AgriLIFE EXTENSION
Texas A&M System

Sampling For Aflatoxin

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Background:

In a field of corn, aflatoxin contamination is not uniformly distributed throughout the field. There are usually areas of the field with higher levels of contamination. However, not all ears in this area will be contaminated and not all kernels of a contaminated ear will have the same degree of contamination. Kernels of a contaminated ear could have concentrations of aflatoxin ranging from 0 ppb to 400,000 ppb, with most kernels having no or very low amounts of aflatoxin.

This distribution of aflatoxin contamination in kernels in the field accounts for the difficulty in accurately determining the aflatoxin concentration of a grain load. Typically, a small percentage of kernels are contaminated with aflatoxin though individual kernels often have extremely high concentrations. For example, a single kernel contaminated with 400,000 ppb aflatoxin in a 10-lb corn sample causes that sample to have an overall concentration of 26 ppb aflatoxin. This skewed distribution of contamination can account for the variability often seen in sampling loads of grain. If a shipment is tested several times, different measurements will be obtained, and often with results having a wide range of values (i.e., variance).

Researchers have found that if aflatoxin contamination is relatively high, a high variance will occur among samples, and many values will be higher than the average. For example, a lot of cottonseed known to have aflatoxin contamination was tested 20 times. The average value was 74 ppb and the range of values was 15-160 ppb. Two of the values were less than the threshold of 20 ppb, and nine of the values were greater than the average. In contrast, the average of another lot was 2.7 ppb, with a range of 0-14. Aflatoxin was not detectable in nine of the samples, while five exceeded the average.

Sampling variability can be reduced by increasing sampling size, or by increasing numbers of sampling units. However, the disadvantage with increasing sample size are increased sampling costs and increased sampling time. Samples for analysis should be 5-10 pounds.

In addition to sampling variability, two additional sources of variability affect final measurement of aflatoxin concentrations, namely, subsampling variability and analytical variability. These latter sources of variability are not as great as sampling variability, and they are easier to minimize than sampling variability.

Subsampling variability refers to the smaller portion of the sample that is actually extracted and analyzed. With corn, the entire sample (usually 10 lb.) must be ground. If the particle size is small enough and the ground corn is sufficiently mixed, variation of aflatoxin among subsamples will be relatively small. The more particles

per unit mass, the smaller the subsampling variability. Established sample preparation protocols should be followed to minimize this variability.

Analytical variability refers to the actual sample analysis. Different steps in the analysis (e.g., extraction, purification) can also contribute to variation. Research shows that variance for different analytical methods is a function of the aflatoxin concentration in the subsample. Established analytical protocols should be followed closely to minimize analytical variability.

How to Obtain a Representative Sample for Analysis:

Proper sampling is critical to obtain accurate analytical results. "Grab" samples (e.g. scooping the top of truck) are useless. Sampling from a moving stream (i.e., continuously removing grain samples while the shipment is being loaded or unloaded) is better than static sampling (i.e., taking a sample from grain on a truck or stored in a bin).

For static sampling, grain should be sampled from different locations in the lot. Distribution of contaminated kernels within a given lot will likely be uneven, consisting of pockets of contamination from (1) grain harvested from highly-contaminated portions of the field; (2) redistribution of lighter, more contaminated grain; or (3) post-harvest aflatoxin production caused by improper storage. A 1-lb sample should be removed for each ton of grain. If the amount of sample is greater than 10 lb, the grain should first be thoroughly blended and then the required amount removed. A riffle divider can be used to reduce the size of a given sample. A double-tube compartmented grain probe should be used to sample a container of grain, and the probe should reach to the bottom of the container. The number of probing points increases with the lot size, and each probe should sample a different portion of the container. Specific recommendations for sampling are outlined in the USDA Grain Inspection Handbook.

NOTE: this handbook is available in PDF format from the following website: www.gipsa.usda.gov/GIPSA/webapp?area=home&subject=landing&topic=landing

For sampling from a moving stream of grain, small samples are taken at pre-determined time intervals during the time the grain is moving. Commercial devices are available for this kind of sampling.

Sampling guidelines are available, e.g., the FAO Technical Consultation on Sampling Plans for Aflatoxin Analysis in Peanuts and Corn (FAO Food and Nutrition Paper 55). This document also recognizes the limitations of sampling for detection of aflatoxin. As an example, Table III-4 in this document lists the probability of accepting corn lots with concentrations of aflatoxin from 0-130 ppb using five different acceptance levels and two sample sizes. The table shows that as the lot concentration of aflatoxin increases, the probability of accepting the lot decreases. If the lot concentration is below the threshold, there is still a probability of rejecting it. However, the probability of making the correct decision increases with an increasing sample size. It also is clear that the probability of making a correct decision increases with repeated sampling. For example, a lot with 22 ppb has a 54% chance of being rejected, but with one additional sample test, this chance increases to 80%, and with yet another sample test, it increases to 90%.

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