

MANAGING MYCOTOXINS IN FEED AFTER HARVEST PART 1



By Dr Max Hawkins, Alltech Mycotoxin Management Team

The summer of 2012 presented the grain and livestock industry with a mycotoxin challenge on a scale not seen for many years. Severe heat and drought covered nearly 90% of north America. European producers faced the same challenge in the south, while the north and west experienced extreme wet conditions.

The global trade of grains further exacerbated the mycotoxin challenge, leading to unforeseen toxin interactions and growing concern about the impact of toxins in one region on the quality of feed in another. The conditions reduced yields, grain and forage quality, and served as a precursor for mould growth and mycotoxin production.

The predominant concern for mould growth in the US, the world's largest exporter of grains, was *Aspergillus flavus*, as it thrives in hot dry weather. This mould and the aflatoxin B1 it produces is a major concern, particularly to dairy farmers, due to its regulation and potential transfer to milk.

Early field scouting confirmed that *Aspergillus* was present in higher than normal numbers. However, these fields tended to be regionalised. In the areas where aflatoxin is concentrated, it is a serious concern and a considerable amount of milk has been condemned.

Fusarium verticillioides was found in nearly 100% of the fields over the entire cornbelt area. Fumonisin B1, B2 and B3, produced by this mould, can have a negative effect on all classes of livestock. It affects ruminant immune status and productivity.

Alltech conducted a harvest analysis throughout the corn growing area of the US. These

samples were analysed using LC-MS/MS technology. Alltech's 37+ Program identifies 37 mycotoxins, including 'masked' toxins. The percent detection for different mycotoxins are shown in Table 1. Fumonisin was found in nearly 100% of the samples, followed by *Penicillium* and the DON Group. It is important to note that over 95% of samples contained more than two mycotoxins.

This presents a compound effect as they can act additively or synergistically and so often have a greater impact on livestock production than may be expected.

There are many instances where mould growth has flourished and continued in storage. This presents an even greater danger for livestock as it is brought out of storage this winter and spring.

Management

Often producers try to find other sources to replace or limit the use of contaminated feedstuffs. However, this year's broad spectrum challenge makes it difficult, if not impossible.

It is also important to realise that most corn and corn silage

Inorganics are made of silica and referred to as clays.

Organics are carbon based and are equivalent to plant fibres. Diaz et al. (*Mycopathologia* 157:223, 2004) has shown polymeric glucomannan adsorbent extracted from yeast cell wall, known as GMA, to be effective in preventing the transfer of aflatoxin into the milk.

GMA has also been shown to be effective in limiting the negative effects of combinations of *Fusarium* mycotoxins.

Broad spectrum mycotoxin

Origin	Aflatoxins ¹	Ochratoxins ²	DON group ³	T-2 type ⁴	Fumonisins ⁵	Penicillium mycotoxins ⁶	Ergot toxins ⁷	Sterigmatocystin	Zearalenone group ⁸	Alternariol
IA	0	20	20	20	100	20	0	0	0	0
NC	67	33	33	0	100	33	0	0	33	0
OH	0	50	0	0	100	50	0	0	0	0
PA	10	10	75	45	90	55	10	10	25	0
SD	0	25	13	13	100	63	0	13	0	13
WI	33	33	33	0	100	67	0	0	33	0

¹Aflatoxins = Aflatoxin B1 + B2 + G1 + G2; ²Ochratoxins = Ochratoxin A+B; ³DON group; Deoxynivalenol (DON) + 15-acetyl DON + 3-acetyl DON + Fusarenon X + Nivalenol + DON 3-Glucoside; ⁴T-2 group = T-2 + HT-2 + Diacetoxyscirpenol (DAS) + Neosolaniol; ⁵Fumonisins = Fumonisin B1 + B2 + B3; ⁶Ergot toxins = 2-bromo-alpha-ergocryptine + Ergocominine + Ergometrine/ergonovine + Ergotamine + Lysergol + Methylegonovine; ⁷Penicillium mycotoxins = Gliotoxin + Patulin + Penicillic acid + Roquefortine C + Mycophenolic acid + Verruculogen + Wortmannin; ⁸Zearalenone group = Zearalenone + α Zearalanol + β Zearalanol+Zearalanone

Table 1. Percent detection of various mycotoxin groups in harvest US corn samples 2012.

The average levels of mycotoxins were also assessed. Many of these levels are at practical limit and require a control programme. However, as they are combined with other ingredients, these levels could pose a more serious threat to livestock production.

Storage

The effects of storage have already been seen in the 2012 crop. Much of the crop was at a more mature stage when harvested and the forage did not compact well. This makes it more susceptible to mould growth and mycotoxin production.

The grain, even though dried sufficiently to <15% moisture, has been subjected to temperature and humidity swings.

samples from across the US were contaminated with multiple mycotoxins, and not just aflatoxin. In fact *Fusarium* is of greatest concern and requires a different approach to maintain livestock health and efficiency of production.

Adsorbents are widely used to manage mycotoxins. These are feed additives that are non-nutritive and are comprised of long polymeric molecules.

They are non-digestible and maintain their characteristics as they pass through the digestive system and are excreted. While in the digestive tract they attract and bind small molecules such as mycotoxins. This prevents the toxins from passing into the blood system and into the organs of the livestock.

These adsorbents are classified as inorganic or organic.

adsorbents are an effective tool to lessen the effect of contaminated feedstuffs on livestock production. They represent the most effective way to utilise the crop you are feeding this year.

Producers need to be sure of the efficacy, traceability and consistency of the mycotoxin binder they choose.

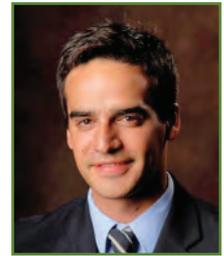
- Has it been tested in-vivo?
- Does it bind essential nutrients (often the case with clay based products)?
- Has its efficacy been demonstrated in independent research?

Protect your herd and your profits!

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MANAGING MYCOTOXINS IN FEED AFTER HARVEST PART 2



by Pedro Caramona, Alltech,
Mycotoxin Management team

Every harvest season delivers a variety of challenges that farmers and producers need to address in order to safeguard the health of their animals and the quality of their feed. This season has already made its mark. Aside from the poor nutritive quality of harvested crops, issues with mould and mycotoxin contamination look set to continue.

2012's growing and harvesting conditions provided optimal conditions for mycotoxin growth. Perhaps the most widely talked about has been the surge in levels of aflatoxin contamination – threatening the livelihoods of producers, the profitability and the security of the industry as a whole. Milk discards and the associated financial losses to producers, together with concern over the safety of the feed and food chains, means that minimising mycotoxin contamination continues to be a top priority for Europe.

Aflatoxins, produced by *Aspergillus* moulds, grow on crops in the field or commodities during storage under excessively dry conditions. Aflatoxins are the most heavily regulated mycotoxins due to their highly toxic and carcinogenic nature. When

Fig. 1. Number of toxins found in samples from Europe's 2012 harvest.

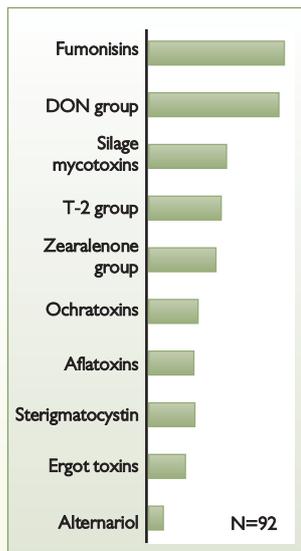
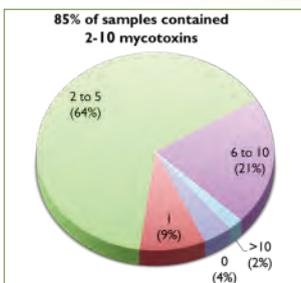


Fig. 2. Percentage detection of various mycotoxin groups in samples from Europe's 2012 harvest.

ingested by dairy cows, between 1% and 6% of aflatoxin B1 in feed is converted to aflatoxin M1 (AFM1) and subsequently appears in the milk. This seemingly small rate of transfer is enough to have significant effects on the health and performance of animals and potentially compromise the safety of the milk supply. Symptoms of aflatoxin contamination in dairy cows include reduced milk yield and quality, decreased immune status leading to increased susceptibility to infection, reduced protein synthesis, liver damage and, most crucially, carcinogenicity (cancer causing).

Due to its highly carcinogenic properties, the EU enforces a regulatory limit of 0.05ppb aflatoxin in milk.

While aflatoxin contamination is undoubtedly of major concern to both industry and consumers, it is important that producers remember that aflatoxin is only one of approximately 500 toxins that animals can be exposed to through the diet. Forages and grains are naturally contami-

nated with multiple mycotoxins, weakening the immune and detoxification systems of dairy cows and reducing the animals' capacity to deal with the more potent aflatoxins. Additionally, mycotoxins derived from *Penicillium* moulds (for example Roquefortine C, Patulin, Penicillic acid) have an antibiotic effect on beneficial rumen microbes, further reducing the detoxification capacity of the animal. Alltech's 37+ findings show the extent and the broad spectrum nature of the mycotoxins challenge in this year's harvest (Fig. 1 and 2).

Risk assessment

An effective mycotoxin management program should play an integral part in herd health management – promoting animal health and protecting producer profits. Methods for the simultaneous analysis of several important mycotoxins, such as liquid chromatography with tandem mass spectrometry (LC MS/MS) enables the rapid quantification of a greater number of mycotoxins at very low levels and thus covers a broader spectrum of potential contamination with different mycotoxins.

Alltech's investment in this technology through its 37+ Programme has allowed not only significant improvement in precision but also the analysis of complex matrices, such as silages and TMR.

Management tools

Recognising the impact of mycotoxins in dairy farms is often difficult since symptoms are non-specific and the synergy between mycotoxins adds another layer of complexity. Mycotoxins can cause acute health or production issues, however, in the majority of cases mycotoxins are a contributing factor to vague, chronic problems, including poor perfor-

mance and higher incidence of disease. In addition to mycotoxin analysis, practical feed and animal management can reduce the risk posed to the animal and future instances of mycotoxin contamination. Alltech's MIKO (mycotoxin hazard analysis) Programme is a systematic audit or management tool for farms and feedmills. Based on HACCP (hazard analysis critical control points) principles, this programme involves the identification of critical control points and the establishment of corrective actions and appropriate monitoring procedures to mitigate the impact of mycotoxins.

Conclusions

- Mycotoxin contamination is inevitable, however, there are steps one can take in order to reduce the risk of impacting animal health and performance as well as producer profit.
- In order to achieve a better assessment of mycotoxin contamination, analyses should be conducted for a multitude of mycotoxins and their additive and synergistic effects must be considered.
- There is a need for implementing integrated strategies to counteract mycotoxin contamination in dairy herds and throughout the production chain, including mould growth and mycotoxin management.
- Continuous investment in analytical technologies that support a more complete understanding of a broad-spectrum mycotoxin challenge will promote technologies designed to mitigate the negative impact of mycotoxins on dairy farms.

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EMERGING MYCOTOXINS IN DAIRY PART 3



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There are many synonyms for mycotoxins; 'hidden killers', 'silent killers', 'thieves' etc. The complexity of dairy feeds and the relatively higher difficulties in precisely analysing mycotoxins in forages strongly support these synonyms.

Knowledge and understanding of their nature and occurrence continues to advance the industry's growing appreciation for the adverse affects of mycotoxins on animal health and performance. We know now that upwards of 500 mycotoxin types exist so if we are to adequately address the challenge posed by these toxins, efforts should be made to analyse for as many as possible.

Emerging mycotoxins

There are a few laboratories in the world capable of analysing 50 or more mycotoxins. Such analyses, however, are restricted to research and regulatory purposes. To bring such capability to the field and to assist animal producers around the world, Alltech established its 37+ Program.

This Program makes use of state-of-the-art UPLC-MS/MS techniques to analyse more than 37 mycotoxins. Table 1 compares the list of mycotoxins that are included in Alltech's 37+ Program with those analysed routinely in the animal industry. For ease of understanding, additional mycotoxins tested for in the 37+ Program are considered emerging mycotoxins.

Fusarium mycotoxins

Many studies have shown that when grains/feeds contain DON, other mycotoxins such as 3-acetyl DON and 15-acetyl DON are also present. Recent studies have shown how masked or conjugated DON can increase overall DON toxicity. Masked/bound DON cannot be detected using conventional methods of mycotoxin analysis, including HPLC.

The dairy industry understands all too well the negative effects of T-2 toxin. But few people know that diacetoxyscirpenol (DAS) is more toxic than T-2 toxin.

Trichothecene Fusarium mycotoxins and fumonisins can sup-

press immunity even when present at lower concentrations. Since performance may not be affected at such low levels often their impact on immunity goes unnoticed.

Zearalenone is known to compromise reproductive performance of dairy cows. Table 1 shows three other forms of zearalenone which are not routinely analysed for in the dairy industry, explaining why we often see reproductive issues in dairy cows even when the actual zearalenone level is very low in the TMR.

Penicillium mycotoxins

Some Penicillium moulds can survive at low pH levels which often occur in silages. Such moulds produce mycotoxins such as patulin, mycophenolic acid and roquefortine C. These mycotoxins have been shown to compromise rumen health and immunity. A recent study from The Netherlands has shown increased incidence of metabolic disorders, compromised anti-oxidant status, higher somatic cell counts and poor milk production

when cows are fed TMR containing mouldy silages. This necessitates testing silages and TMR for these emerging mycotoxins – not only aflatoxins and Fusarium mycotoxins.

Other emerging mycotoxins

Apart from Fusarium and Penicillium mycotoxins, many Aspergillus, Alternarium and ergot mycotoxins, which are not routinely analysed, are also included in Alltech's 37+ Program. The toxicity of ergot toxins in dairy animals is well understood and the ability to analyse many allows for a more precise understanding of the potential toxicity. Gliotoxin is often detected in silages and has been linked to haemorrhagic bowel syndrome (HBS) in cows.

Mycotoxin management

In light of the unexpected toxicity from emerging and unknown mycotoxins, the use of a HACCP-like approach such as Alltech's MIKO Program to control mycotoxins in feed mills and farms is highly recommended.

Conclusion

The role of lesser known and emerging mycotoxins, along with routinely analysed mycotoxins, should be considered in determining the total toxicity to dairy cows. The non-specific symptoms and subtle nature of the mycotoxin challenge warrants the implementation of mycotoxin control strategies all along the mycotoxin production chain rather than waiting for the devastation to happen. ■

Table 1. Comparison of routine mycotoxin analysis vs. Alltech's 37+ Program.

Mycotoxin group	Routine analysis	37+ program
Aflatoxins	Aflatoxin B ₁	Aflatoxin B ₁ , B ₂ , G ₁ , and G ₂
Ochratoxins	Ochratoxin A	Ochratoxin A and B
Type A trichothecenes/T-2 Group	T-2 toxin	T-2 toxin, DAS, HT-2 toxin, neosolaniol
Type B trichothecenes/DON Group	DON	DON, 3-acetyl DON, 15-acetyl DON, Nivalenol, Fusarenon-X, masked DON
Fumonisin	Fumonisin B ₁	Fumonisin B ₁ , B ₂ , and B ₃
Zearalenone group	Zearalenone	Zearalenone, α-zearalenol, β-zearalenol and zearalanone
Other penicillium and aspergillus mycotoxins	-	Patulin, roquefortine C, penicillic acid, mycophenolic acid, gliotoxin, sterigmatocystin, verruculogen, wortmannin
Ergot mycotoxins	-	2-bromo-alpha-ergocryptine, ergocornine, ergometrine, ergotamine, lysergol, methylergonovine
Alternaria toxins	-	Alternariol
Total number analysed	6	38

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