

How to take grain and feed samples for mycotoxins detection



Why is it difficult to identify mycotoxins in grains and feed?

A common concern for nutritionists, veterinarians and technical personnel working in animal production is the difficulty in identifying mycotoxins in the feed consumed by animals showing typical lesions caused by mycotoxins. Below, we are listing the most important causes for this problem:

- ① **Uneven distribution of mycotoxins (grain and feed).**
- ② **Sampling errors.**
- ③ **Laboratory techniques used.**
- ④ **Presence of conjugated or masked mycotoxins.**

1. Uneven distribution of mycotoxins. Unlike the protein or moisture content present in corn or soybeans, mycotoxins are not evenly distributed. The main reason is that fungi do not grow everywhere, they do it in specific places. This trend can start in the field. Some grains can contain high levels of mycotoxins while others do not. The same phenomenon can occur in trucks, ships, and other compartments where grains or feed are stored.

2. Sampling. A correct analysis means the determination of the average contamination of the batch of grain or finished feed are evaluated. If proper sampling procedures are not followed, analytical results are likely to underestimate the true mycotoxins concentration. That is, if only areas with low or no contamination are sampled, no real results will be obtained. False negative results can occur in the tests, which is the result of inadequate sampling and poor preparation of the sample to be analyzed. When few incremental samples are taken or the total lot sample is too small, it is much more common to "lose" a contaminated grain than to "find" it.

The chances of identifying mycotoxins in feed increase if they are taken from the feeders located inside the farm because that feed has been stored inside the farm silos for about 5 to 7 days, and close to a day in the feeders inside the house where animals are raised. The more time feed is exposed to the environmental conditions (high moisture and temperature) present in a farm, the more likely it is to identify mycotoxins.

3. Lab techniques. Results may vary depending on the type of test. Some tests are more specific than others with the ability to detect lower levels of mycotoxins (higher sensitivity). For example, HPLC (high precision liquid chromatography) has a higher sensitivity than Elisa (enzyme-linked immunoassay). In other words, it is capable of detecting lower levels of mycotoxins in the sample. In Elisa's case, the possibility of false positives is higher than in HPLC.

There are tests that are more specific for the detection of mycotoxins, among them is HPLC. Techniques as liquid chromatography coupled to tandem mass spectrometry (LC / MS / MS) have been recommended widely because of its specificity and speed, among other characteristics. LC/MS/MS is highly advanced and simultaneously analyzes hundreds of metabolites and mycotoxins, something that cannot be done with HPLC or Elisa.

4. Conjugated or masked mycotoxins.

This represent a new challenge that makes it difficult to determine the real concentration of these toxins in the food that humans and animals consume. They consist of derivatives of mycotoxins not detectable with conventional analytical tests because the chemical structure of the mycotoxin has changed during the growth of the plant in the field, before the grains are harvested. The agents that catalyze the development of these variations in the integrity of mycotoxins are enzymes produced by plants that commonly act in the detoxification process. The mycotoxins adhere to other nutrients such as sugars (glucose), fatty acids or amino acids.



Where to take samples

To determine which ingredients are contaminated or when mycotoxins levels increase during feed preparation in a feed mill, it is necessary to take samples in the following areas:

- ① Reception area (raw materials).
- ② After grinding grains and before mixing grains.
- ③ Storage warehouse, once the feed has been prepared and stored.
- ④ Feeders (in the farm)

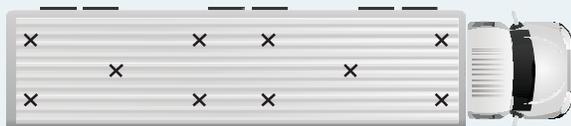
Taking samples from trucks or railroad cars that transport the grains (corn in this case) to the feed mill.

- Use a bulkhead gauge or probe with a minimum length of 1.5 m, with a minimum of ten compartments.
- Take samples in the areas marked by X in the truck's diagram shown below. Probe should go as deep as possible to obtain product from multiple layers of the load.
- The total weight of all the samples taken must be at least 10 Kg for trucks with a loading capacity of 20 to 50 MT and for trucks between 13 to 20 MT, the total of all the samples taken must be 6 Kg.
- From the total of samples taken, 21 subsamples of 200 g should be taken.
- In very modern plants there are pneumatic and automatic probes that take the samples directly.
- The more sample points we have when sampling, the more accurate the results will be.

Sampling points

Size of the composite sample, following the recommendations of the European Union.

Loads from 20 to 50 MT = 10 Kg of composite sample

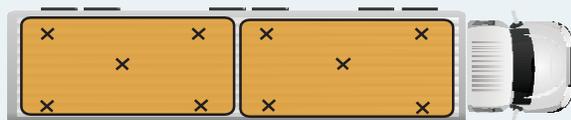


Loads from 13 to 20 MT = 6 Kg of composite sample



To reduce sampling variability:

- ① Collect 2 composite samples per truck.
- ② Weight of each composite sample = 5 Kg.



5 Kg

5 Kg



▶ Sampling at the reception area (grains and raw materials)

- 21 samples of 200 g each are taken, for a total of 4,200 g.
- The 4,200 g are mixed and split in half (2,100 g).
- Take these 2,100 g, mixed them again and take half (1,050 g).
- Out of the 1,050 g obtained, take 500 g.
- These 500 g are sent to the lab to run the test.

Taking "dynamic" or moving material samples within the feed mill during the manufacturing process.

- ① Drill a hole with a drill of approximately 1.5 inch in diameter in the bottom of the conveyor that carries the ground raw material to the storage tanks for mixing.
- ② A pipe with a stopcock is connected to this hole to keep it closed when no sample is taken. The tubing must end in a container that is removed at the time the sample is taken.
- ③ When the raw material is passing, the tap is opened and product is dropped, preventing it from spilling out of the container used to collect the sample.

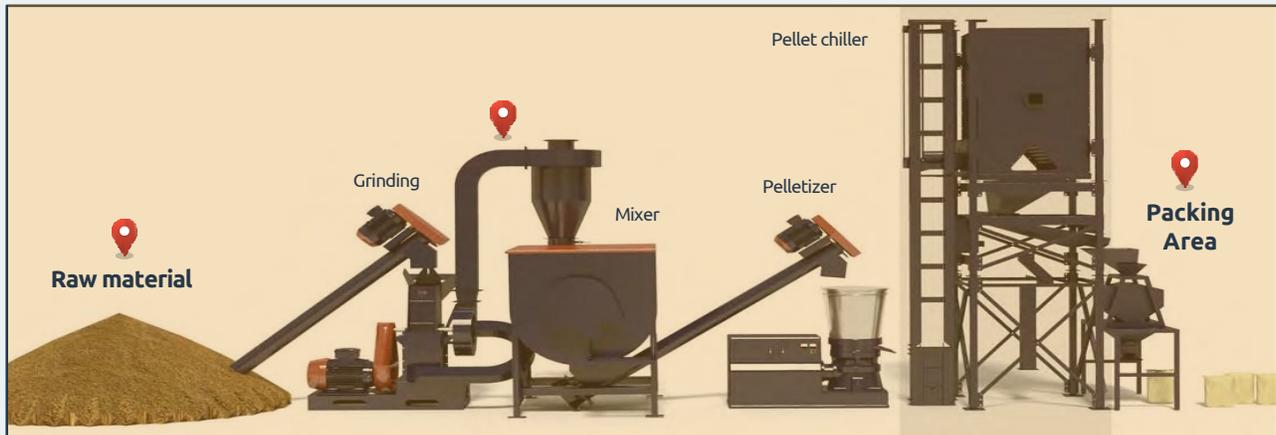
GRAIN SAMPLER MANUAL AND HYDRAULIC



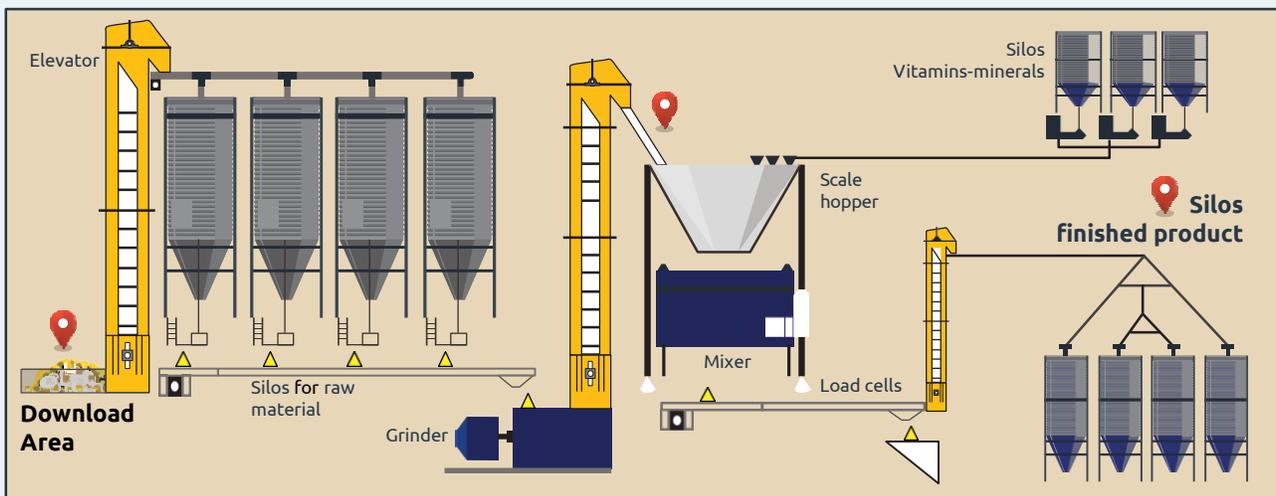
- ④ For every 30 MT that pass through the pipe, a minimum of 10 kg of sample must be taken and then proceed with the preparation of the sample as explained for the raw material reception area (21 samples of 200 g).
- ⑤ This pipe must be easily accessible for cleaning and must be placed on the finished product conveyor, after mixing, to facilitate sampling.
- ⑥ The sampling containers must be properly washed and disinfected to avoid contamination.

Sampling points in two types of feedmills 📍

Plant 1



Plant 2



▶ Taking samples from feeders

After distributing the feed in the farm, take samples with one of the following methods:

- ① Take a sample of 50 g of feed for every 100 square meters in poultry farms. In pig farms take 50 g from each feeder. Once collected, all the samples are mixed and split as explained before to obtain a final volume of 500 g that is sent to the lab.
- ② Divide the house into 10 equal sections and take a 100 g sample from a feeder in each section. Mix the 10 samples of 100 g to obtain 1.0 Kg and divide the sample in two, obtaining a final sample of 500 g, which is sent to the lab.

Does your anti-mycotoxin additive meet the basic TOP and FACTS?

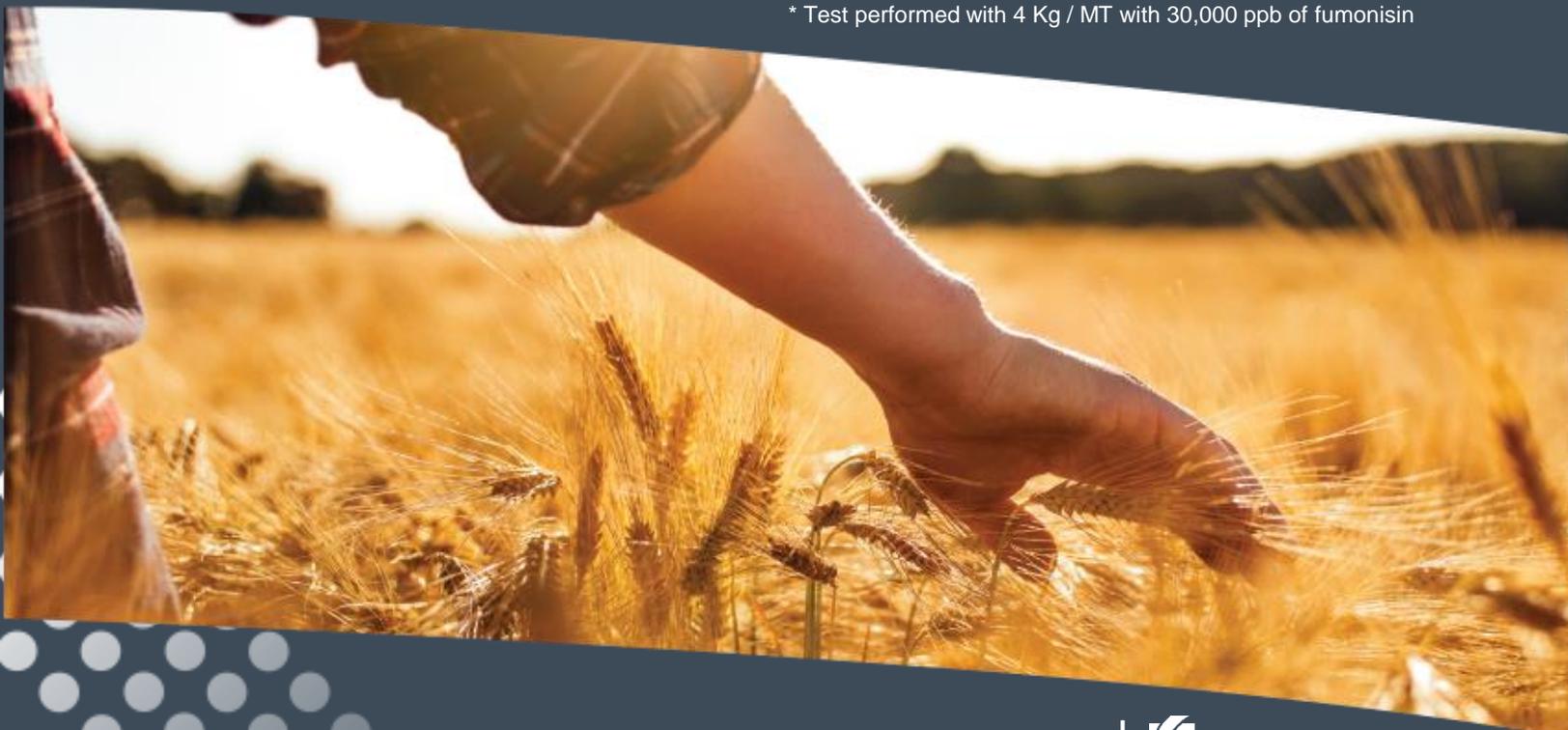
TARGET ORGAN PROTECTION

MYCOTOXIN	ORGAN	MYCOAD	MYCOAD AZ
Aflatoxin	Liver	YES	NO
Ochratoxin	Kidney	YES	NO
T-2 Toxin	Oral lesion	YES	YES
Fumonisin	Heart / Lung / Liver	YES	YES
Zearalenone	Reproductive tract	N/A	YES*
DON	Liver	N/A	YES

Facts	MYCOAD	MYCOAD AZ
<i>In vivo</i> dosage with TOP results	2.5 Kg / MT	1 Kg / MT*
Recommended commercial dosage	2.5 Kg / MT	1 Kg / MT
The clay is always obtained from the same mine	YES	YES
Approved in Texas , against Aflatoxin, USA	YES	N/A
Approved in the European Union against Aflatoxin. Regulation #1831 / 2003 (1m 588)	YES	N/A
ENDOTOXIN adsorption	N/A	YES
Efficacy approved by LAMIC in BRAZIL and other institutions against the following number of mycotoxins	4	4
Efficacy approved by LAMIC and other institutions in different types of animals	6	6
Nutrient absorption	NO	NO
<i>In vitro</i> efficacy test every:	100 MT	18 MT

* Test performed with 4 Kg / MT with 30,000 ppb of fumonisin

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www.agrimprove.com
agrimprove@agrifirm.com

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