Aflatoxin analysis of dairy feeds and milk in the Greater Addis Ababa milk shed, Ethiopia

Dawit Gizachew, Barbara Szonyi, Azage Tegegne, Jean Hanson and Delia Grace

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Aflatoxins

- Toxic secondary metabolites produced by *Aspergillus* fungi
- Contaminate a variety of foods such as corn, oil seed and animal feed
- One of the most toxic forms of aflatoxin (AFB1) is converted to AFM1 and excreted in milk by lactating animals that consume contaminated feed
- Highly carcinogenic; cause liver cancer, stunting and immunosuppression

*Aspergillus flavus*  (Maize breeding program at Texas A&M University)
Effect of aflatoxins on human health

AFB1 is potent carcinogen. It can cause liver cancer

Liver of a rat fed with high doses of aflatoxin B1
Notice the induced tumours in the liver
(http://www.ansci.cornell.edu)
Aflatoxin Regulatory Guidance

FDA Mycotoxin Regulatory Guidance

A Guide for Grain Elevators, Feed Manufacturers, Grain Processors and Exporters

National Grain and Feed Association
1250 Eye St., N.W., Suite 1003, Washington, D.C., 20005-3922
Phone: (202) 289-0873 Fax: (202) 289-5388
Web Site: www.ngfa.org

August 2011

FDA’s Action Levels for Aflatoxin

FDA has established the following action levels for aflatoxins present in human food, animal feed and animal feed ingredients as indicated in Chart 1.

<table>
<thead>
<tr>
<th>Intended Use</th>
<th>Grain, Grain By-Product, Feed or other Products</th>
<th>Aflatoxin Level [parts per billion (p.p.b.)]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Human consumption</td>
<td>Milk</td>
<td>0.5 p.p.b. (aflatoxin M1)</td>
</tr>
<tr>
<td>Immature animals</td>
<td>Corn, peanut products, and other animal feeds and ingredients, excluding cottonseed meal</td>
<td>20 p.p.b.</td>
</tr>
<tr>
<td>Dairy animals, animals not listed above, or unknown use</td>
<td>Corn, peanut products, cottonseed, and other animal feeds and ingredients</td>
<td>20 p.p.b.</td>
</tr>
<tr>
<td>Breeding cattle, breeding swine and mature poultry</td>
<td>Corn and peanut products</td>
<td>100 p.p.b.</td>
</tr>
<tr>
<td>Finishing swine 100 pounds or greater in weight</td>
<td>Corn and peanut products</td>
<td>200 p.p.b.</td>
</tr>
<tr>
<td>Finishing (i.e., feedlot) beef cattle</td>
<td>Corn and peanut products</td>
<td>300 p.p.b.</td>
</tr>
<tr>
<td>Beef, cattle, swine or poultry, regardless of age or breeding status</td>
<td>Cottonseed meal</td>
<td>300 p.p.b.</td>
</tr>
</tbody>
</table>

The following additional policies and legal provisions concerning aflatoxin also are important:

- **FDA Blending Policy:** Importantly, with respect to aflatoxin, FDA currently generally does not permit corn containing aflatoxin to be blended with uncontaminated corn to reduce the aflatoxin content of the resulting mixture to levels acceptable for use as human food or animal feed. However, on occasion FDA has relaxed its “no-blending” policy in

EU

0.05 ppb

4 ppb
Study locations: the greater Addis Ababa milk shed

- Includes Addis Ababa, Debre Zeit, Sebeta, Sendafa and Sululta
- It serves as a major supplier in and around Addis Ababa
- The sector is commercial and uses concentrate feeding
Feed analysis of aflatoxin B1 (AFB1) using enzyme-linked immunosorbent assay (ELISA)
Study methods

• Study participants:
  • 100 dairy farmers
    • 27 from Addis Ababa, 23 from Debre Zeit, 9 from Sebeta, 31 from Sendafa and 10 from Sululta
  • 5 feed producers
  • 5 feed processors
  • 9 feed traders

• A semi-structured questionnaire was administered to all study participants

• 100 gram samples of each feed type were collected
All dairy farmers used concentrates every day to feed cattle of all ages

- Ingredients in concentrates feed include:
  - Wheat barn (100%)
  - Noug seed cake (73%)
  - Pea hulls (37%)
  - Maize grain (12%)

Noug cake

Pea hulls and wheat bran
Results of feed analysis

Summary of aflatoxin (AFB1) contamination of feed in the value chain (N=156)

<table>
<thead>
<tr>
<th>Value chain actor</th>
<th>AFB1 levels (ppb) in feed samples</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>&lt;5</td>
</tr>
<tr>
<td>Milk producer</td>
<td>a0</td>
</tr>
<tr>
<td></td>
<td>b0</td>
</tr>
<tr>
<td>Feed manufacturer and trader</td>
<td>a0</td>
</tr>
<tr>
<td></td>
<td>b0</td>
</tr>
</tbody>
</table>

aNumber of contaminated samples  
bPercentage of AFB1 contaminated feed samples  
cThe highest AFB1 concentration was 419 ppb from milk producer
Concentration of AFB1 (ppb) in individual dairy feed ingredients

<table>
<thead>
<tr>
<th>Sample</th>
<th>Mean ± SD (ppb)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Noug cake</td>
<td>362 ± 38</td>
</tr>
<tr>
<td>Wheat bran</td>
<td>15 ± 6</td>
</tr>
<tr>
<td>Maize grain</td>
<td>18 ± 11</td>
</tr>
<tr>
<td>Brewer's dry yeast</td>
<td>15 ± 4</td>
</tr>
</tbody>
</table>

The fate of wheat bran and noug cake in the peri-urban dairy value chain

Crop Farmer
- Grain producers (wheat)
- Oil seed producers (noug)

Factory
- Flour factory (wheat bran)
- Oil factory (noug cakes)

Processor
- Mixed dairy feed including wheat bran and noug cake

Dairy farmer
- Fed to cattle of all ages daily

(Traders)

(Traders)

(Traders)
Noug Seed (Guizotia abyssinica)

Beside its use as oil seed and animal feed, noug is sold in the local market for consumption.
Milk analysis of aflatoxin M1 (AFM1) using enzyme-linked immunosorbent assay (ELISA)
Conversion of AFB1 to AFM1

Animals, under the influence of the cytochrome P_{450} oxidase system found in their microflora and own cells, hydroxylate aflatoxin B$_1$ (AFB1) to aflatoxin M$_1$ (AFM1)

Figure 1: The Formation of Aflatoxin M$_1$ from Aflatoxin B$_1$
Study Methods

• Study participants:
  • 100 dairy farmers
    • 27 from Addis Ababa, 23 from Debre Zeit, 9 from Sebeta, 31 from Sendafa and 10 from Sululta
  • 10 milk collectors

• A semi-structured questionnaire was administered to all study participants

• 50 ml of raw milk samples were collected
Results of milk analysis from milk producers and collectors

Summary of aflatoxin (AFM1) contamination of milk in the value chain (N=110)

<table>
<thead>
<tr>
<th>Value chain actor</th>
<th>AFM1 levels (ppb) in milk samples</th>
<th>c1.00 &gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>&lt;0.05</td>
<td>0.05-0.10</td>
</tr>
<tr>
<td>Milk producer</td>
<td></td>
<td>48</td>
</tr>
<tr>
<td>Milk collector</td>
<td></td>
<td>3</td>
</tr>
</tbody>
</table>

|                   |        | 30        | 30        | 10        | 30       |

\(^{a}\) Number of contaminated samples  
\(^{b}\) Percentage of AFM1 contaminated milk samples  
\(^{c}\) The highest AFM1 concentration was 4.98 ppb from milk producer.

## Aflatoxin contamination level in milk in some African countries

<table>
<thead>
<tr>
<th>Country</th>
<th>Region</th>
<th>AFM1 (ppb)</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kenya</td>
<td></td>
<td>0.68</td>
<td>Kang'ethe and Lang'a, 2009</td>
</tr>
<tr>
<td>Nigeria</td>
<td>Ogun State</td>
<td>2.04</td>
<td>Atanda et al., 2007</td>
</tr>
<tr>
<td>South Africa</td>
<td></td>
<td>0.2-1.5</td>
<td>Dutton, 2003</td>
</tr>
<tr>
<td>Sudan</td>
<td>Khartoum state</td>
<td>0.22-6.9</td>
<td>Elzupir and Elhussein, 2010</td>
</tr>
</tbody>
</table>

Conclusion

- Noug (*Guizotia abyssinica*) cakes are widely used in the greater Addis Ababa milk shed as cattle feed and have been found to be contaminated with AFB1
- Intervention studies, including chemical detoxification of the feed, might be necessary to minimize the aflatoxin contamination along the value chain
- Together with the Ministry of Health, there is need to study aflatoxin in human urine and breast milk to examine the impact on human health
- Further studies are required on the impact of aflatoxin on human and animal health in Ethiopia
Acknowledgements

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better lives through livestock

ilri.org