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Prevention of mycotoxin effects in dairy cows by adding an anti-mycotoxin product in feed

Z. Zouagui[1], M. Asrar[1], H. Lakhdissi[1], E.H. Abdennebi[2*]

1. Department of Medicine, Surgery and Reproduction, IAV Hassan II, Rabat, Morocco
2. Department of Veterinary Biological and Pharmaceutical Sciences, IAV Hassan II, Rabat, Morocco

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EH Abdennebi
Pr.abdennebi@yahoo.fr
+212673383382

Abstract

The main objective of this work was to assess the impact of using an anti-mycotoxin product on liver function, milk production and reproductive performances of dairy cows subjected to a high risk of mycotoxins. The evaluation criteria consisted of liver function tests including total blood proteins, urea, total bilirubin and transaminases activities. They also include an assessment of different fertility parameters, milk production and milk somatic cell counts of individual treated and non-treated animals.
As compared to the control cows, results of these investigations showed that the anti-mycotoxin product significantly improved liver function and reproductive parameters of treated cows. Additionally, the production and the quality of milk, as tested by its somatic cell numbers, were improved. The observed positive effects may, most probably, be explained by the important binding properties to mycotoxins of the clay and the inactivated bacteria and yeasts used as ingredients of the anti-mycotoxin mixture.

1. Introduction

Mycotoxins are secondary metabolites produced by fungi from harvesting to storage and distribution of food. These substances are produced mainly by five species of fungi (*Fusarium* sp, *Penicillium* sp. *Aspergillus* sp. *Claviceps* sp. *Alternaria* sp.). Growth of these microorganisms and production of their mycotoxins are mainly influenced by the presence of fungal spores, the nature and the density of cultures, the climate and mechanical damages of plants [1,2]. In livestock production, and particularly in ruminants which are considered as a filter for mycotoxins, several studies have shown significant negative effects on health and productivity of these animals. In dairy cows, mycotoxins are frequently incriminated in poor reproduction performances, low production and poor quality of milk [3-8].
Since it is difficult to effectively control mycotoxins in the environment and in animal food, the use of anti-mycotoxin substances is one way to prevent, or at least to attenuate, their deleterious effects. In poultry and pork, the use of such substances has now become a common practice [9,10]. However, in ruminants, and especially in dairy cows, few studies are now available on this issue. Some preliminarily investigations have shown that the addition of certain products in the diet improves productivity of cows exposed to the risk of mycotoxins. Thus, the objective of this study was to investigate the benefit, if any, of the use of an anti-mycotoxin product on milk production and reproduction performances in a dairy farm with a high risk of mycotoxins.

2. Materials and methods

2.1. *Experimental animals*

This study was carried out in a dairy farm in the region of Gharb (Morocco), where a total of 600 dairy cows were conducted in an intensive and modern management. A mixed ration (MR), with corn silage as the main forage, was fed to the animals throughout the year. Livestock was free from any contagious disease and a medical prevention, associated with bio-security measures, was followed to prevent any disease of cattle. An integrated program on reproduction was also implemented at this farm

2.2. Mycotoxin assessment

To assess the mycotoxin risk in the studied farm, samples of the MR were taken as described by Mahieu [10] and analyzed for the major mycotoxins in the Development Laboratory and Analysis of Ploufragan (France), using the high pressure liquid chromatography associated to a mass spectrophotometer (HPLC/MS/MS).
Flowing this analysis, which revealed the presence of high levels of some mycotoxins (see results section),

2.3. *Experimental design*

36 Holstein cows in early lactation were randomly divided into two groups of 18 animals each. The first group received the anti-mycotoxin product, while the second was kept as a control without any treatment. The anti-mycotoxin product is a mixture of the clay with yeast enzymes and mineral adsorbents. This product is available under the trade name of Mycofix®. The characteristics of the experimental animals at the beginning of this study are described in table 1.

Table 1: Characteristics of the experimental animals

Parameters	Control lot	Treated lot
Average age of cows (year)	4± 08	4±0.9
Lactation stage	1	1
Cumulative milk production/year (liter)	8137±1300	7590±1268
Number of days of lactation	320±32	329±52
Average milk yield / day (liter)	25.5 ±2.8	23.1±3.7
Calving interval (days)	401±39.7	429±42.8

Each day, animals of the treated group were individually given an oral dose of 30 g of the anti-mycotoxin during a period of 100 days. Health status as well as reproduction and milk production parameters were regularly monitored in both groups of animals, prior to and during 100 days after calving. Blood samples were taken at day 0 (corresponding to the day of calving) and at the 30^{th}, 60^{th} and 100^{th} day post-calving.

2.4. *Laboratory investigations/Blood chemistry and milk parameters*

Blood samples were later used for the determination of selected biochemical parameters such transaminase activities (ALT and AST), blood urea, total bilirubin and total blood proteins. Milk samples were also taken, at the same times as for blood, except for the 100 day post calving. Immediately after sampling, the quality of milk was monitored by the determination of its somatic cell count using an electronic counter. Results of these investigations were analyzed by the statistical test of independence ($\chi 2$).

3. Results and Discussion

3.1. *Food analysis*

The analysis of food samples from the MR revealed relatively high levels of mycotoxins, especially those of fuminosines (Table 2).

Table 2: Major mycotoxins detected in feed of the studied animals (MR) with their corresponding levels

Mycotoxin	Deoxinivalenol (DON)	Diacetoxyscirpenol (DAS)	Fuminosins (FB)	Zearalenone (ZEA)
Concentration (µg/kg)	1021	1791	2339	595

Although ruminants have normally important capabilities of detoxification, due to their rumen flora, data from the literature indicate that the detected levels of contamination present a real risk to these animals [10]. Furthermore, as conducted in the intensive fashion, these cows became more sensitive to diseases, particularly acute acidosis and subclinical metabolic disturbance, which would amplify the effects of mycotoxins. Even cattle are able to metabolize mycotoxins, the resulting metabolites may also be as toxic as the original substances or have additive and even synergistic effects [6, 7].

3.2. *Biological profile*

As compared with the control animals, results of the biochemical analysis clearly indicate that the anti-mycotoxin product had a positive effect on studied parameters of treated animals. As indicated by the transaminase activities, liver function of treated animals was significantly improved, especially after the first month of treatment ($p < 0.05$) (table 3). Accordingly, a significant increase of total proteins and a decrease of blood urea and total bilirubin were recorded starting from day 30 ($p < 0.05$) (table 3). These effects may, most probably, be explained by the binding properties to mycotoxins of the clay and of the inactivated bacteria and yeast present in the used anti-mycotoxin product. It has been reported that certain strains of bacteria and yeasts have parietal structures capable of binding to mycotoxins [11,12,13]. The clay has an important adsorbing power to mycotoxins, without interfering with the bioavailability of vitamins and other nutrients [14]. In parallel, since the liver function was improved in treated animals, the hepatic detoxifying power and capacity of metabolism of mycotoxins might also be increased and, therefore, there would be an important elimination of mycotoxin metabolites, such as aflatoxin P, Q1 or B2 [7, 11].

Table 3: Results of the biochemical parameters (mean ± standard deviation)

Parameter	Day 0		Day 30		Day 60		Day 100	
	Controls	Treated cows	Controls	Treated cows	Controls	Treated cows	Controls	Treated cows
AST (IU/l)*	22±34	18±23	83±40	55±35	100±45	101±43	81±28	86±31
ALT (IU/l)**	14±9.0	12±10	41±28	54±7.1	47±14	48±20	46±25	52±27
Total proteins (g/l)	80±5.0	83±8.0	79±25	90±7.0	91±4.0	92±3.0	92±9.0	92±7.0
Total Bilirubin (mmol/l)	38±5.0	38.3±7.0	56±6.0	41±5.0	33±6.0	35±7.0	30.4±2.0	28.7±3.0
Urea (mmol/l)	-	-	7.7±4.0	6.2±3.0	7.2±2.0	7.8±5.0	-	-

*AST: Aspartate transferase
**ALT: Alanine transferase

3.3. *Milk production and quality*

At the peak of lactation, the treated cows produced an average of 32.9 liters per day of milk against 23 liters for the control cows (table 4). The estimated standard quantities expected for 305 days are, thus, 6580 and 4600 liters for the treated and the control cows, respectively. Mycotoxins are known to affect dairy cow performances, including milk production [5,6,10,13]. According to Whitlow and Hagler [6], T-2 toxin, at concentrations of 300 to 500 ppb, decrease food ingestion and milk production of dairy herds. Dacetoxyscirpenol (DAS) has also similar undesirable effects as those of the T-2 toxin [15, 16]. Consequently, the very high concentrations of DAS and fuminosins detected in the MR given to the studied animals are very likely to be responsible for the observed differences between the two groups of cows and confirm the positive effect of the used antimycotoxin product.

Table 4: Milk production recording from control and treated cows
(Mean ±standard deviation in liters).

	Day 0 (calving day)	Day 30 (post calving)	Day 60 (post calving)
Controls	25.5±7.0	23±8.0	26.5±9.0
Treated cows	27.4±8.0	32.9±11	29.7±9.0

Results of somatic cell counts showed a significant difference between the two groups of animals ($p < 0.05$) throughout the test period. The average numbers were 300,000 cells /ml in treated animals and 519 000 cells/ml in the controls (table 5). The immunosuppressive effect of mycotoxins, due to their related hepatotoxicity, seems to be the most plausible explanation for these differences [10,13].

Table 5: Somatic cell counts (thousands / ml) (mean ± standard deviation)

	Day 30	Day 60	Day 100
Controls	234±23	156±21	519±11
Treated cows	343±38	244±24	300±17

3.4. *Reproduction parameters*

The summary of reproductive parameters is depicted in table 6. The comparison of the time-intervals between calving and first artificial insemination (AI) and between calving and fertilizing AI revealed that these times were shorter in the treated cows. Treated cows had also the highest overall pregnancy rate (50% against 27.2% for the control group) with a slight improvement of the pregnancy rate after the first insemination (up to 35 %).

Table 6: Results of the reproductive parameters (main standard ± deviation)

Parameters	Control cows	Treated cows
Calving-1st AI (days)*	70±17	61±12
Calving.-FAI (days)**	130±12	94±42
Pregnancy rate after 1st AI.	19%	35%
Overall pregnancy rate	27.2%	50%
% of cows with 3 or more AI	33%	22%

* Calving-1st AI= Time-interval between calving and 1st artificial insemination

**Calving-FAI= Time-interval between calving and fertilizing artificial insemination

In both groups, the percentage of cows that had 3 or more effective artificial inseminations exceeded the maximum threshold of 15% adopted in such breeding, However, in treated animals this parameter was less than that obtained from the non-treated animals (22% against 33%).

The relatively poor reproductive performances observed in non-treated cows could be related to zearalenone present in the mixed feeding ration (MR). Zearalenone is a major toxin produced by the *Fusarium* molds. Since its chemical structure is similar to that of the estrogen hormones, this chemical is well known by its estrogenic activities [5]. In fact, in cattle, zearalenone causes reproductive disorders and various modifications at the genital organs, mainly when its concentration in feed is near to 400 ppb [6,17,18]. Consequently, the detected levels of zearalenone in the feed of the studied cows (595 mg/kg) are sufficient to produce undesirable effects in non-treated animals and the improvement of the reproductive performances of the treated animals indicates that the anti-mycotoxin product is also effective against zearalenone.

Conclusions

This study, which aimed to explore the effect of an anti-mycotoxin product Mycofix® on the performances of dairy cows subjected to a significant mycotoxin risk, revealed that this product, given at a daily oral dose of 30 g allowed the improvement of liver function, reproductive performances and milk production and quality.

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