nitrogen stress, and spacing of plants. Plants that experience higher relative levels of humidity, periods of extreme fluctuations in humidity or temperature, stressful environments due to drought, weather, insects, and other stressors, or overcrowding are much more likely to become infected with *A. flavus*. Hell and Mutegi<sup>11</sup> report that aflatoxin levels in samples of Ugandan maize were higher in the samples collected from more humid areas than samples taken from drier regions; similar results were reported in samples from Nigeria under similar conditions.

Causal relationships have been shown between soil temperature and aflatoxin levels. A study completed in southern Texas demonstrated that aflatoxin levels are much lower in crops grown during winter months than in summer months, as soil temperatures had a great influence on *A. flavus* growth. In addition, researchers rotated crops, including corn (maize), cotton, and sorghum, between experimental fields. They found that all crops showed reduced levels of aflatoxin contamination during winter months, however corn consistently demonstrated higher levels of aflatoxin than the other test crops.<sup>12</sup>

Soil samples gathered after harvest showed higher incidences of aflatoxin



“Environmental stressors greatly impact the health and virility of crops.”

contamination in fields that had hosted corn than the other test crops year-round. These results occurred in both Bt (a species of genetically modified corn) as well as the non-Bt corn samples. The highest amounts of aflatoxin were found in fields with corn residues from cobs containing grain left in the fields.<sup>13</sup> As cotton and sorghum are much less prone to aflatoxin contamination than corn, these findings suggest that rotating corn with crops such as cotton, sorghum, or other plants less susceptible to aflatoxin contamination, as well as staggering the growing season, may allow for a healthier growth environment and generally decrease aflatoxin exposure.<sup>14</sup>

### **Environmental Stress**

Climate has a direct causal impact on crop growth and health. Strains of *A. flavus* are common between the latitudes of 40° degrees North and 40° degrees South worldwide. This includes irrigated deserts as well as in warm humid climates with the combination of heat and wetness as a key factor in facilitating fungal growth. In temperate regions, aflatoxin contamination is more likely during times of drought. Extreme climates can be difficult for plants, leaving them susceptible to damage by pests and to aflatoxin contamination. However, in extreme climates, some crops may be affected differently by aflatoxin than others. Although cotton-seed contamination has been positively correlated with periods of rain and increased humidity, drought is a major factor in the contamination of corn and peanut crops.<sup>15</sup>

Environmental stressors greatly impact the health and virility of crops. Plants that sustain stress from insects or climate are not as healthy as plants that do not; just like people, a stressful environment leaves plants susceptible to disease. Drought stress is a major contributor to pre-harvest aflatoxin contamination. Research shows that a higher level of stress-related or so-called defense proteins found in corn may cause different genes to predominate, leading to a different end-product at harvest. This can make it difficult to predict how any given harvest may be affected by drought, or how susceptible it may be to aflatoxin contamination. Research suggests that resistant strains should be bred to express higher levels of defense proteins, making crops more resilient to stressors such as drought, and consequently less susceptible to aflatoxin contamination.<sup>16</sup>

## **B.4 PLANT BREEDING FOR RESISTANCE**

Since the early cultivation of crops, humans have worked toward the development of stronger, more versatile, and more efficient plants. As suggested in the previous section, crops may be bred to express particular genes or proteins which make them less susceptible to environmental stressors. Other plants have been bred to produce greater quantities of food, called *higher yielding varieties*. In many instances, crops are now being genetically modified to strengthen natural properties and/or to combine beneficial genes from other plants or organisms to create more successful strains. Stronger plants are less susceptible to aflatoxin contamination. Two cases highlighted here are of Bt corn and participatory plant breeding (PPB) technologies.

One successful instance of genetic modification technology has been that of Bt corn. *Bacillus thuringiensis* (Bt) is a species of bacteria found throughout the world and is renowned for its insecticidal properties as a natural pesticide. First used as an insecticidal spray, the insertion of Bt DNA directly into corn has allowed for the production of insecticidal toxins by the plant itself. As a result, Bt corn has proved less susceptible to insect penetration. By 2000, Bt corn constituted a significant amount of the planting done in the United States,

*“The participatory plant breeding (PPB) approach provides stakeholders with the opportunity to consult and participate in technology and dissemination, thus increasing the probability that techniques and plant strains will be adopted by the community.”*

Argentina, and Canada. Many European countries, however, have been slower in adopting genetic modification technology as it is still uncertain what the long-term effects of genetic modification may be. The impact on the environment at large is still largely unknown. The elimination of certain pests will affect the insects and animals that feed on them, and the introduction of any new pesticide almost certainly leads to the eventual birth of resilient “superbugs.” Release of Bt corn, and any genetically modified plant, warrants further region-based research and consistent documentation.<sup>17</sup>

Peanuts are an important crop to East Africa as they are generally resilient, and high in food energy. One pound of peanuts provides the same energy or protein as 2 pounds of beef, 1.5 pounds of cheddar cheese, 9 pints of milk, or 36 medium-sized eggs. Consumed raw, blanched, roasted, crushed, as livestock feed, or as the oil extracted from the crop, peanuts are extremely versatile and nutritious. However, they are prone to many of the same issues plaguing other cash crops—disease, pests, and drought. Okello, Biruma, and Deom<sup>18</sup> describe a breeding practice, *participatory plant breeding* (PPB), that is currently employed in parts of Uganda and other areas of Africa. This program is largely a collaboration between farmers and plant breeders to select and cultivate strains of peanuts that would make the best additions to a formal breeding program. The PPB approach provides stakeholders with the opportunity to consult and participate in technology and dissemination, thus increasing the probability that techniques and plant strains will be adopted by the community. This collaboration also ensures that breeders and farmers become more familiar with one another’s needs, skills, motivations, challenges, and successes. PPB should be linked with the formal breeding system to ensure a continuous flow of novel genetic variability and an informal seed supply system which can spread new and promising varieties to local farms and communities.

PPB allows community members to develop stronger and more resilient plants through collaboration with one another and regional scientists. A practical application of technology in the field, plants that are bred to be stronger and more resilient to disease, pestilence, and climate are consequently also more resilient to damage attributable to aflatoxin exposure. As discussed earlier in this section, plants weakened by harsh weather conditions and insect invasion are more likely to show higher levels of aflatoxin. Okello, Biruma, and Deom propose a direct correlation between new, resilient plant strains and lower incidences of toxicity. In addition, community collaboration may also increase regional morale.

“...certain processing methods may significantly decrease aflatoxin levels in the end product.”

## PREVENTION THROUGH POST-HARVEST HANDLING

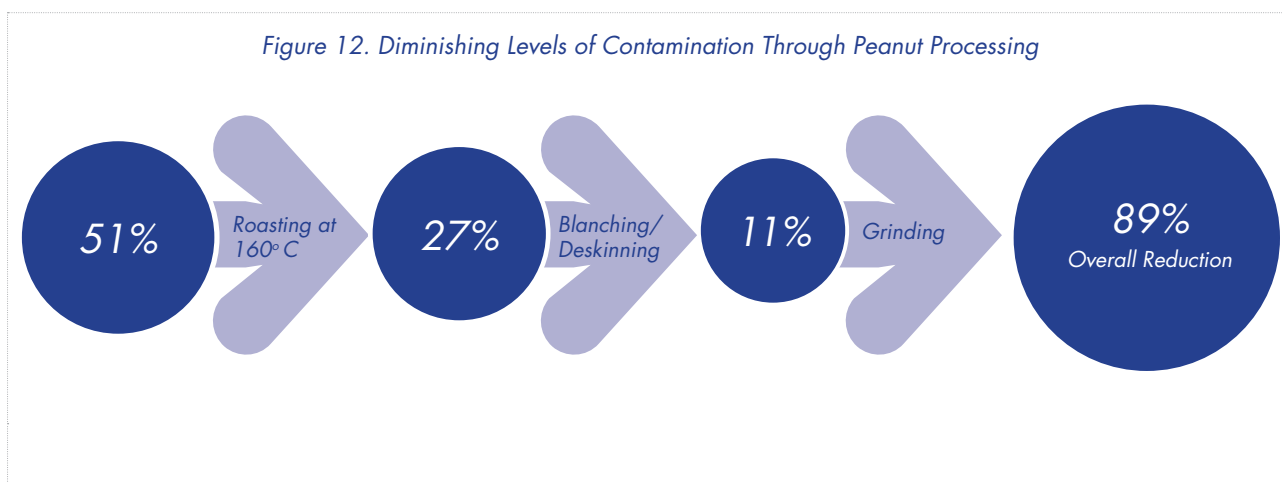
Despite the demonstrated effectiveness of biocontrols and other pre-harvest control methods, aflatoxin contamination remains endemic across Africa. Outlined below are several strategies for post-harvest use to eliminate or limit the spread of aflatoxin contamination throughout an entire harvest. These interventions include food processing, storage strategies such as drying and improving conditions, and measures that are suitable and appropriately tailored for the agro-ecological zone.

### C.1 PROCESSING

Many research studies have demonstrated that while processing procedures cannot completely eliminate aflatoxin exposure in harvested crops, certain processing methods may significantly decrease aflatoxin levels in the end product. Scudamore<sup>19</sup> notes a study completed in the United Kingdom, which demonstrated that a large portion of the mycotoxins found in harvested oats were concentrated in the hull of the plant. By removing the hull, only 5 to 10 percent of the mycotoxins remained in the groat, which forms the basis for human consumption of oats. Similar results have been demonstrated in pistachio crops. The hull acts as a natural barrier to the pistachio. However, pistachios harvested with cracked, stained, or otherwise damaged shells had higher levels of aflatoxin than pistachios with intact shells.<sup>20</sup> This suggests that crops with damaged or weaker hulls or shells are more susceptible to aflatoxin exposure and may require separate storage or more selective processing procedures to eliminate or diminish the threat of aflatoxin exposure, spread, or contamination.

One study on large-scale peanut butter production documented diminishing levels of aflatoxin throughout the processing of contaminated peanuts. Researchers found that roasting the peanuts had a dramatic effect on the levels of aflatoxin found in the nuts. The overall process reduced the levels of aflatoxin by 89 percent as shown in **Figure 12**.<sup>21</sup> Although the roasting process does not heat the nuts anywhere near the melting point of aflatoxin strains, 237°C–289°C, researchers assume that heating the nuts to 160 degrees Celsius damages the chemical structure of aflatoxin enough to reduce aflatoxin levels by half. Similar results have been shown when roasting coffee and pistachios.

Figure 12. Diminishing Levels of Contamination Through Peanut Processing



*“When leaks occur, documentation of environmental fluctuation within storage areas may help determine the potential for aflatoxin contamination in stored crops.”*

Scudamore<sup>22</sup> reviewed several methods of processing cereal-based products and found that in many cases, processing is not enough to decrease mycotoxin amounts to a safe level in foods. For example, he found that processing contaminated wheat to produce white bread, as opposed to whole grain bread, had different results. The white bread had much lower levels of mycotoxins, while the whole grain bread contained higher levels of mycotoxins. He concluded this was due to the level of processing the wheat flour. Flour for white bread is processed more thoroughly; however, it contains much less nutritional value than whole grain flour. He also noted that cereal-based products are manufactured using a number of different methods, which include cooking in water under raised temperatures, fermentation, baking, frying, drying, toasting, and extrusion. Each of these methods can have a different effect on diminished or intensified levels of mycotoxin in a food end-product.

High temperatures can diminish the threat of aflatoxin contamination; however, it should be noted that standard home-based cooking processes are generally insufficient to reduce aflatoxin to safe levels. Although baking can often decrease aflatoxin levels by as much as 48 percent, cooking and canning generally have little effect. Scudamore<sup>23</sup> reviews studies demonstrating that only 23 percent of mycotoxin contamination is lost during the home-based meal preparation of typical African corn porridge. Canning contaminated foods only resulted in a loss of 15 percent of mycotoxins.

## **C.2 STORAGE STRATEGIES**

### **Drying**

Researchers agree that the first step to successful storage of crops begins with minimizing the time between harvesting and drying whenever possible. Heated drying should be used when possible to avoid spoilage that can occur when crops are allowed to dry naturally, particularly when weather is humid at the time of harvest. Harvested grain, for example, at the time of harvesting typically contains 16 to 20 percent moisture. To prevent mold spoilage, wheat should be dried to at least 14.5 percent moisture. Grain is heated at low temperatures to dry it quickly. The heat does not eliminate aflatoxin exposure; but eliminating excess moisture in harvested grains is an effective method of inhibiting mold and fungal growth. Heat drying is effective at limiting the spread of *A. flavus* and other harmful fungi which can produce mycotoxins like aflatoxin.<sup>24</sup>

### **Storage Conditions**

Research has shown that fluctuations in temperature and humidity within storage silos and other buildings can lead to increased risk of mycotoxin exposure and contamination in harvested crops. Measures should be taken to minimize leaks in buildings. When leaks occur, documentation of environmental fluctuation within storage areas may help determine the potential for aflatoxin contamination in stored crops. Insect activity within storage facilities can also increase the temperature and moisture level in nearby crops. Besides causing potentially widespread damage and crop spoilage, increasing moisture levels in sections of stored crops can lead to fungal growth, which in turn can lead to the production of mycotoxins in food stores.<sup>25</sup>

Many farmers are now using silo bags to guard against insect infiltration and moisture. Silo bags are relatively inexpensive and hold between 180–200 tons of grain. Made of plastic, the bags are composed of three layers and are approximately 60 meters long with a diameter of 2.74 meters. The bags are

*“All storage methods were more effective in dryer regions, demonstrating that warm wet climates create an environment especially suited to fungal growth in stored crops. Storing maize near or above the fireplace using any method also correlated to lower levels of aflatoxin...”*

waterproof and have a certain level of gas-tightness, meaning that oxygen and carbon dioxide levels in the bags depend on the balance between respiration, the loss of carbon dioxide, and the entrance of oxygen into the bag. For long-term storage, this impermeability can lead to fungal growth; however, silo bags may be used as a short-term storage method when necessary.<sup>26</sup>

### **Climatic Effects**

A survey completed by African farmers across several agro-ecological zones demonstrated the variety of storage methods employed across zones and the problems that farmers have experienced. Farmers reported using the following methods: setting aside a room in the house to store crops; tying bushels in trees; suspending in a clay pot over the fireplace; storing in woven baskets kept over the fireplace; collecting in an outdoor wired crib; placing on top of a bamboo strip platform; tying maize bundles over the fireplace; storing in a *Rhumbu*, a traditional silo made of mud or grass; placing on an *Oba*, a woven grass mat supported on tree stumps; or placing in glass bottles, large barrels, or a silo, or polyethylene bags. The most common method of storage across zones was the use of storage bags, despite the higher reports of problems with fungi in the more humid areas. Researchers found that bags also contained the highest levels of aflatoxin. Bags also did not seem particularly effective at managing insect activity.

All storage methods were more effective in dryer regions, demonstrating that warm wet climates create an environment especially suited to fungal growth in stored crops. Storing maize near or above the fireplace using any method also correlated to lower levels of aflatoxin, leading researchers to assume that the dry smoke makes stored crops less susceptible to fungal growth and insect infestation.<sup>27</sup>

A study by Hell, Cardwell, Setamou, and Poehling<sup>28</sup> further demonstrated that different storage methods are better suited to different climates. They found that in many cases, storage methods were used in different agro-ecological zones without adaptation for use in that particular zone. For example, the use of the *Ago*, or large woven basket, in dryer, southern zones can be an effective storage method; however, when transplanted to damper, northern climates, this storage method becomes a haven for fungal growth and aflatoxin contamination. Researchers also found that the use of smoke to dry harvested crops, as well as storing crops above or near a smoke source, such as a fireplace, was an effective inhibitor of fungal growth. **Figure 13** highlights key findings from this study.

Figure 13. Rate of Insect and Fungal Problems by Storage System and Agro-ecological Zone

ZONE	STORAGE SYSTEM	PROBLEM	
		FUNGI	INSECTS
<b>Humid Forest</b> <i>(high humidity)</i>	Platform	29	34
	Bag	61	23
	Fireplace	50	35
	Pot	0	7
	Bottle	0	25
<b>Mid-Altitude</b> <i>(cool climate)</i>	Platform	0	13
	Bag	50	49
	Rhumbu	0	60
	Floor	0	20
	Tree	0	0
<b>Southern Guinea Savannah</b> <i>(annual rainfall 100cm–150cm)</i>	Bag	0	58
	Crib	40	13
	Fireplace	0	0
	Rhumbu	0	19
	Oba	0	27
<b>Northern Guinea Savanna</b> <i>(wet season lasts 4–6 months)</i>	Bag	0	84
	Rhumbu	0	28
	Floor	0	11
	Basket	0	20
<b>Sudan Savanna</b> <i>(dry season lasts 6–8 months)</i>	Bag	0	58
	Rhumbu	0	39
	Floor	0	7
	Basket	0	7

*“...implementing a package or set of procedures to prevent aflatoxin contamination of crops is more effective than traditional post-harvest procedures.”*

### **C.3 Implementation Package**

Research shows that implementing a package or set of procedures to prevent aflatoxin contamination of crops is more effective than traditional post-harvest procedures. In a study on peanut farming and consumption at the subsistence level in Sub-Saharan Africa, researchers surveyed 20 farms, half of which used a package of post-harvest measures and half of which followed the commonly followed post-harvest procedures. Levels of aflatoxin found in the blood of more than 600 community members spanning both groups were similar. Five months after harvest and peanut storage, samples were taken again. Researchers found that aflatoxin levels found in community members who had consumed peanuts harvested and stored by farmers who had used the package of post-harvest measures were 57 percent less than aflatoxin levels in community members who had consumed peanuts from farmers who had used the commonly followed post-harvest measures. The post-harvest package delivered to farmers in the first group included information and suggestions on various methods to reduce aflatoxin contamination:

- Identify peanuts that are moldy or have damaged shells.
- Use mats to dry peanuts to avoid humidity during the drying process.
- Judge the thoroughness of sun drying peanuts.
- Use natural fiber bags for storage.
- Store bags of peanuts on pallets instead of on the floor.
- Use insecticide.

Any one of these methods has been demonstrated to have an impact on aflatoxin contamination in the local population, however when used as a package these measures reduced post-harvest exposure in the food chain by more than half.<sup>29</sup>



*“In many cases, co-products or by-products of processed food that have levels of aflatoxin contamination that are too high for human consumption are instead used for animal feed...”*

*“... aflatoxin levels were highest in countries with warm, humid temperatures, such as Nigeria, Kenya, and Ghana, reaching greater than 70 percent contamination in feed samples.”*

## ANIMAL-FEEDING PRACTICES

In many cases, co-products or by-products of processed food that have levels of aflatoxin contamination that are too high for human consumption are diverted to animal feed if the contamination levels are still relatively low.<sup>30</sup> However in high doses animals may become ill, and their by-products then contain levels of aflatoxin that are still too high for human consumption, thus rendering products dangerous and inconsumable. It is difficult for researchers to predict the level of aflatoxin that may be present in dairy products after a cow has consumed contaminated feed. A certain amount of the aflatoxin will be absorbed and processed by the animal's body; however, the animal's health, long-term exposure to aflatoxin, and the amount of aflatoxin consumed at any given time are variables that can be difficult to track.

### D.1 CONTAMINATION LEVELS IN BY-PRODUCTS

Studies have shown that milk, cheese, and eggs can also be contaminated with aflatoxin. Aflatoxins have been found in fresh and sun-dried meats and poultry. Many farmers in developing countries have reported that they were not aware that milk, eggs, and meat could be contaminated by aflatoxins and were misinformed on ways to prevent exposure.

#### Dairy and Milk

In a 2009 study, researchers took 830 animal feed and 613 milk samples from urban centers in Kenya. Samples were analyzed for aflatoxin B<sub>1</sub> and aflatoxin M<sub>1</sub>, the form of the mycotoxin found in milk and other dairy products. Researchers found that 86 percent of feed samples were contaminated with aflatoxin B<sub>1</sub> and 67 percent of these samples exceeded the Food and Agricultural Organization (FAO) and World Health Organization (WHO) limits for aflatoxin. Approximately 72 percent of the milk came from small dairy farmers and 84 percent from large and medium scale farmers; however, 99 percent of all pasteurized, marketed milk was contaminated with aflatoxin M<sub>1</sub>. Of the contaminated milk, 20 percent, 35 percent, and 31 percent of that milk, respectively, was above the FAO and WHO limits for aflatoxin. Sixty-seven percent of urban smallholder dairy farmers had no knowledge that milk could be contaminated with aflatoxin.<sup>31</sup> Similar studies have been done in the Middle East, further demonstrating that this is a global health issue.<sup>32</sup>

Research remains sparse on the effects of processing milk products in the elimination of aflatoxin. Although there are no conclusive studies on most dairy-related manufacturing processes, the manufacturing of cheese with contaminated milk actually increases the concentration of aflatoxin M<sub>1</sub>—at least three times higher in soft cheeses and five times higher in hard cheeses. Of note, one misconception in dairy production may be that pasteurization would reduce aflatoxin contamination. In fact, treatments like pasteurization and sterilization have no effect on the amount of aflatoxin M<sub>1</sub> in dairy products.<sup>33</sup>

#### Meat and Eggs

Researchers have found a causal effect between levels of aflatoxin in the feed of laying hens and aflatoxin residues in their eggs. Even relatively low levels of aflatoxin in contaminated feed resulted in eggs contaminated with aflatoxin. Further, aflatoxin B<sub>1</sub> was found to be stable in naturally contaminated eggs for up to 20 minutes in boiling water. Although hard-boiling is an effective way

of eliminating other contagions such as salmonella and E. coli, it has little to no effect on the reduction of aflatoxin levels within the egg. The best way to decrease aflatoxin contamination in eggs is to prevent it altogether.<sup>34</sup> Other studies have shown mixed results, suggesting that very low levels of aflatoxin may not affect eggs, but may have an effect on hens' livers, where the toxin is stored. Further research should be conducted as to the long-term effects of an aflatoxin-contaminated diet on egg-laying and the characteristics of eggs.

A 2010 study sampled fresh and sun-dried meats sold in urban centers throughout Nigeria. The samples were analyzed for contaminants including aflatoxin. Aflatoxin was found in every sample. Aflatoxin levels were higher in fresh samples, but sun-dried samples had higher levels of other fungal contaminants, assumed to come from the use of contaminated tools in the slaughter and/or drying process. Although aflatoxin levels were lower than permissible limits, researchers suggest that withholding aflatoxin-contaminated feed from livestock for 3 to 4 weeks prior to slaughter may be enough to clear toxins from muscle tissue.<sup>35</sup>

Figure 14. Aflatoxin Contamination Across Country and Feed Type

Percentage of Contaminated Samples in African Countries						
Algeria	Egypt	Ghana	Kenya	Nigeria	S. Africa	Sudan
0%	19%	72%	78%	94%	6%	54%
Percentage of Contaminated Samples Across African and Middle Eastern Countries						
Maize	Wheat	Finished Feed	Soybean	Other Feed		
22%	6%	50%	8%	59%		

## D.2 EFFECTS ON LIVESTOCK

Livestock are affected by aflatoxin through the ingestion of contaminated animal feeds. Different species are more susceptible to aflatoxins than others. The effect of aflatoxins on poultry, especially chickens and turkeys, and pigs has been documented.

### Contamination of Animal Feed

Researchers conducted a survey of sampled animal feed from Africa and the Middle East throughout 2009. More than 300 samples were gathered and analyzed for mycotoxin levels. The team found that aflatoxin levels were highest in countries with warm, humid temperatures, such as Nigeria, Kenya, and Ghana, reaching greater than 70 percent contamination in feed samples. More than half of the samples retrieved from Sudan showed aflatoxin contamination, comparable to a 2009 study which demonstrated 64 percent contamination of animal feed.<sup>36</sup> Researchers also found that across all countries sampled in Africa and the Middle East, maize, finished feed, and other feedstuffs, which included smaller amounts of grass, alfalfa, cotton seed, sunflower meal, gluten, sorghum, barley, fish meal, and peanuts, had higher levels of contamination than other types of feed. Ultimately, researchers found a relationship between the pattern of aflatoxin contamination and the origin of the commodity.<sup>37</sup> **Figure 14** above highlights key findings from this study.

*“Supplementing potentially aflatoxin-contaminated feed with milk thistle has shown some positive effects in broiler chicks...”*

*“Clay additives have shown promise in inhibiting the effects of aflatoxin contamination...”*

### **Documented Effects on Poultry and Pigs**

Poultry is commonly considered highly susceptible to the effects of aflatoxin, including aflatoxicosis. Different species are more susceptible than others; turkey poults are the most sensitive, while chickens are the most resistant. Researchers studied the effects of aflatoxin-contaminated feed on the growth of turkey poults. Birds receiving 100–1,000 ppb of aflatoxin showed signs of aflatoxicosis such as poor feathering, apathy, and pallor of the feet and beak. Some birds receiving 500–1,000 ppb experienced convulsive crises. Higher levels of aflatoxin toxicity corresponded to lower levels of feed intake, which resulted in lower weight gain in poults fed contaminated feed.<sup>38</sup>

Studies show that chickens consuming diets contaminated with aflatoxin show renal lesions,<sup>39</sup> lower growth performance, diminished immune performance, and lower survival rates than birds fed contaminant-free diets.<sup>40</sup> Birds fed mycotoxin-rich diets had also developed fewer antibodies than birds fed clean feed. Supplementing potentially aflatoxin-contaminated feed with milk thistle has shown some positive effects in broiler chicks—possibly due to the ability of milk thistle to support the immune system through antioxidant, free-radical scavenging action that preserves the effects of other antioxidants in food, as well as other positive effects on the immune system. The addition of milk thistle into contaminated diets also showed a positive effect on weight gain in broiler chicks.<sup>41</sup>

Pigs are also highly susceptible to aflatoxin poisoning. Aflatoxin has similar effects on swine as on poultry such as compromised immune system, poor weight gain, reduced feed intake, and organ damage. Adverse effects have been shown both at both high and low doses; doses as low as 175 ppb—in combination with other contaminants—have been shown to cause hardening of the liver and damage to related vessels. Clay additives have shown promise in inhibiting the effects of aflatoxin contamination; however, further research is required before wide-scale use is recommended.<sup>42</sup>

*“...there are alternatives to discarding tainted foodstuffs...”*

*“Ammoniation is currently the most economically viable decontamination process.”*

## HANDLING CONTAMINATED COMMODITIES

When contamination does occur, there are alternatives to discarding tainted foodstuffs, such as the processing procedures summarized in section C.1 Processing. Other alternatives include treating contaminated crops with binding agents or other substances to eliminate the aflatoxin or finding alternative uses for crops beyond consumption.

### E.1 TREATMENT

Studies have shown that foods contaminated by aflatoxins can be detoxified through the use of inorganic salts and organic acids, ammoniation, and use of aflatoxin B<sub>1</sub> binding agents.

#### **Inorganic Salts and Organic Acids**

Shekhar, Singh, Khan, and Kumar<sup>43</sup> demonstrated the efficacy of six chemicals in the degradation of aflatoxin levels in stored maize. These nontoxic chemicals are safe for use with foods and include sodium carbonate, sodium bicarbonate, potassium carbonate, ammonium carbonate, acetic acid, and sodium propionate. The research team showed that each of these chemicals, derived from inorganic salts or organic acid, were effective in reducing the concentration of aflatoxin up to 88 percent. Further tests were completed which demonstrated that the organic acids (acetic acid and sodium propionate) were effective in reducing aflatoxin concentration up to 69 percent in ambient storage conditions for eight months.

Another study recorded the effects of citric acid in the detoxification of rice contaminated with aflatoxin. Citric acid is another organic acid found in fruits and commonly used to adjust flavoring in fruit and vegetable juices and candy. This food additive acts as a natural antimicrobial preservative, slowing food spoilage. Researchers found that when applying citric acids to rice that was low in levels of aflatoxin (containing less than 30 ppb), aflatoxin spores were completely degraded. In rice which contained higher levels of aflatoxin (containing 30 or more ppb), 97.22 percent of the aflatoxin spores were degraded.<sup>44</sup>

#### **Ammoniation**

Of all decontamination processes outlined in this document, ammoniation is currently the most economically viable. Ammonia in a gaseous form is added to crops in a sealed area and allowed to permeate for 1 to 2 weeks. In a study on artificially contaminated corn, ammoniation procedures destroyed 90 percent of the aflatoxin.<sup>45</sup> This practice typically makes products previously unsafe for consumption, safe for livestock consumption by decreasing aflatoxin levels to a safe range for animals.

Studies on broiler chickens demonstrate the effects of ammoniation on animal feed. Researchers assembled two groups of chicks; one group was fed aflatoxin-contaminated maize, while the other group was fed aflatoxin-contaminated maize that had been treated with ammonia. After 6 weeks, the chicks that were fed aflatoxin-contaminated maize showed a significant increase in mortality rate as compared to the chicks that were fed the treated feed. Dietary intake, weight gain, and feed-conversion rate was suppressed in chicks fed the aflatoxin contaminated diet, while chicks fed the treated feed showed normal growth.<sup>46</sup>

In a 2008 study, researchers showed that treating corn with ammoniation procedures did not affect the general health of broiler chicks. Broilers were

separated into four groups: group A was fed normal clean corn; group B was fed clean corn which had been treated with ammonia; group C was fed corn contaminated with 1000 ppb of aflatoxin B<sub>1</sub>; group D was fed corn contaminated with 1000 ppb of aflatoxin B<sub>1</sub> which was then treated with ammonia. Broilers fed with contaminated corn which had not been ammoniated had a significantly higher mortality rate than broilers in other groups. Broilers in all other groups had comparable mortality rates.<sup>47</sup> **Figure 15** highlights the key findings of this study.

Figure 15. Mortality Rates of Broiler Chicks Fed Contaminated and/or Ammoniated Feed

	Condition	AfB <sub>1</sub> (ppb) in feed	Mortality rate
<b>Group A</b>	<b>Uncontaminated corn</b>	0	5.8%
<b>Group B</b>	<b>Ammonia-treated uncontaminated corn</b>	0	6.5%
<b>Group C</b>	<b>Contaminated corn</b>	650	22.5%
<b>Group D</b>	<b>Ammonia-treated contaminated corn</b>	3.5	8.7%

### Binding Agents Effective on Aflatoxin B<sub>1</sub>

Several studies are being done regarding the use of clays, (e.g., NovaSil) to decrease the threat of aflatoxin. One study proposes that smectite clay be administered to contaminated food to act as a binding agent. The clay binds with aflatoxin B<sub>1</sub> molecules, shielding them from absorption into the digestive system when consumed and allowing them to pass harmlessly through the body.<sup>48</sup> Other studies are examining the effects of bacteria applied to contaminated feed as a binder.<sup>49</sup> Researchers experiencing high reduction rates, however, are constrained to specific strains of bacteria. These theories require much more research prior to large-scale use.

## E.2 ALTERNATIVE USES

Alternate uses of aflatoxin-contaminated crops include animal feed (only if levels are low), in the wet milling industry, and in ethanol production. In the wet milling process, corn is steeped in water and sulfur dioxide to swell the kernels. When the process is complete, nutrients from the kernels will have entered the liquid and can be condensed, processed, and used for animal feed. The remaining portion of the kernel can be used for oil or starch, which can be further processed into fructose syrup.<sup>50</sup> The production of ethanol is another potential use for contaminated fuels. Ethanol is a biofuel currently being used as an environmentally friendly additive to fossil fuels. However, one by-product of ethanol production is a substance often used for animal feed which, following ethanol production using contaminated corn, then becomes a more concentrated contaminated source of aflatoxin. Animal feed resulting from each of these alternatives should be monitored to ensure it meets regulated aflatoxin standards for livestock feed.<sup>51</sup>

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# 4

*“Standards for aflatoxin limits vary across foodstuffs, country or region, and the intended use of the food.”*

*“...aflatoxin cost Africa \$750 million USD each year in exports of cereals, dried fruit, and nuts.”*

## TRADE

### Summary

Mycotoxins impact the trade of several agricultural products including cereals, oilseeds, root crops, dried fruits, and coffee beans, which form the agricultural economic foundation of most developing African countries. Standards for aflatoxin limits vary across foodstuffs, country or region, and the intended use of the food. Classic technologies for detecting and quantifying mycotoxins can include high performance liquid chromatography (HPLC), fluorescence or mass spectrometry detection, thin-layer chromatography, ELISA (enzyme-linked immunosorbent assay), gas chromatography, and flame-ionization chromatography.

International guidelines on aflatoxin are provided by the Codex Alimentarius Commission established by the Food and Agricultural Organization of the United Nations (FAO) and the World Health Organization (WHO). In addition the U.S. Department of Agriculture (USDA), the European Union (EU), Canada, and many African and Asian countries have established specific regulations on acceptable levels of aflatoxin in human food and animal feed. Trade regulations on aflatoxin have been imposed for decades and are becoming increasingly strict: the acceptable range of aflatoxin for humans is from 0.5 ppb in milk to as much as 20 ppb in processed foods.

Nontariff barriers are barriers to trade that are not based in laws, treaties, or official regulations. They are often contained within in the rules and regulations of a country relating to trade or product sanitary and phytosanitary standards. Key nontariff barriers include technical barriers, such as safety standards, electrical standards, environmental standards, health standards, and other protective codes. Compliance costs associated with the rejection of food products due to a failure to meet regulations can be significant. For example, Europe is the most important market for African exports. Between 1989 and 1998 the EU imported approximately 66 percent of its peanut exports from African countries.<sup>1</sup> Under more stringent EU harmonization guidelines, exports of cereals and cereal preparations in 1998 could have declined by 59 percent, or \$177 million. While adoption of more lenient Codex standards would have increased exports of cereals and cereal preparation by 68 percent, or \$202 million in 1998, standards have not been relaxed.

Dr. Kofi Annan, the former UN Secretary General, has called on the Western nations to redress a global trade that is disadvantageous to the developing countries as a result of rejected commodities that are short on quality. He cited a World Bank study which revealed that the European Union regulation on aflatoxin cost Africa \$750 million each year in exports of cereals, dried fruit, and nuts.<sup>2</sup>



## INTRODUCTION

Rising standards and lower limits for aflatoxin contamination have had an enormous impact on the ability of developing countries in Africa to export goods. Regulations differ across countries and regions and across food and feed types. Despite science-based recommended standards, regulations have become more and more strict, making exportation of goods more and more difficult for developing countries. Several methods of testing exist to analyze aflatoxin levels. Thin layer chromatography, ELISA, and rapid testing methods are inexpensive testing methods, although not as precise as more expensive tests. The primary barrier to trade is the strict aflatoxin limits set by Africa's key trade partner, the EU. Estimates put losses due to stringent EU limits at between \$400 and \$450 million annually.

## GENESIS OF AFLATOXIN STANDARDS

Although the United States, the EU, and Canada have imposed trade regulations on aflatoxins for decades, regulations are becoming increasingly strict. However, strict legal limits are being increased and/or being imposed with limited regard to regionally focused research or the ability of developing nations to meet newly imposed standards. The Codex Alimentarius Commission, established by the FAO and WHO, provides international guidelines.

Figure 16. USDA Regulations for Aflatoxin Levels in Food

Product	Total Aflatoxin Level (ppb)
<b>Food for Human Consumption</b>	20
<b>Milk</b>	0.5
<b>Beef Cattle</b>	300
<b>Swine (over 100 lbs.)</b>	200
<b>Breeding Beef Cattle, Swine, or Poultry</b>	100
<b>Immature Animals</b>	20
<b>Dairy Animals</b>	20

Source: National Grain and Feed Association. [www.ngfa.org/files/misc/Guidance\\_for\\_Toxins.pdf](http://www.ngfa.org/files/misc/Guidance_for_Toxins.pdf)

### B.1 MULTINATIONAL STANDARD SETTING

Established by the FAO and WHO in 1963, the Codex Alimentarius Commission is intended to protect consumer health and to ensure fair practices in food trade. Although recommendations are science-based and nondiscriminatory, it is important to note that the Codex does not impose control over regulations. Recommendations made by this commission are advisory in nature. Dohlman<sup>3</sup> proposes that food safety regulations which are stricter than Codex recommendations can impose an unfair economic burden, particularly on exporter nations with a lower gross income (2008). In 1997, the Joint FAO/WHO Committee on Food Additives (JECFA) demonstrated the possible impacts of two alternative standards (10 ppb and 20 ppb) on human health using two example populations and diets: a European diet with 1 percent of the population testing positive for hepatitis; and a Far Eastern diet with 25 percent of the population testing positive for hepatitis. For regions with higher rates of hepatitis, this small difference in the limit of aflatoxin could save as many as 300 cancer deaths per billion people each year. **Figure 16** above highlights U.S. aflatoxin standards that are regulated by the USDA.

*“Harmonized regulations proposed by the EU established a standard of 4 ppb of total aflatoxin strains allowed in cereals and peanuts intended for human consumption...”*

Harmonized regulations proposed by the EU established a standard of 4 ppb of total aflatoxin strains allowed in cereals and peanuts intended for human consumption; of that 4 ppb, no more than 2 ppb may be composed of aflatoxin B<sub>1</sub>. Foodstuffs intended for further processing are permitted higher levels of aflatoxin, as some processing procedures decrease aflatoxin levels. Despite the relaxation of standards for food intended for further processing, limits set by the EU are still much lower and stricter than Codex suggestions, as well as standards set in many developing countries, leading many to protest these levels as unfair trade regulations that limit the export potential of developing countries. Several factors influence aflatoxin tolerance limits: (1) the availability of toxicological data, including hazard identification and characterization; (2) the availability of data on aflatoxin occurrence within and across commodities; (3) the availability of analytical methods; (4) domestic trade interests and foreign regulations; and (5) the domestic food supply situation. The importance of each of these factors varies over time and across countries. Each of these factors should be taken into account when developing regulations on aflatoxin limits.<sup>4</sup>

From 1986 through 1994, the eighth round of multilateral trade negotiations was held by the General Agreement on Tariffs and Trade (GATT), now known as the World Trade Organization (WTO). Negotiations included over 123 countries and were named the Uruguay Round Agreement on Agriculture. One of the outcomes was the Agreement of the Application of Sanitary and Phytosanitary (SPS) Measures, which includes a series of understandings on how SPS measures for animals and plants should be regulated in trade. Countries agreed to ensure that SPS measures used would not discriminate against trade partners. Countries may use reasonable SPS measures, or combinations of measures, on goods to bring food contaminants into an acceptable range. SPS measures are protective measures applied in many situations:<sup>5</sup>

- SPS measures can protect human or animal life from risks arising from additives, contaminants, toxins, or disease-causing organisms in their food.
- SPS measures can protect human life from plant or animal carried diseases.
- SPS measures can protect animal or plant life from pests, diseases, or disease-causing organisms.
- SPS measures can prevent or limit other damage to a country from the entry, establishment, or spread of pests.

## **B.2 EUROPEAN HARMONIZATION**

In 1998, the EU announced common regulations for maximum allowances of aflatoxin in imported foodstuffs. However, empirical evidence on the trade impact of tightening standards is extremely limited. Little baseline information has been produced to inform trade policy and decision making. Otsuki et al.<sup>6</sup> reviewed a 1997 study by FAO which uncovered several issues impacting compliance by developing countries: lack of funds allocated to research aflatoxin, scarcity of highly trained personnel, inadequate facilities for safe aflatoxin research, lack of maintenance of laboratory facilities, and inadequate infrastructure. Otsuki also notes that although more sensitive equipment is being made available for the testing of aflatoxin levels in foodstuffs, the development of successful farming techniques is still lagging.

*“As of 2003, aflatoxin regulations existed for five countries in Africa, including Kenya and South Africa.”*

## AFLATOXIN STANDARDS FOR HUMANS AND ANIMALS

Standards for aflatoxin limits vary across foodstuffs, country, or region, and the intended use of the food. Outlined below are limits for each of these categories.

### C.1 HUMAN CONSUMPTION

**European Union:** The EU has some of the strictest standards for mycotoxins, including aflatoxin, in the world. The limit for aflatoxin is 4 ppb.<sup>7</sup>

**United States:** The United States has adopted 20 ppb as the maximum level for aflatoxin<sup>8</sup> and 0.5 ppb for milk<sup>9</sup>.

**Canada:** A standard of 15–20 ppb has been set for finished food products.<sup>10</sup>

**Asian Countries:** Chinese levels of aflatoxin B<sub>1</sub> in peanut butter and sesame paste cannot exceed 20 ppb. Regulations are based on the EU Commission Directive 2003/121/EC, which states that for peanuts, nuts, dried fruit, and cereals intended for direct human consumption, the measurement uncertainty and correction for recovery should be taken into account if one or more of the subsamples exceed the maximum limit beyond a reasonable doubt.<sup>11</sup>

**African Countries:** Aflatoxin remains largely unregulated throughout Africa. As of 2003, aflatoxin regulations existed for five countries in Africa, including Kenya and South Africa. Standards range between 10 ppb and 20 ppb.<sup>12</sup> For many countries, regulations have not yet been mandated or strongly enforced.

### C.2 ANIMAL FEED

Widespread concern about potential effects of aflatoxin in humans and animals—as well as possible transfer of aflatoxin residues into edible animal tissues and milk—has led to regulatory actions in the EU and the United States. Products exceeding these levels may be seized and destroyed.<sup>13</sup> Of note, researchers predict that it may take approximately 72 hours on aflatoxin-free feed for milk to become aflatoxin-free;<sup>14</sup> others suggest that withholding aflatoxin-contaminated feed from livestock for 3 to 4 weeks prior to slaughter may be enough to clear toxins from muscle tissue.<sup>15</sup> Outlined below are regulations for animal feed:

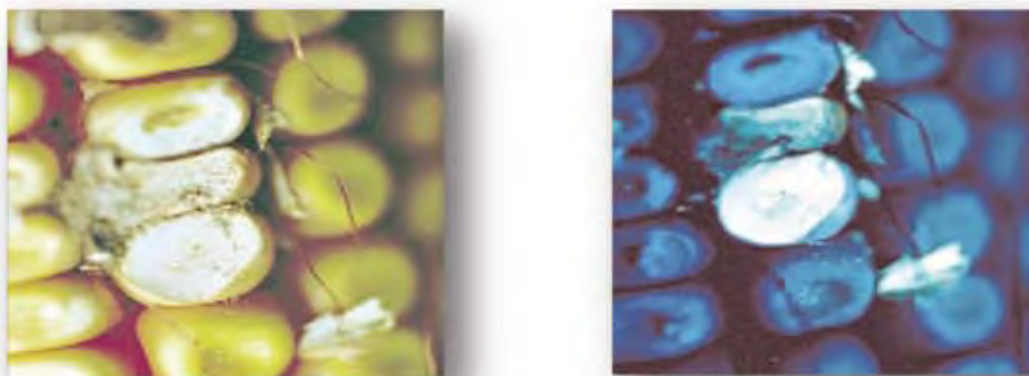
**European Union:** Maize intended for feed cannot exceed 20 ppb of aflatoxin B<sub>1</sub>.<sup>16</sup>

**United States:** For mature non-lactating animals, aflatoxin limits have been set at 100–300 ppb, depending on the feed type and the animal species.<sup>17</sup>

## TESTING PROCEDURES

There are several testing methods and technologies that can be used in identifying the level of contamination in various foodstuffs. Technologies range in effectiveness and expense. Classic technologies for quantifying mycotoxins include high-performance liquid chromatography (HPLC), fluorescence (see **Figure 17**) or mass spectrometry detection, thin-layer chromatography, gas chromatography, and flame-ionization chromatography. Concentration data for the presence of mycotoxins in food and feed are needed by researchers, policymakers, and risk managers; there is no single testing method for mycotoxins. Gas chromatography, HPLC, and liquid chromatography are very accurate tests, however the equipment necessary to complete these tests is very expensive and requires a high level of expertise to conduct.

*Figure 17. Contaminated Corn under Normal and Ultraviolet Light*



*Source: Cassel et al, 2001. South Dakota State University*

A variation, the thin-layer chromatography test, is a simple, inexpensive test for aflatoxin; however this test typically is not as sensitive and lacks precision. Another method, ELISA testing, requires a simple preparation and inexpensive equipment. ELISA testing is highly sensitive, good for screening, and can be used to test for related mycotoxins in addition to aflatoxin, but it can sometimes lead to false-positive results. Rapid testing using a membrane-based card test, antibody-coated tube, and/or immunodot cup tests are simple, fast (procedures take 5–10 minutes), inexpensive, and generate quantitative or semi-quantitative data. Rapid testing can also generate false positive results, and the methods lack sensitivity in testing results that approach regulatory limits.<sup>18</sup>

*“Compliance costs associated with the rejection of food products can be significant, particularly to peanut exporters.”*

## TRADE BARRIERS

Compliance costs associated with the rejection of food products can be significant, particularly to peanut exporters. The U.S. peanut industry estimated in 2001 that complying with EU sampling standards would result in an additional \$150 per lot for raw peanuts. The industry calculated a rejection rate of approximately 30 percent for U.S. exports. African exporters of peanuts depend heavily on the European market. Between 1985 and 1998, 56 percent of African exports of edible peanuts, 61 percent of exported peanut oil, and 74 percent of oil seeds were sold to countries in the EU. Researchers estimate that the impact of tightening aflatoxin B<sub>1</sub> standards by a 10 percent reduction in acceptable levels would lead to an 11 percent reduction of exports of edible peanuts to the EU. This estimated loss totals more than \$238,000, approximately 36 percent of the total trade value exchanged in 1998.<sup>19</sup>

### **E.1 IMPACT OF MYCOTOXINS ON COMMODITY TRADE**

Mycotoxins impact the trade of several agricultural products, including cereals, oilseeds, root crops, dried fruits, and coffee beans, which form the agricultural economic foundation of most developing African countries. Under more stringent EU harmonization guidelines, exports of cereals and cereal preparations in 1998 would have declined by 59 percent, or \$177 million. Adoption of more lenient Codex standards could have increased exports of cereals and cereal preparation by 68 percent, or \$202 million in 1998. For edible nuts and dried and preserved fruits, the estimated decline in African exports to the EU was \$220 million (47%) under EU harmonization guidelines; under Codex standards, increased exports are estimated at \$66 million (14%). Wu estimates export losses at \$450 million under EU export guidelines, almost five times higher than if U.S. standards were adopted (2004). In 2007, the African Peanut Council estimated that the annual cost of implementing a program to reduce aflatoxin contamination to allowable levels for EU export could reach \$7.5 million.<sup>20</sup>

*“Nontariff barriers are barriers to trade that are not based in laws, treaties or official regulations.”*

## **E.2 NONTARIFF BARRIERS TO TRADE**

Nontariff barriers are barriers to trade that are not based in laws, treaties or official regulations. They are often buried in the rules and regulations of a country relating to trade or product *standards*. Key nontariff barriers can take different forms:<sup>21</sup>

- Subsidies, payments, or assistance to domestic producers and businesses can make domestic businesses more competitive as compared to foreign competition.
- Emergency import protection to counteract sudden surges in imports that could damage the local economy can put foreign products at a disadvantage.
- Administrative barriers, such as unnecessary procedures with respect to customs inspections, can keep imports from entering the country.
- Industrial and commercial practices, embargoes, or boycotts can destroy foreign businesses.
- Technical barriers, such as safety standards, electrical standards, environmental standards, health standards, and other protective codes can drive up the cost of doing business.
- Practices such as social or cultural forces, monetary exchange controls, foreign government procurement policies, licensing schemes, and even corruption can have negative consequences for trading partners.

“...Kenya’s trade policies have become the most important regulator of regional maize flow...”

## TRADE FLOWS

Top agricultural exports for Africa include maize and peanuts. Examples of trade flows for Kenya—as well as a brief overview of the impact of World Food Programme activities in Africa—are outlined below.

### F.1 IMPORTS AND EXPORTS

Europe is the most important market for African exports; between 1989 and 1998, the EU imported approximately two-thirds of its peanut exports from African countries prior to raising its standards.<sup>22</sup> While intraregional trade throughout Africa continues to be robust, aflatoxin contamination limits how much Africa can enter global export markets. As discussed earlier, experts estimate losses at more than \$400 million due to stringent EU standards alone. Without significant measures to hamper aflatoxin exposure of key crops such as maize, cereals, and peanuts, East African Community (EAC) exports cannot hope to overcome the key barriers to external trade which include compliance costs of rejected food and safety standards.

### F.2 U.N. WORLD FOOD PROGRAMME PURCHASES

In 2011, the U.N. World Food Programme (WFP) purchased 2.4 million metric tons (mt) of food worldwide amounting to \$1.23 billion. Top food products procured in 2011 include wheat (31%; 751.2 mt), maize (17%; 410.2 mt), and blended foods or cereals (14%; 350.0 mt). The WFP purchased 713,654 mt (\$305.2 million) in Africa. Total metric tonnage and value have been shown in **Figure 18**.<sup>23</sup> Researchers note that food purchases in Africa have proven to be cost-efficient as well as effective for supporting small-scale farmers.<sup>24</sup> The WFP standards and protocols in testing aflatoxin levels have been instrumental in raising awareness with farmers, traders, and governments regarding the prevalence of aflatoxin contamination and the need for policies and programs to address it.

Figure 18. Quantity and Value of Food Purchased from Select African Countries in 2011

Country	Quantity Metric Tonnage (mt)	Value (US\$ thousands)
South Africa	109,683	53,361
Tanzania	64,992	20,031
Kenya	57,961	22,867
Ghana	6,710	3,673
Niger	3,526	1,684

WFP purchases are an integral component of agricultural trade in developing regions of the EAC, particularly as agricultural standards on aflatoxin-prone food remain stringent in key global trade partners such as the United States and the EU. Compliance costs associated with the rejection of food products due to a failure to meet regulations can be significant. As the economy continues to waver, the question remains as to whether WFP will continue to meet increasing demands as the trending value of goods purchased has surpassed the trending quantity of goods purchased.



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## GLOSSARY

### **Aflatoxicosis**

The poisoning that results from ingestion of aflatoxins in contaminated foods or feed. Onset may be acute—due to a single, or a few moderate to high doses of aflatoxin—or chronic—due to the long-term exposure to aflatoxin-contaminated foods or feed. Symptoms of acute aflatoxicosis may include abdominal pain, vomiting, enlarged liver and/or liver damage, fever, hemorrhage, difficulty breathing, poor digestion, or convulsions. Chronic aflatoxicosis may result in liver damage, cirrhosis of the liver, liver cancer, and/or growth and developmental delays in exposed children.

### **Aflatoxin**

Aflatoxins are highly carcinogenic toxins produced by the fungus *Aspergillus flavus* that are commonly found in soils and infect grains, nuts, seeds and legumes. Aflatoxins have a negative impact on health and have been associated with liver cancer, growth retardation and stunting in children, suppression of the immune system, liver cancer—and more recently, linked with HIV and tuberculosis.

### **Ammoniation**

Ammonia is diluted, applied to crops in a water vapor, and allowed to permeate the feed. The ammonia destroys fungus or mold growth and has a cleansing effect on treated feed. Ammoniated feed is safe for animal consumption and can make feed more efficient when used in the short-term.

### **Biocontrol**

A method of inhibiting pests by disrupting their ecological status in the local environment through the introduction of natural organisms such as parasites or pathogens. An example of this is the introduction of a non-toxigenic strain of *A. flavus* to displace the toxigenic strain.

### **Carcinogen**

Any substance that is an agent directly involved in causing cancer due to its ability to damage or disrupt the cellular metabolic processes. Carcinogens may increase the risk of cancer by altering cellular metabolism or damaging DNA directly in cells, which interferes with biological processes. The cell alteration induces uncontrolled, malignant division, ultimately leading to the formation of tumors, and the DNA damage leads to the cell becoming a cancer cell.

### **Cirrhosis**

A condition in which the liver slowly deteriorates and malfunctions due to years of chronic injury. It is a consequence of chronic liver disease characterized by replacement of liver tissue by fibrosis, scar tissue, and regenerative nodules (lumps that occur as a result of a process in which damaged tissue is regenerated),

which leads to the loss of liver function. Cirrhosis is most commonly caused by alcoholism, hepatitis B and C, and fatty liver disease, but it has many other possible causes. Scar tissue replaces healthy liver tissue—partially blocking the flow of blood through the liver—and impairs the liver’s ability to control infections; remove bacteria and toxins from the blood; process nutrients, hormones, and drugs; make proteins that regulate blood clotting; and produce bile to help absorb fats—including cholesterol—and fat-soluble vitamins.

**Fumonisin**

A mycotoxin derived by the *Fusarium verticillioides* and *Fusarium moniliforme*. It occurs as Fumonisin B<sub>1</sub> and Fumonisin B<sub>2</sub>. **Fumonisin B<sub>1</sub>** occurs mainly in maize (corn), wheat and other cereals. Human exposure is greatest in regions where maize products are the dietary staple. Fumonisin B<sub>1</sub> causes increased apoptosis in the liver or kidney of fumonisin-treated animals followed by regenerative cell proliferation. While the acute toxicity of fumonisin is low, it is the known cause of two diseases which occur in domestic animals with rapid onset: equine leukoencephalomalacia and porcine pulmonary oedema syndrome. **Fumonisin B<sub>2</sub>** is more toxic than fumonisin B<sub>1</sub> and frequently contaminates maize and other crops.

**Mycotoxin**

It is a toxic secondary metabolite produced by organisms of the fungus kingdom, commonly known as molds. The term “mycotoxin” is usually reserved for the toxic chemical products produced by fungi that readily colonize crops. One mold species may produce many different mycotoxins and/or the same mycotoxin as another species. The six major groups of mycotoxins include aflatoxins, patuilins, fusariums, ochratoxins, citrinins, and ergot alkaloids.

**Toxigenic**

Something that produces poison or toxin; conversely, *non-toxigenic* is something that is not able to produce poison.











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