Aspergillus fumigatus – A Threat to Healthy Cows
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Mycotoxins Impact Animal Health and Performance: Many livestock producers are aware of the common problems associated with molds and mycotoxins. In general, several species of molds (Aspergillus, Fusarium, and Penicilium) are known to produce common mycotoxins (aflatoxin, zearalenone, trichothecenes, vomitoxin and fumonisin) when growing conditions are adequate (organic nutrient source, temperatures ranging from 23 to 140°F, moisture levels >70%, pH 4-7 and oxygen >0.05%). These mycotoxins, or secondary metabolites of molds, may contribute to losses in performance, reduced growth rate, feed refusal, diarrhea, irregular estrous cycles, abortion, interference with disease resistance and immune function, and vomiting in various livestock species (CAST, 2003).

While there is considerable information in the public domain on the impact of mycotoxins on animal performance, until recently, little emphasis has been placed on understanding the direct effect that certain mold species may have on the development of mycoses or mold-related health disorders in livestock. Much of this knowledge has come from the ongoing investigation of the link between Hemorrhagic Bowel Syndrome (HBS), immunosuppression and the mold, Aspergillus fumigatus (AF).

The Dominant Infesting Fungus in Livestock
Aspergillus fumigatus (AF) is a ubiquitous, fast-growing, saprophytic fungus that sporulates abundantly, releasing thousands of airborne conidia from each conidial head. All humans inhale several hundred AF conidia per day.

Healthy people and animals rarely show adverse effects as the conidia are eliminated by the actions of the innate immune system (Latge, 1999). Immunosuppressed people are much more susceptible to invasive aspergillosis as a result of failure of the immune system to reduce and eliminate the pathogenic effect of the conidia. There has been a 14-fold increase in human aspergillosis over the past 15 years as a result of increased use of immunosuppressive therapies associated with cancer, AIDS and organ-transplant therapies (TIGR website; Latge, 1999).

Aspergillus fumigatus and aspergillosis can affect animals as well. It is the dominant infecting fungus in ruminant livestock (Jensen, 1989). Common entry points into the animal are the lungs and the gastrointestinal tract. In ruminants, AF may enter the GI tract at the omasum and Peyer’s Patch immune tissue. Placentitis and pneumonia are secondary infections occurring as a result of the spread of AF through the blood from the primary gastrointestinal lesions. Aspergillosis can account for up to 20% of bovine abortions (Sarfati, et. al., 1996).

McCausland, et.al., 1987, reported a 78% incidence of Aspergillus fungal hyphae in the placentas of aborted cattle, with abortions occurring between 6 and 8 months of gestation. Puntenney, et. al., 2003, found a high degree of correlation between the incidence of AF infections and HBS with the bleeding into the jejunum resembling observations in humans infected with AF. Sockett, et. al., 2004, has also reported a high degree of correlation between the incidence of HBS and the presence of AF.

The pathogenicity of AF is attributed to three primary virulence factors. Aspergillus fumigatus produces compounds called siderophores which compete with iron-binding proteins to steal iron from normal biological functions in the host to support fungal growth. The fungus also contains specific types of lipid compounds as cellular constituents which allow fungal organisms to effectively evade the immune system by inhibiting specific pathways responsible for activation of the complement and phagocytic defense mechanisms. Thirdly, AF produces proteases which facilitate the penetration of fungal hyphae from the site of infection (lungs, GI tract) to the surrounding tissues (Rhodes, et. al., 1992).
infection. In addition to the virulence factors described earlier which allow the organism to evade destruction and elimination from the body, AF in the hyphal form also produces a mycotoxin called gliotoxin. Gliotoxin has been shown to induce apoptosis (cell death) of lymphocytes and macrophages (Zhou, et. al., 2000). Gliotoxin has also been shown to go through a redox cycle, generating ROS which damage healthy cells in the organs of the host (Zhou, et. al., 2000). Stanzi, et. al., 2005, also demonstrated that gliotoxin interfered with the generation and activation of T lymphocytes. Low levels of gliotoxin target and kill antigen-presenting cells (monocytes and dendritic cells), resulting in impaired antigen presentation. Reduced or impaired antigen presentation could result in reduced antibody production and impaired adaptive immune system response to viruses, bacteria or vaccines.

**Aspergillus fumigatus Prevalence Advances**

Aspergillus fumigatus has been detected in a variety of feeds, forages and in the blood and tissues of dairy cows throughout the United States. In 2003, the Department of Animal Sciences at Oregon State University began offering testing of feeds, blood and tissues for the presence of AF DNA. The method used involves analysis of fungal DNA based upon the DNA sequences located within a number of unique regions of the AF genome. To date, it is the only quantitative assessment of AF commercially available. Over 1200 blood samples have been analyzed and 44% of these have tested positive (Table 1).

**Table 1: Summary: results from blood samples submitted for A. fumigatus (AF) DNA testing by region.**

<table>
<thead>
<tr>
<th>Region of US</th>
<th>Traces</th>
<th>Positive</th>
<th>Positive +</th>
</tr>
</thead>
<tbody>
<tr>
<td>NE</td>
<td>51.8</td>
<td>19.6</td>
<td>28.6</td>
</tr>
<tr>
<td>SE</td>
<td>29.3</td>
<td>32.3</td>
<td>37.2</td>
</tr>
<tr>
<td>MW</td>
<td>44.7</td>
<td>22.9</td>
<td>32.4</td>
</tr>
<tr>
<td>SW</td>
<td>51.9</td>
<td>33.1</td>
<td>15.0</td>
</tr>
<tr>
<td>NW/W</td>
<td>42.4</td>
<td>20.2</td>
<td>37.6</td>
</tr>
</tbody>
</table>

A. fumigatus DNA determined by real-time PCR by Oregon State University, Immunobiology Laboratory. Traces levels periodically detected in cattle, generally not of concern (0-1 GU x 10⁴/ml). Positive: cattle in upper 25 percentile of AF levels detected in cattle, and may be cause of concern (1.3 GU x 10⁴/ml). Positive +: cattle in the upper 10 percentile of AF levels detected in cattle, and are cause for concern (3>100 GU x 10⁴/ml).

**Table 2: A. fumigatus in silage from Texas-New Mexico dairies.**

<table>
<thead>
<tr>
<th>Silage</th>
<th># Samples</th>
<th>Top</th>
<th>Face</th>
<th>Slab</th>
<th>Bags</th>
</tr>
</thead>
<tbody>
<tr>
<td>Corn</td>
<td>64</td>
<td>135,107,243</td>
<td>2,515</td>
<td>19,655</td>
<td>217,355,304</td>
</tr>
<tr>
<td>Triticale</td>
<td>2</td>
<td>ns</td>
<td>ns</td>
<td>24,645</td>
<td>195,440</td>
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<tr>
<td>Sorghum</td>
<td>16</td>
<td>317,148</td>
<td>812</td>
<td>19,797</td>
<td>129,636</td>
</tr>
<tr>
<td>Wheat</td>
<td>13</td>
<td>2,797,964</td>
<td>1,701</td>
<td>1,031,470</td>
<td>ns</td>
</tr>
<tr>
<td>Alfalfa</td>
<td>7</td>
<td>ns</td>
<td>2,206</td>
<td>3,126</td>
<td>ns</td>
</tr>
</tbody>
</table>

*Ave. AF spore counts (spores/gram) by sample collection location*
Limit Exposure to *Aspergillus* through Silo Management

Following best management practices for proper storage and rotation of feed ingredients and ensiled forages is paramount to reducing exposure to AF growth. \( A.\ fumigatus \) is extremely tolerant of wide ranges in temperature, 20 to 50°C (68 - 122°F), and grows best in dry, hot conditions. Moisture content should fall between 68-72% for corn silage and between 64-68% for legume haylages going into bunker or drive-over silos. Silage pits should be rapidly filled and immediately packed for best results. Corn silages should reach a pH of 4.0 or less and legume forages should reach a pH below 4.5 for proper fermentation to occur. Bunker silos should be immediately and effectively covered to reduce exposure to air. The spoiled layer at the top of the silo should be carefully discarded. Water troughs and feed bunks should be cleaned regularly to reduce the potential for explosive mold growth (Puntenney, 2003).

**Management Practices to Reduce Molds and Mycotoxins**

- Manage commodities and silos to minimize or eliminate molds
- Improve feedbunk and pen management
- Feed effective fiber and soluble carbohydrates
- Increase cow comfort to reduce stress
- Adopt good nutrition practices for a positive impact on immunity and disease resistance
- Mitigate the risks associated with molds and mycotoxins by contacting your Prince Representative

**Strong Immune System the Best Defense**

Building the immune system is also another strategy to allow the dairy cow to effectively deal with the immune challenges associated with the inhalation or consumption of \( A.\ fumigatus \). Recent studies have measured significant changes in the expression of several immune markers associated with neutrophil migration and phagocytic activity when diets are supplemented with specific nutritional compounds. Wang, et. al. reported a significant change in immune status in ruminants fed these specific nutritional components as measured by L-selectin, a neutrophil-adhesion molecule responsible for movement of neutrophils into tissue being attacked by pathogen (Wang, et. al., 2004; Wang et. al., 2005).

Recently, Forsberg et. al., measured dramatic changes in three neutrophil killing mechanisms when these specific nutritional components were included in the diet of ruminants. These killing mechanisms are associated with a 65% (\( p<0.01 \)) change in phagocytic activity, a two-fold (\( p<0.01 \)) change in ROS or neutrophil respiratory burst activity and a significant difference (\( p<0.05 \)) in DNA NET extrusion (Forsberg et. al., 2007). Neutrophil killing mechanisms are necessary for the dairy cow’s immune system to recognize, trap and destroy pathogens, such as \( A.\ fumigatus \), bacteria or viruses.

Changing the activity of these immune mechanisms will impact individual cow health and overall herd productivity and profitability.
A Cow’s Healthy Immune System
Starts with Good Nutrition

Good herd health starts with good nutrition, and good nutrition is required to help maintain a healthy immune system in dairy cows.

When cows are under stress from the strains of production and reproduction, or molds and mycotoxins in feed and pathogens in the environment, their natural immune system goes to work fighting off those challenges. In an ongoing fight, the cow’s immune system itself can begin to weaken.

That’s why good nutrition is so important, including sufficient energy, fiber, vitamins, trace minerals and the special nutritional supplements your nutritionist or veterinarian may recommend.

Maintaining a Healthy Immune System can:
• Fight pathogens in the environment
• Fight the effects of molds and mycotoxins in feed
• Reduce somatic cell count
• Reduce cases of mastitis
• Reduce cases of metritis
• Reduce death loss and culls
• Help increase milk production
• Help improve reproduction

Literature Cited

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Through Dynamic Quality Assurancesm Prince Agri Products has earned the American Feed Industry Association Safe Feed/Safe Food certification for the production and delivery of a safe and quality feed supply for the growth and care of food producing animals. Company facilities at Bremen-IN, Marion-IA, and Quincy-IL hold valid certification under this program.