

## REPORT OF THE USAHA/AAVLD COMMITTEE ON ENVIRONMENT AND TOXICOLOGY

Co-Chairs: Wilson Rumbelha, MI

Larry Thompson, MO

David Ailor, DC; A. Catherine Barr, TX; Karyn Bischoff, NY; Tim Evans, MO; Francis Galey, WY; Tam Garland, TX; L. Wayne Godwin, FL; Ramesh Gupta, KY; Jeffery Hall, UT; Jeffrey Hamer, PA; William Hare, MD; Brent Hoff, ON; Steve Hooser, IN; Laurent O'Gene Lollis, FL; Randall Lovell, MD; Travis Mays, TX; David Meeker, VA; Gavin Meerdink, IL; Sandra Morgan, OK; Michelle Mostrom, ND; Lee Myers, GA; Eileen Ostlund, IA; Elizabeth Parker, DC; Robert Poppenga, CA; John Rathje, IA; John Reagor, TX; Jane Robens, MD; Nick Schrier, ON; Lori Smith, KY; Patricia Talcott, WA; Larry Thompson, MO; Gary Weber, MD.

The Committee met on October 1, 2011 at the Adam's Mark Hotel in Buffalo, New York from 3:30 p.m. to 6:30 p.m. There were 37 attendees present.

Dr. Larry Thompson opened the meeting at 3:40 pm, welcomed everyone, and handed out the meeting agenda. Larry then handed out attendance sheets.

Dr. Thompson indicated he was asked by Dr. Gary Osweiler, who has a family commitment and could not attend, to serve as the USAHA co-chair for the 2011 meeting and to serve as the USAHA co-chair for the remainder of Gary's term, with the approval of the USAHA president.

Dr. Thompson indicated that at the end of this meeting there will be elections for a new scribe/secretary to take over for Dr. Randall Lovell immediately after the 2011 meeting and for a new AAVLD co-chair to take over for Dr. Rumbelha after the 2012 meeting.

Dr. Wilson Rumbelha then opened the floor for questions to Dr. Renate Reimschuessel, FDA/CVM. Renate had just presented a talk in the same room entitled "Vet-LRN – Center for Veterinary Medicine's Laboratory Response Network Focusing on Animal Feeds and Drugs" as part of the AAVLD Toxicology Scientific Session. Instead of repeating this talk to the same audience, Renate answered many questions from attendees at the USAHA/AAVLD Committee on Environment and Toxicology as part of her year 1 update on Vet-LRN. These questions included, but were not limited to, the possible purchase and/or lease of equipment for use by participating laboratories and the development of standardized protocols by the labs. Renate encouraged everyone to contact her at [Renate.Reimschuessel@fda.hhs.gov](mailto:Renate.Reimschuessel@fda.hhs.gov) if anyone had additional comments, concerns or questions or if they wished to submit a proposal.

Dr. Tim Evans, Veterinary Medical Diagnostic Laboratory, University of Missouri, Columbia presented a talk entitled Practical Aspects of Ergot Contaminated Feeds. Tim indicated that ergotism is an ancient disease caused by various alkaloids, especially ergopeptine alkaloids, produced by *Claviceps purpurea*. This fungus produces ergot bodies (sclerotia) that are found on many grasses (including, but not limited to, fescue, brome and timothy) and many cereal grains (especially wheat, rye, barley and oats, but NOT on corn). These ergot bodies vary widely in size and shape with the largest ones being found on barley.

Clinical signs in animal species are due primarily to vasoconstrictive and hypoprolactinemic effects of the alkaloids and often include decreased milk production (even agalactia) and dry gangrene of the extremities (feet, tongue, comb, etc.). The mare is one of the most sensitive animals to the alkaloids and agalactia (or decreased milk production) is a hallmark clinical sign. Clinical signs in cattle are dependent on environmental temperatures with summer slump (increased body temperature and decreased production parameters) occurring during hot weather and fescue foot (sloughing of hoofs and/or feet) occurring during cold weather.

Dr. Evans indicated there is anecdotal evidence of adverse effects in mares (agalactia or decreased milk production) when the total dietary concentration of ergopeptine alkaloids exceeds 50 ppb. There is anecdotal evidence of adverse effects in other livestock when the total dietary concentration of ergopeptine alkaloids exceeds 100 to 200 ppb. The ergopeptine alkaloid concentration of ergot bodies varies widely from ~0.01% to 1% with the weighted average of ~0.2% to ~0.3%. Thus, if ergot bodies comprise 0.1% of the complete ration, this equates to approximately 0.1 ppm (100 ppb) to 10 ppm (10,000 ppb) in the feed with a

likely mean of about 2-3 ppm (2,000 to 3,000 ppb). Ergot alkaloids in animal feeds are measured at the University of Missouri using an HPLC-MS method.

Dr. Evans also discussed the life cycle of the fungus (including both the sexual and asexual reproductive stages) and the ways to best prevent/control this disease (deep tilling of the soil, awareness of clinical signs, screen harvested grains and avoid use of these grain screenings in feedstuffs, etc.). Lastly, Tim indicated that ergot alkaloids have not been demonstrated in milk, but do have the potential to bioaccumulate in fat.

Dr. Wilson Rumbelha then opened the floor for the reporting of mycotoxin results from the states. He reminded attendees of the preliminary results provided prior to the meeting from Alabama, California, Colorado, Idaho, Illinois, Indiana, Iowa, Kansas, Kentucky (both the Breathitt Veterinary Center in western KY and the University of Kentucky in Lexington, KY), Missouri, Nebraska, New York, North Dakota, Oklahoma, Pennsylvania, Texas, Wisconsin, Wyoming and Ontario, Canada.

Dr. Michelle Mostrom, North Dakota State University, also reported finding 50 ppm of deoxynivalenol (DON) in wheat straw and up to 20 ppm DON and 1-2 ppm of zearalenone in corn silage this year. Although high levels of aflatoxins (700 – 800 ppb) in southern corn (only the general location of the state where the corn was harvested from was provided on the accession) have been found by her lab, no affected animals have been reported. Much of the corn crop in the upper Midwest has not been harvested so everyone needs to realize that the preliminary results presented at this meeting may change as more data becomes available.

Texas VMDL has reported levels of up to 1,373 ppb of aflatoxins, although only 19 of 80 tests from 56 accessions (mostly in corn) were positive for aflatoxins. The Office of the Texas State Chemist tested 594 samples from a variety of matrices for aflatoxins with the highest level found being 768 ppb.

Missouri has recently found high levels in aflatoxins in the 2011 crop. Illinois recently found 1,300 ppb aflatoxins (B<sub>1</sub>+B<sub>2</sub>+G<sub>1</sub>+G<sub>2</sub>) in a corn sample and 8 ppb (aflatoxin M<sub>1</sub>) in a milk sample from a dairy.

Corn gluten meal has been found with 3-7 ppm of zearalenone without any DON. It is unclear if this is due to concentration of this mycotoxin during processing or represents elevated levels of zearalenone in the corn used to produce the corn gluten meal.

DON was recently found in grain at 15-20 ppm from a Midwest state.

Finally, the 500 ppm vomitoxin (DON) reported by Kentucky (Western) in six feed samples was incorrect and should have been 5 ppm.

Dr. Rumbelha indicated we were done with all the non-business items and recessed the meeting for a 10 minute break at 4:45 p.m.

Dr. Thompson called the meeting back to order at 4:56 p.m.

Dr. Bob Poppenga, California Animal Health & Food Safety Laboratory, then provided the Report of the Mission Statement Subcommittee. This subcommittee was chaired by Dr. Poppenga and its members included Drs. Dwayne Hamar, John Reagor and Larry Thompson. After discussion at teleconferences prior to this meeting, the subcommittee developed the following proposed mission statement:

The mission of the USAHA/AAVLD Joint Committee on the Environment and Toxicology is to provide the USAHA and AAVLD with guidance, based upon sound scientific principles, on chemical residues that negatively affect or have the potential to negatively affect animal health, food or feed safety or the environment.

After Dr. Poppenga led discussions on this proposed mission statement for several minutes, a consensus was reached by the 27 members still attending the meeting for the following new mission statement of this joint committee:

The mission of the USAHA/AAVLD Joint Committee on the Environment and Toxicology is to provide guidance, based upon sound scientific principles, on chemical residues and toxicants that negatively affect or have the potential to negatively affect animal or public health, food or feed safety, or the environment.

Dr. Steve Hooser, Animal Disease Diagnostic Laboratory, Purdue University, then provided the Report of the Proficiency Testing Subcommittee. This subcommittee was co-chaired by Dr. Hooser and Dr. Cat Barr and its members included Drs. Jeff Hall, Walt Hyde, Bob Poppenga, and Nick Schrier. This subcommittee had two conference calls prior to the meeting.

Dr. Jeff Hall, Utah State University, has lyophilized and prepared for shipping a bovine liver from a lead poisoning case. This liver will be sent to Dr. Hyde at NVSL (National Veterinary Service Laboratory) who will then ship the samples to the participating labs. The subcommittee asks the participating labs to analyze the liver for as many elements as possible and send their results back to Dr. Hyde at NVSL who will prepare a report of all the findings.

It was reported that NVSL may have up to \$30,000 to help fund parts of this proficiency testing (PT) for the next year. The PT Subcommittee will be preparing a report on the costs related to the above proposed proficiency testing (acquiring tissues, preparing tissues for shipping, analyzing samples for homogeneity, shipping to participating labs, correspondence with labs, &/or preparing final report, etc.) and will submit a proposal to NVSL in hopes of securing funds that may be available. The co-chairs of this subcommittee were asked to send a letter of thanks to Dr. Beth Lautner at NVSL for her past support of our PT program.

If funding from the NVSL is not obtained, then participating labs will likely need to pay for this proficiency testing services by subscription and this subcommittee will need to develop plans to collect and utilize these funds.

Ideas for inclusion in future proficiency testing included the following:

Nitrates in forage	Vitamin E and selenium in liver and serum
Metals in animal feed	Insecticides in liver
Anticoagulants in liver	MGA in animal feed
DON in wheat straw	Nitrates in ocular fluid
Alkaloids, especially strychnine and nicotine, in gut contents and/or urine	

It was noted that additional testing on the stability of nitrates in ocular fluid will need to be conducted before this fluid could be seriously considered and that AAFCO already has a proficiency testing program for elements in animal feed.

The PT Subcommittee currently has contacts in 33 labs performing toxicology testing in animal tissues/fluids or animal feeds. Putting information about the toxicology PT program on the AAVLD list serve was believed to be a good public relations move.

All aspects of the PT program described above, including ways to obtain long term funding from NVSL and/or Vet-LRN and ways to rotate who prepares samples for the PT program, will be discussed at the toxicology retreat in Canada.

Dr. Jeff Hall was unable to attend the meeting so Dr. Wilson Rumbelha presented slides and led the discussion on Survey Results of State Reporting Requirements for Toxicology. Wilson believes there has been a good overall response to the survey that was sent to all 50 states by Jeff on July 28, 2011, but does not know exactly how many states have responded.

Dr. Wilson Rumbelha was copied on the survey responses from 10 states and believes there are no reporting requirements for toxin-induced diseases in many states. There are considerable differences in the reporting requirements for toxin-induced diseases between states with such requirements. Wilson showed or discussed the language for the reporting requirements for toxin-induced diseases in Michigan, Iowa, Utah, Maine, and North Dakota. Dr. Cynthia Gaskill indicated that Kentucky has recently added toxicants to the list of reportable diseases.

Following discussion, there was a general agreement that this committee should work on developing model language for the reporting of toxin-induced diseases that provides broad authority, but is not too intrusive. Issues related to confidentiality of client records, enforcement of the laws, and the large number of reportable diseases already on the books will need to be considered when writing this model language. The committee will also need to be transparent and open and work closely with the AVMA (as this organization is re-writing their Model Practice Act) and with State Veterinary Medical Associations & State Departments of Agriculture (who often coordinate and implement any animal emergency responses).

### **Committee Business**

The action items from this discussion were as follows: Dr. Jeff Hall or a committee designee will tabulate the results from the 50 state survey and report back to the committee in 2012. The committee will communicate its concerns about the lack of reporting requirements for toxin-induced diseases in many states to the USAHA and AAVLD. The committee will explore the development of model language for the reporting of toxin-induced diseases.

Dr. Larry Thompson opened the floor to any new business. Nominations were made and seconded for the Scribe/Secretary and incoming AAVLD Co-Chair. By unanimous voice votes Dr. Cynthia Gaskill was elected as the new Scribe/Secretary and Dr. Tim Evans was elected as the new incoming AAVLD Co-Chair and will take office after the 2012 meeting.

A motion to adjourn was made by Dr. Tam Garland and it was seconded and unanimously approved by voice vote.

**USAHA/AAVLD COMMITTEE ON ENVIRONMENT AND TOXICOLOGY  
MYCOTOXIN ROLL CALL 2011**

**Alabama: John Roberts**

Last year from October 2010 until present, at the Alabama Diagnostic lab (Thompson Bishop Sparks State Diagnostic lab) we had: Aflatoxin tested in 24 feed samples and there were 6/24 with levels between 4-10 ppb and one sample of pig feed 1/23 with a level of 28 ppb.

We tested two dog feed samples for DON and it was not detected above 0.5 ppm in any sample.

We tested 4 samples of feed with corn for fumonisins and had 4/4 had detection of low level between 0.76-2.0 ppm.

Zearalenone was not requested in that time period.

**California: Elizabeth Tor**

Some positive feed samples for DON, 1-2ppm. Few cases with stomach contents and urine tested positive for both: Penitren A and Roquefortine C.

**Colorado: Dwayne Hamar**

No report of suspected mycotoxins.

**Idaho: Patricia Talcott**

Not much from the PNW, to my knowledge. Few perennial ryegrass staggers, fescue and ergot cases.

**Indiana: Christina Wilson**

Positive mycotoxins for the past year (number of samples positive at that level):

DON: 1 ppm (7), 2 ppm (3), 4 ppm (1), 5 ppm (2), 8 ppm (1), 10 ppm (2)

Zearalenone: 1 ppm (4), 2 ppm (1)

**Illinois: Gene Niles**

We're not seeing any so far but I'm expecting plenty of aflatoxin and fumonisin with the dry weather here.

## **Iowa: Paula Imerman**

	# tests	# positive
aflatoxins	50	0
vomitoxin	32	9
ochratoxin	32	0
fumonisin	12	12
T2 toxin	32	0
zearalenone	32	1
aflatoxins-liver	4	0

## **Kansas: Deon van der Merwe**

We did not have many mycotoxin cases the past year in Kansas, but we had some interesting ones.

We received several (around 10) calls regarding ergot in wheat during early to middle July. Some of the calls were related to wheat that had been refused by buyers because of the presence of ergot sclerotia. My impression was that most of the problems occurred in Nebraska or close to the Kansas Nebraska border. We recommended that samples be sent to Missouri VMDL for ergot alkaloid testing.

I had a call last week related to aflatoxin in corn in North-Central Kansas that was reported to contain 40 ppb (the test was not performed by us), and refused by a buyer. We have not had positive aflatoxin test results in samples submitted to us thus far this year. Many producers are concerned, however, due to the poor state of the corn crop in many areas.

A corn-based horse feed sample tested positive for Fumonisin (15 ppm) in early June.

There was an interesting case of penitrem A poisoning in two dogs that had consumed garbage in November 2010. The dogs displayed "classic" signs of strychnine poisoning, but no strychnine was identified. 25-100 ppm penitrem A was found in vomitus samples. Both dogs died.

## **Kentucky (Western): Ramesh Gupta**

From Breathitt Veterinary Center in Western Kentucky:

Six feed samples out of 15 samples contained aflatoxin B1 (13-50 ppb) and aflatoxin B2 (2-4 ppb).

In another case, six feed samples contained vomitoxin (500 ppm).[verify]

## **Kentucky: Cynthia Gaskill**

We have not had much in the way of feed mycotoxins this year. We send our feed samples to North Dakota for analysis. I expect we will be getting more samples submitted in the next few months. We had a very late planting season here because of all the heavy rains this spring. We have had a few tremorgenic mycotoxin (penitrem A and Roquefortine) cases in small animals.

## **Missouri: Tim Evans**

Ergots---149 samples, 74 positives MANY OF THESE WERE FROM PACIFIC NORTHWEST  
Ergot/ergovaline---36 samples; 1-ergot only, 1 ergot and ergovaline, 6 ergovaline only for a total of 8 positives. (MANY OF THESE WERE OUT OF STATE).

Ergovaline---9 samples and 2 positives

Mycotoxin screen---153 samples; 10 vomitoxin and zearalenone, 43 vomitoxin only, 2 zearalenone only for a total of 55 positives. MANY OF THE POSITIVE MYCOTOXINS (VOMITOXIN AND ZEARALENONE) CAME FROM OHIO.

Aflatoxin---41 samples and 6 positives MOST OF THE POSITIVES WERE FROM CORN GROWN IN TEXAS.

Vomitoxin---5 samples and 3 positives MAINLY MISSOURI

Fumonisin---16 samples and 12 positives CURRENTLY DON'T HAVE FEEL FOR DISTRIBUTION.

Aflatoxin M1---4 samples and no positives

Oosporein---52 samples and no positives

Ochratoxin---8 samples and no positives

## **Nebraska: Michael P Carlson**

Ergot contamination of the 2011 winter wheat crop was greater than usual. Seems to be more prevalent along the KS-NE border. We are in the process of having some specimens analyzed for ergot alkaloid content.

According to the extension plant pathologist here at UNL with whom I have worked on the ergot problem, the incidence of wheat scab and concurrent DON and zearalenone contamination in the 2011 NE wheat crop was not extraordinary, even though the weather conditions during the spring and early summer that favored *Claviceps* infection should also have favored *Fusarium* infection, too. Supporting that notion, I got no calls about Fusarium-related problems, just calls about ergot.

**New York: Karyn Biscoff**

We got definitive diagnoses on at least 2 cases of penitrem toxicosis, affecting about half a dozen dogs altogether, back in November and December of 2010. We aren't seeing anything right now, though there are rumors of "mycotoxin in barley" but I think that's due to some positive tests for DON coming out of PA and the Midwest. We haven't actually seen any DON in barley here.

**North Dakota: Michelle Mostrom**

North Dakota has just started harvesting wheat – low levels of DON in some Red River Valley wheat (< 1 ppm). No corn harvest started.

Low level of DON (<2 ppm) and zearalenone (0.5 to 1.0 ppm) in corn silage from some Midwest states.

We have found high concentrations of aflatoxin in southern corn (up to 700 to 800 ppb aflatoxins). Fumonisin in corn harvest have been low, with 18 ppm the highest that we tested from southern corn. Sorry that I can't be specific on states, but some samples come in from other companies and only know general location.

**Oklahoma: Sandra Morgan**

We lost a lot of our corn crop due to drought, but the corn we are testing has ranged from 0-570ppb aflatoxin with an average of 200ppb. The Fumonisin tests we have run have been low so far. There have been some suspected ergot and tremorgenic mycotoxins cases, but no confirmation.

**Ontario, Canada: Brent Hoff**

In Ontario 79 samples of wheat were tested for DON

- 1) 3 > 6.00 ppm
- 2) 6 4-6 ppm
- 3) 8 2-4 ppm
- 4) 6 1-2 ppm
- 5) rest <1.00 ppm

**Pennsylvania: Lisa Murphy**

We don't really get enough samples to comment on PA

**Texas (College Station): Tam Garland**

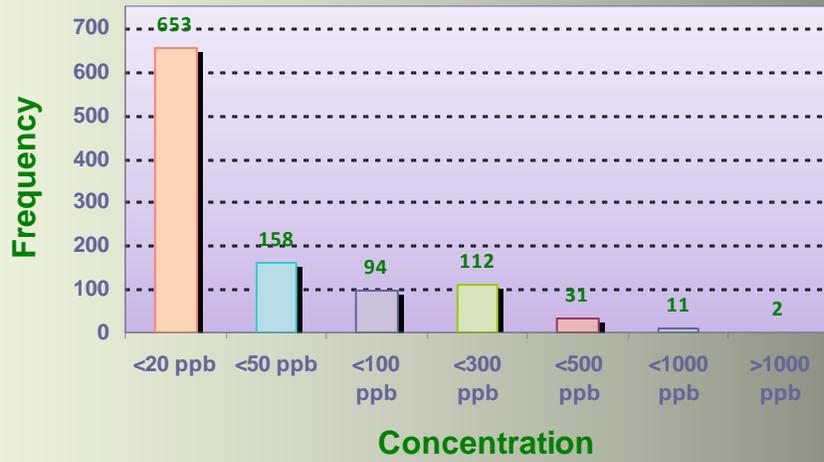
Since 1 Jan 11 we have done 56 aflatoxin accession but 80 test of which 19 were positive for aflatoxin. Most of samples were corn but we had a few dog foods which where thankfully negative. Our samples ranged from 20 ppb to 1,373 ppb. In addition to our samples the Office of the Texas State Chemist (a feed regulatory arm) has tested 594 samples for aflatoxin in a variety of matrixes with numbers ranging from none detected to 768 ppb.

Since 1 Jan 11 we have had 18 fumonisin accessions but 27 test, of which 4 were positive for fumonisin. Most of our samples were feed, but we did have one case that insisted on testing dog food, which was negative. Our samples ranged from none detected, to 5 ppm and up to 157 ppm.

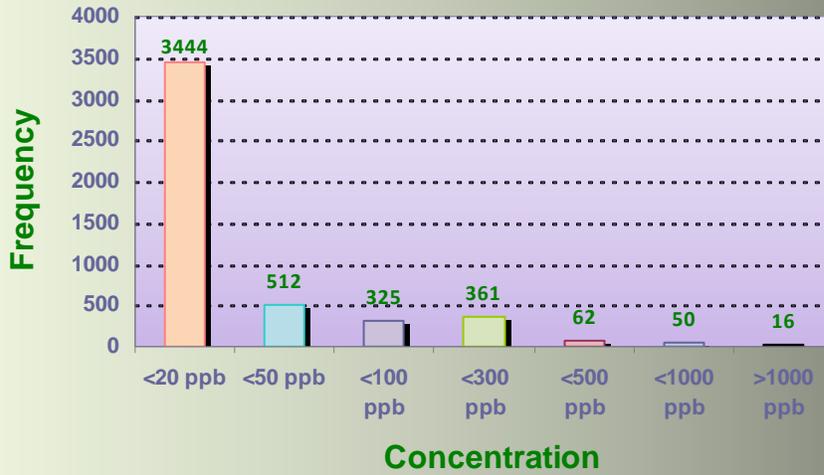
In addition to our samples, the Office of the Texas State Chemist (a feed regulatory arm) has tested 527 samples for fumonisin with numbers ranging from none detected, and between 1 and 16 ppm.

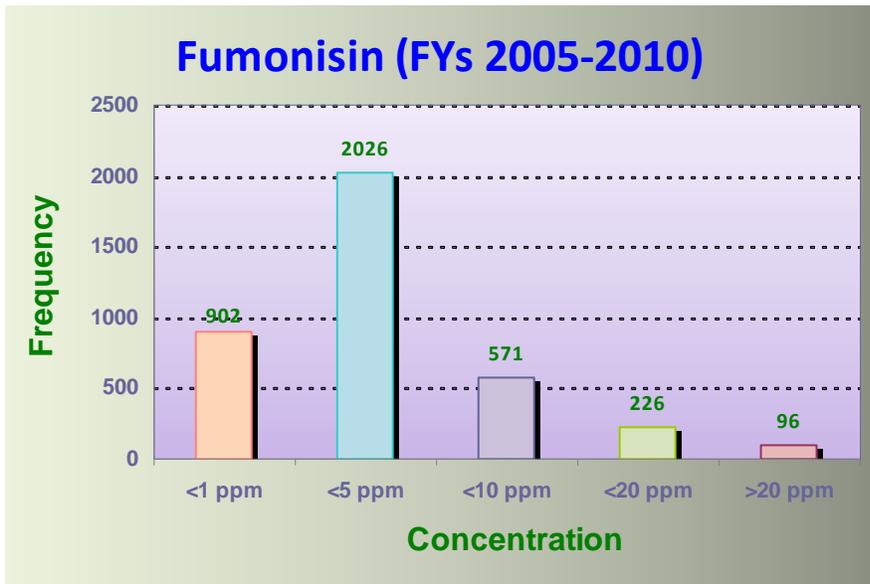
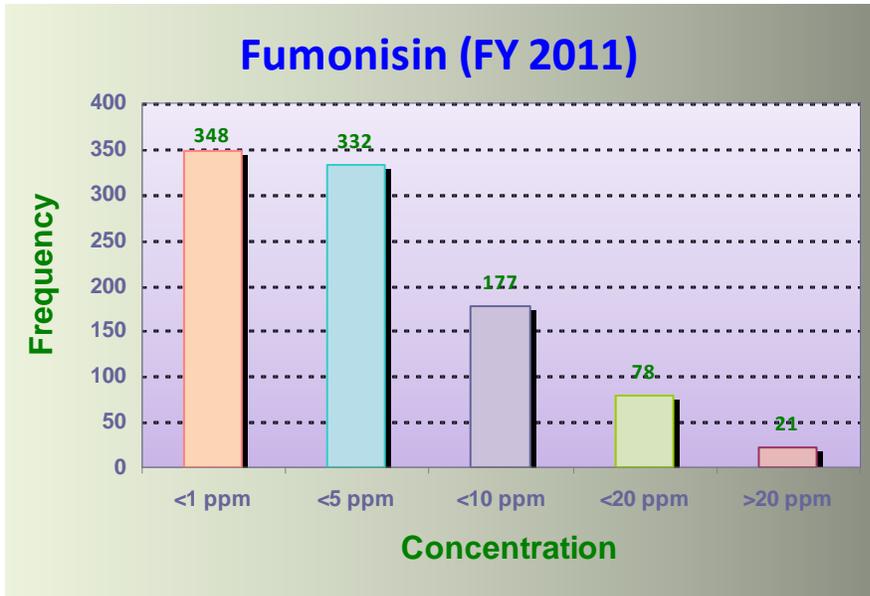
**Texas (Texas State Chemist FY 2011 and FY 2005-2010): Lynn Post**

## Aflatoxin (FY 2011)



## Aflatoxin (FYs 2005-2010)





**Wisconsin: Dave Zoromski**

Wisconsin has not seen much in the way of mycotoxins submitted to the laboratory. I believe that most owners and veterinarians are sending samples directly to North Dakota or Iowa to avoid our accession and shipping fees. In the past year, we have only had 6 submissions that we forwarded to North Dakota. Of these, 5 had levels of DON (0.5, 1.4, 1.5, 1.6, 2.2 ppm). The Department of Agriculture, Trade and Consumer Protection may have additional test information. I will contact them to see what they may have done.

**Wyoming: Merl Raisbeck**

Haven't heard of any in our area.