



Biomonitoring mycotoxins: A new avenue to measure the impact of toxins on animals

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At Innovad®, we want to ensure that we have the means to do what we promise by continually upgrading our resources and skills to support our years of experience in the food/feed industry. As part of this commitment, we are collaborating with the University of Ghent to fund research on a unique new way of looking at diagnosing toxins impact on animals.



The biggest challenge in the mitigation of toxins contamination is the ability to properly detect the risk we are confronted with....

The presence of mycotoxins in feed impairs animal health and performance leading to economic losses.. Monitoring mycotoxin exposure is important to diminish associated losses.



Up till now, mycotoxins are mostly determined **in feed**. Feed analysis gives a good indication of the risk, especially the amount to which the animals are exposed. A <u>risk</u> assessment is hereby a useful tool, it combines characterization of possible health hazards with the amount to which the animals are exposed. However, this information is general and no information is given about the impact on the animals itself. Analyzing such feed samples could sometimes lead to misleading information due to the presence of hotspots and the difficulty of determining masked mycotoxins, both leading to underestimation of the risk.

Our latest research focus on the **exposure** assessment identifying **biomarkers** and developing the appropriate analytical method to determine toxic metabolites in animal biofluids. Such Biomonitoring could give more reliable results providing an indication of the real animal intoxication level and its economic impact. **A link** can also be made between concentrations in the feed and in the boological fluids.





Biomarkers for Biomonitoring

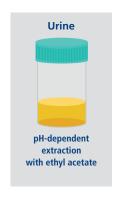
Biomonitoring is determining the exposure of animals to mycotoxins with the use of so-called biomarkers. Biomarkers are the molecules that are related to the exposure and that can be found in the biological matrices of the animals. Two types of biomarkers can be distinguished: direct (exposure) and indirect (mechanism/effect) biomarkers. The latter are non-specific and associated with either the effect or mechanism of the toxins. (I.e: change in So/Sa-ratio after fumonisin exposure or the change in liver/serum enzymes after the administration of aflatoxin B1) The effect-based biomarkers are even less specific than the mechanism-based. A typical example is the alteration in feed intake after administration of deoxynivalenol. The direct biomarkers are specific and directly linked to the exposure. This type of biomarker is often the mycotoxin itself or their phase I and II metabolites. For biomonitoring, the direct biomarkers are the most interesting.

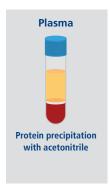
Matrices of Biomonitoring

Detection of biomarkers occurs in easily accessible matrices such as **blood**, **urine** and **feces**.

 Direct biomarkers or biomarkers for exposure, for example the rapidly absorbed mycotoxins can be detected in blood. Peak concentrations can be seen after few hours. The concentration in blood is directly related with the exposure. **Plasma** is thus a good matrix to measure mycotoxins a few hours after exposure.

- In urine the rapidly absorbed mycotoxins and their metabolites are also found. The relation with exposure is more difficult than in blood because the concentration depends on the amount of urine produced. Therefore, it is necessary to include creatinine as a non-food related marker to correct for the variation in production.
- Feces is a useful matrix for the mycotoxins whose absorption in the gastro-intestinal tract is low. They are directly excreted in feces. The concentration found can slightly deviate from the exposure due to detoxification by the intestinal microflora.







MEASURING COMPLETE EXPOSURE

Modified mycotoxins can convert back to their original forms during degestion and so contribute to the overall effect of the toxins.

Biomonitoring includes these effects and gives thus a very accurate estimation of the risk.

Advantages of biomonitoring

INDIVIDUAL ANIMAL INFORMATION

Biomonitoring measures the exposure at an individual level while feed analysis shows a general risk. Exposure at an individual level takes variation in food consumption and ADME parameters into account and integrates contamination from all different sources leading to a more accurate estimation and possible use in toxicokinetic studies.

AVOID MISLEADING SAMPLING

Although feed is an accessible matrix, it is difficult to sample correctly due to the presence of hotspots.

Urine, feces and plasma/blood in contrast are easy to sample correctly and also allow sequential sampling.

ADDITIONAL ANALYSIS BEYOND FEED

Feed is a consumable. After consumption it is no longer available.

In this scenario, biomonitoring might be of help in diagnosing a mycotoxin related problem.

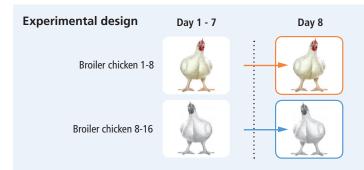




Biomonitoring analysis as a new tool to evaluate mycotoxin detoxifyers

Mycotoxin detoxifiers like **Escent® S** are products that diminish the effects associated with toxins. A possible way to prove the efficacy of these products is doing in vivo absorption tests. In these trials the concentration of mycotoxins in animal matrices is detected and the difference in concentration with and

without detoxifier is determined. This difference is a measure for the efficacy of the product. This type of test gives accurate results and should in the future replace in vitro tests or in vivo test based on non specific parameters such as growth performance and feed conversion.



16 broiler chickens were acclimatized during one week.

After one week 8 chickens received mycotoxins and the 8 other chickens received mycotoxins plus **Escent® S** (2.4 kg/ton feed). The selected mycotoxins were aflatoxin B1 (20 ppm in feed) and deoxynivalenol (5 ppm in feed).

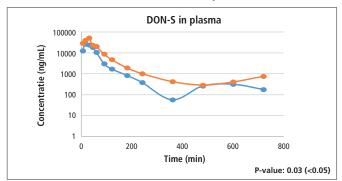
Escent® S and the mycotoxins were administered using an intra-crop bolus. Plasma was collected.

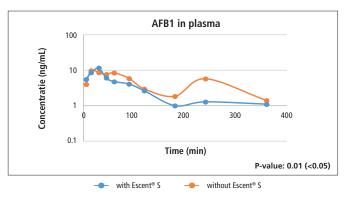
Results

After administration of DON, the best biomarker for exposure was DON-sulphate. DON-sulphate has a higher area than DON itself. After administration of AFB1, AFB1 remained the best biomarker for exposure.

Escent® **S** significantly decreased the concentration of mycotoxins (deoxynivalenol and aflatoxin B1) in plasma of broiler chickens. Therefore, this research is an additional way of proving the efficacy of **Escent**® **S**.

Results of the trial in plasma





Reference:

Related to biomarkers: general

T. Baldwin; R.Riley; N.Zitomer; K.Voss; R. Coulombe Jr.; J.Pestka; D. williams, A. Glenn, The current state of mycotoxin biomarker development in humans and animals and the potential for application to plant systems, *World Mycotoxin J. (2011)*.

A. Vidal, M. Mengelers, S. Yang, S. De Saeger, M. De Boevre, Mycotoxin Biomarkers of Exposure: A Comprehensive Review, Compr. Rev. Food Sci. Food Saf. 0 (2018). doi:10.1111/1541-4337.12367.

Related to the advantages of biomonitoring

N. Broekaert, M. Devreese, T. van Bergen, S. Schauvliege, M. De Boevre, S. De Saeger, L. Vanhaecke, F. Berthiller, H. Michlmayr, A. Malachová, G. Adam, A. Vermeulen, S. Croubels, In vivo contribution of deoxynivalenol-3-B-d-glucoside to deoxynivalenol exposure in broiler chickens and pigs: oral bioavailability, hydrolysis and toxicokinetics, *Arch. Toxicol. 91 (2017) 699–C712. doi:10.1007/s00204-016-1710-2*.

Related to new tool to evaluate mycotoxin detoxifiers

M. Devreese, A. Osselaere, J. Goossens, V. Vandenbroucke, S. De Baere, M. Eeckhout, P. De Backer, S. Croubels, New bolus models for in vivo efficacy testing of mycotoxin-detoxifying agents in relation to EFSA guidelines, assessed using deoxynivalenol in broiler chickens, Food Addit. Contam. - Part A Chem. Anal. Control. Expo. Risk Assess. 29 (2012) 1101–C1107. doi:10.1080/19440049.2012.671788.