Short Report #034
YEAR 2016

Effect of Toxy-Nil® on the reduction of aflatoxin M_1 levels in milk of lactating dairy cows fed aflatoxin B_1

Aim

The objective of this research was to establish the efficacy of dietary mycotoxin binder Toxy-Nil® in reducing transfer of aflatoxins from diet to milk of lactating dairy cows challenged with an aflatoxin B_1-contaminated diet.

Introduction

Aflatoxins are mycotoxins produced by fungi of the genus Aspergillus, present in 20 to 30% of feeds globally, and estimated to exceed $250 million in annual losses to crop and livestock operations in the USA alone. Aflatoxin B1 is the major toxin produced by Aspergillus and it is considered the most carcinogenic naturally-occurring toxin. It is quickly absorbed in the upper gastrointestinal tract of animals, being detected in plasma of dairy animals by 5 minutes after ingestion. After absorption, aflatoxin B1 is metabolized in the liver to several constituents including aflatoxin M1, a hepatotoxin, carcinogen and immune suppressor excreted through milk and urine. Transfer rates from feed to milk have been reported to range from 0.25% to 6.2%. In the US, the Food and Drug Administration (FDA) has set the limit for aflatoxin M1 in milk at 0.5 µg/kg. In Europe, the limit is set at 0.05 µg/kg.

Toxy-Nil® is a reliable and cost-effective solution to neutralize mycotoxins and minimize their negative effects on different types of cattle. Toxy-Nil® is also the solution of choice against aflatoxin M_1 in milk.

Toxy-Nil® benefits:

- ✓ The reliable solution against aflatoxin M_1 in milk
- ✓ Better growth performance and nutrient utilisation in calves and beef cattle
- ✓ Effective protection against several mycotoxins

Experimental design

Twenty-four mid-lactation Holstein cows housed in a free-stall facility were blocked based on days in milk, milk yield, and parity. The experiment was conducted as a complete randomized block design. Cows within a block were randomly assigned to receive one of three treatments:

- Control: no aflatoxin B_1 and no mycotoxin deactivator
- Aflatoxin: 2.8 mg of aflatoxin B_1/cow/day
- Aflatoxin+Toxy-Nil®: 2.8 mg of aflatoxin B_1 + 100 g of Toxy-Nil®/cow/day

The experiment comprised three 7-day periods, which included an acclimatization period from -7 to -1 days, an experimental period, during which aflatoxin B_1 and mycotoxin deactivator were fed to cows as per treatment design (days 1 to 7), and a recovery period, during which all cows received the diet without mycotoxin or product and were monitored until levels of aflatoxin M_1 in milk were undetectable (days 8 to 14).

The measurements and calculations were: reduction in aflatoxin M_1 concentrations in milk and urine, reduction in aflatoxin M_1 excretion through milk and urine and reduction in aflatoxin transfer from feed to milk.

Results

Treatments had no effect on performance parameters such as feed intake, milk yield, milk composition, or somatic cell count (SCC) (data not shown) due to the short-term exposure. Level of aflatoxin B1 biomarker of exposure (aflatoxin M_1) in the milk and urine dropped when Toxy-Nil® was fed to cows (table 1). That means the product was effective to reduce bioavailability of aflatoxin B1 in the gastrointestinal tract significantly. Transfer of aflatoxin B1 from feed to milk averaged 1.0 and 2.7% in cows fed contaminated diet with and without Toxy-Nil® respectively. In other words, Toxy-Nil® reduced aflatoxin M_1 in milk by 63.4% ($p<0.001$) and in urine by 51.1% ($p<0.001$). The clearance rate of aflatoxin M1 in milk did not differ ($p\geq0.40$) between contaminated control and Toxy-Nil® fed group (27.1 and 27.4%/day respectively).

Table 1. Effects of aflatoxin B_1 and dietary inclusion of Toxy-Nil® on dairy cows

Variable	Control	Aflatoxin	Aflatoxin + Toxy-Nil®	SEM1
Intake of aflatoxin, µg/kg of diet	0.0^a	106.5^b	107.6^b	2.9
Intake of adsorbent, % of diet	0.00^a	0.00^a	0.38^b	0.01
Aflatoxin M_1 in milk, µg/kg	0.00^a	0.57^b	0.23^c	0.04
Aflatoxin M_1 excretion in milk, µg/day	0.0^a	20.5^b	8.1^c	1.7
Aflatoxin transfer to milk, %	0.00^a	2.74^b	1.00^c	0.23
Clearance of aflatoxin M_1 from milk, %/day[2]	0.00^a	27.10^b	27.35^b	1.33
Binding efficiency of product[3]	0.00^a	0.00^a	63.44^b	8.23
Aflatoxin M_1 concentration in urine, µg/l	0.54^a	14.17^b	6.93^c	1.46
Aflatoxin M_1 excretion via urine, µg/day	15.4^a	521.6^b	225.8^c	53.1
Aflatoxin transfer to urine, %	0.00^a	18.63^b	8.06^c	1.89

$^{a-b}$ Means within row with different superscript letters differ ($p\leq0.05$)
[1] Pooled SEM
*[2] Clearance was calculated as: (aflatoxin M_1 in milk for each day)*100/(average of aflatoxin M_1 in milk on day 6 and day 7)*
*[3] Binding efficiency calculated as: 100-(aflatoxin M_1 in milk on day 6 and day 7)*100/(average of aflatoxin M_1 in milk on day 6 and day 7 for contaminated control)*

Daily feeding of 2.8 mg of aflatoxin B_1 to lactating dairy cows resulted in a few differentially expressed genes in blood leukocytes, but elicited numerous changes in gene expression in milk somatic cells (data not shown). In total, about 130 common genes expressed differently in milk somatic cells of cows fed aflatoxin B_{11} and they were represented by functional clusters for glycoproteins and milk proteins, milk secretion, signalling, protein binding, transporter activity, response to steroid hormones and by protein domains for α- and β-caseins. Approximately 20% of those differently expressed genes were "mammary gland-specific". Many of the differentially expressed genes in milk somatic cells as well as in blood leukocytes were modulated in cows fed Toxy-Nil®.

Conclusion

Inclusion of 100 g of Toxy-Nil® per cow per day in the diet significantly reduced aflatoxin M_1 concentration in milk of cows consuming total mixed ration containing approximately 5 times the FDA limit of 20 µg and 20-times the EU limit of 5 µg of aflatoxin B_1/kg of diet dry matter. Supplementation with Toxy-Nil® also reduced the time required post-challenge for aflatoxin M1 concentrations to drop below the legally accepted (by FDA) limit for milk. Results suggest that Toxy-Nil® significantly reduced bioavailability of aflatoxin B_1 in the gut.
In addition, the level of mycotoxin deactivator used (0.37%) indicated that it is a very effective product, since the milk aflatoxin M_1 reduction was much higher than in other studies conducted to evaluate the efficacy of many different commercial products including pure clays.