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Evaluation of adsorbents on the health and productivity of finishing steers fed a naturally mycotoxin-contaminated diet

Avaliação de adsorventes na saúde e produtividade de novilhos terminados em confinamento submetidos à dieta naturalmente contaminada por micotoxinas

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Highlights

Both Adsorbents showed similar hepatoprotective effects in mycotoxin-challenged cattle.
The Mixed Adsorbent reduced the immunosuppression caused by mycotoxins.
The mixed adsorbent modulated ruminal papillae and improved feed conversion.

Abstract

Mycotoxins are common contaminants in cattle diets that compromise animal performance and health. Despite the demonstrated efficacy of adsorbents in poultry and swine, studies involving feedlot cattle, particularly those fed naturally contaminated diets, are limited. This study evaluated the impact of a mixed adsorbent (MA) and an inorganic adsorbent (IA) on the hepatic, ruminal, respiratory, and immune health and productive performance of 36 crossbred steers (½ Angus-Nelore; average weight of 501.7 ± 33.2 kg and 11 ± 5 months of age) fed naturally contaminated diets containing aflatoxin, zearalenone, deoxynivalenol, and fumonisins over 63 days. The treatments included a control, MA (bentonite,

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yeast cell wall, and silymarin), and IA (bentonite, clinoptilolite, and activated charcoal from babassu). Parameters related to immune, hepatic, respiratory, and ruminal functions as well as animal performance were assessed on days 0, 21, 42, and 63. Immunologically, MA treatment resulted in higher neutrophil and lymphocyte counts, whereas the IA group exhibited only increased neutrophil counts on day 63 compared to the control group. Both treatments resulted in a higher albumin/globulin ratio, indicating a more efficient immune response. The hepatic profiles revealed a tendency for the control group to have higher AST levels (p=0.07) and greater hepatic degeneration (p=0.07) than the treated groups. In the rumen, the MA group showed thickening of the keratin layer and connective tissue, suggesting an increased absorptive area. The average daily gain was higher in the MA group (1.362 kg) than in the control (1.198 kg) and IA (1.266 kg) groups. Feed conversion efficiency was higher in the MA group (7.37) than in the control (7.94) and IA (8.47) groups. These results confirm that adsorbents, particularly MA, mitigate the adverse effects of mycotoxins, thereby enhancing the performance and health of feedlot steers.
Key words: Feed conversion. Fumonisin. Hepatic degeneration. Pneumonia. Reactive oxygen species (ROS).

Resumo

Micotoxinas são contaminantes frequentes em dietas de bovinos, podendo comprometer o desempenho e a saúde animal. Apesar da eficácia de adsorventes em aves e suínos, estudos com bovinos confinados, especialmente com dietas naturalmente contaminadas, ainda são escassos. Esta pesquisa avaliou o impacto de adsorventes misto (AM) e inorgânico (AI) na saúde hepática, ruminal, respiratória, imunidade e desempenho produtivo de 36 novilhos ½ Angus-Nelore (501,7 ± 33,2 kg de peso médio, 11 ± 5 meses de vida) alimentados com dietas naturalmente contaminadas por aflatoxina (AFLA), zearalenona (ZEA), desoxinivalenol (DON) e fumonisina (FM), durante 63 dias. Os tratamentos incluíram controle (C), AM (bentonita, parede celular de levedura e silimarina) e AI (bentonita, clinoptilolita e carvão ativado de babaçu), diariamente por 63 dias. Avaliaram-se parâmetros imunes, hepáticos, respiratórios, ruminais e desempenho animal em D0, D21, D42 e D63. Imunologicamente, o AM apresentou maiores contagens de neutrófilos e linfócitos, enquanto o AI exibiu apenas maiores contagens de neutrófilos em D63 em comparação ao controle, e ambos tratamentos promoveram maior fração albumina/globulina indicando resposta imunológica mais eficiente. No perfil hepático, a C apresentou tendência a ter teores mais altos de AST (p=0,07) e degeneração hepática (p=0,07) que os grupos tratados. No rúmen, o AM apresentou espessamento de estrato córneo e tecido conjuntivo em relação aos demais tratamentos, sugerindo uma maior área absortiva. No desempenho, o ganho de peso diário (GPD) foi maior no grupo AM (1,362 kg) comparado ao controle (1,198 kg) e ao AI (1,266 kg). A conversão alimentar foi melhor no AM (7,37) em relação ao controle (7,94) e ao AI (8,47). Os resultados confirmam que os adsorventes, principalmente o AM, atenuam os danos das micotoxinas, melhorando o desempenho e a saúde dos novilhos confinados.
Palavras-chave: Conversão alimentar. Degeneração hepática. Espécies reativas de oxigênio (ERO). Fumonisina. Pneumonia.

Introduction

Finishing steers in fedlot is a strategy to maximize productivity in limited areas, through the use of grain-rich diets characterized by rapid ruminal fermentation (Zanella et al., 2022). However, the high temperatures and humidity typical of the Brazilian climate pose a significant risk, favoring the contamination of these grains by mycotoxins, toxic metabolites produced by filamentous fungi that can proliferate during planting, harvesting, or even during storage (Ogunade et al., 2018).

Mycotoxins such as aflatoxins (AFLA), fumonisins (FM), deoxynivalenol (DON), zearalenone (ZEA), and ochratoxin A (OTA) are frequently found in the diets of Brazilian cattle, including in feed, silage, and by-products (Biscoto et al., 2022; Custódio et al., 2017). Besides that, the concentrations of mycotoxins detected in these diets were within the limits established by Brazilian legislation, which imposes restrictions solely on AFLA (Ministério da Agricultura e Pecuária [MAPA], 2025). Nonetheless, these levels were still capable of causing reduced animal performance and hepatic lesions (Batista et al., 2023). Although chronic exposure to and the simultaneous presence of multiple mycotoxins have been associated with immunosuppression and decreased productive performance in animals, international regulations and guidelines are therefore more restrictive (Chhaya et al., 2022; Elliott et al., 2019).

One of the most promising measures to reduce mycotoxicosis is the use of adsorbents added to the diet, which form stable and non-absorbable complexes with the toxins in the gastrointestinal tract, thereby decreasing their bioavailability (Ogunade et al., 2018). Organic adsorbents, composed of plant fibers, yeasts, and *Trichoderma* spp. enzymes, show significant potential in combating AFLA, DON, ZEA, and OTA. In contrast, inorganic adsorbents, based on phyllosilicates such as bentonite, zeolite, and activated charcoal, exhibit more specific action against AFB1, albeit with limited effectiveness against other mycotoxins (Xu et al., 2022).

While adsorbents such as β-glucan, montmorillonite, and bentonite have shown effectiveness in mitigating the effects of mycotoxins in cattle (Oliveira et al., 2021), there is a pressing need for more comprehensive and standardized in vivo testing. Most of the available tests are conducted in vitro, which may not accurately reflect the effects observed *in vivo*, where factors such as animal metabolism, dietary interactions, ruminal microbiota, and physiological responses significantly influence the outcomes (Debevere et al., 2020; Kihal et al., 2020).

Thus, a systematic approach becomes essential to compare the efficacy of mycotoxin adsorbents in naturally contaminated diets, especially in intensive production systems where exposure is more common. In this context, the present study evaluated the effect of two specific adsorbents, a mixed absorbent and an inorganic absorber, on the immunity, health, and performance of feedlot cattle fed a naturally contaminated diet.

Material and Methods

This experiment was approved by the Animal Use Ethics Committee (CEUA-UNICENTRO: 17/2024).

Animals, experimental design, and treatments

Thirty-six intact male ½ Angus × Nelore steers were used, with an average initial body weight (BW) of 501.7 ± 33.2 kg and an average age of 11 ± 5 months. All animals had been previously dewormed. The experimental design was a completely randomized design comprising a control diet (C), without the inclusion of mycotoxin adsorbents; a diet with a mixed adsorbent (MA), based on bentonite (>690.00 g kg^{-1}), yeast cell wall, and silymarin (>500.00 mg kg^{-1}) (Mastersorb Gold®, Grasp Indústria e Comércio Ltda., Curitiba-PR, 10 g $animal^{-1}$ day^{-1}); and a diet with an inorganic adsorbent (IA), comprising bentonite, clinoptilolite, and activated babassu charcoal (Notox LS®, Mineração e Pesquisa Brasileira Ltda., Cargill Alimentos Ltda., Urussanga-SC, 10 g $animal^{-1}$ day^{-1}).

Feeding management

The animals originated from a commercial farm (50 km from the experimental site) and were housed in feedlot pens (two animals per pen, 15 m² area). Each pen contained a concrete feed bunk (2.30 m length, 0.60 m width, 0.35 m depth) and an automatic drinking water dispenser. The allocation of animals to the experimental units was based on the BW recorded on the final day of the adaptation period.

The experimental period lasted 64 days, preceded by a 21-day adaptation period during which the original diet (50% corn silage and 50% concentrate) was gradually replaced with the experimental diet (35% corn silage and 65% concentrate). The diets were provided *ad libitum*, and the beginning of adsorbent supplementation was designated as Day 0 (D0). Animal health and performance were evaluated every 21 days (D0, D21, D42, and D63). The animals were fed twice daily at 06h30 and 17h30. Voluntary feed intake was recorded daily by weighing the amount of feed offered and orts from the previous day. Feed intake was adjusted daily to maintain feed refusal at approximately 3% of the total feed offered, in order to avoid component diet selectivity. The diet was provided as a total mixed ration (TMR) (Table 1). The adsorbents or the concentrate isolate (control treatment) were homogenized with 50 g of concentrate and applied *top-dressed* daily at a small amount in the TMR. After these consumptions, the total TRM was offered for animals.

Table 1
Chemical composition of corn silage and concentrate, average values of the experimental diet based on total dry matter, and average mycotoxin concentration in the diet of feedlot steers

Chemical composition	Corn silage	Concentrate[1]	Experimental diet (35% Corn Silage 65% concetrate)
Dry Matter, (DM) g kg^{-1}	281.4	892.9	678.9
Mineral Matter, g kg^{-1} DM	42.5	81.1	67.6
Crude Protein, g kg^{-1} DM	78.4	161.3	132.3
Ether Extract, g kg^{-1} DM	36.2	39.2	38.1
Starch, g kg^{-1} DM	288.2	386.4	352.0
Neutral Detergent Fiber, g kg^{-1} DM	500.0	268.3	349.4
Acid Detergent Fiber, g kg^{-1} DM	299.0	96.0	167.0
Lignin, g kg^{-1} DM	43.9	13.6	24.2
Total Digestible Nutrients, g kg^{-1}	673.1	813.4	764.3
Mycotoxins	**Experimental diet (μg kg^{-1})***		**Level of concern[2] (μg kg^{-1})**
Desoxynivalenol	3455		1423
Aflatoxin (B1+B2+G1+G2)	10		12
Fumonisins (B1+ B2)	89944		4544
Zearalenone	642		100

Experimental diet- 35% corn silage and 65% concentrate Concentrate Composition: wheat bran (16.0%), soybean meal (5.1%), soybean hulls (14.0%), barley rootlets (5.0%), corn germ (23.0%), barley grains (5.0%), ground corn (25.0%), degummed soybean oil (2.8%), sodium chloride (0.6%), calcitic limestone (2.0%), common salt (0.5%), cattle-grade urea (0.5%), and vitamin–mineral premix* (0.5%) [1]Level of guarantee of the premix per kg of concentrate: vit. A: 14,000 IU; vit. D3: 1,800 IU; vit. E: 75 IU; Monensin sodium: 40 mg; S: 0.70 g; Mg: 0.12 g; Na: 3.0 g; Co: 1.0 mg; Cu: 18 mg; I: 1.1 mg; Mn: 29.0 mg; Se: 0.35 mg; and Zn: 72.2 mg. *based on the calculation of the percentages of silage and concentrate. [2]Source: mycotoxin Level of concern[2] acording Batista et al. (2023).

Mycotoxin analyses were conducted at the Laboratory of Mycotoxicological Analyses of the Federal University of Santa Maria, utilizing validated protocols. Mycotoxins were identified and quantified using liquid chromatography (HPLC-Agilent 1100, Agilent Technologies, Santa Clara, CA) coupled with tandem mass spectrometry (5000 spectrometer, Applied Biosystems, Framingham, MA). This methodology enabled the simultaneous detection of aflatoxins (AFLA) B1, B2, G1, G2, zearalenone (ZEA), deoxynivalenol (DON), and Fusarium toxins B1 and B2 (FB1 and FB2) in a single chromatographic run. The limits of quantification for each mycotoxin, expressed in μg kg^{-1} of dry matter (DM), were as follows: AFLA=1; ZEA=10; DON=140; and fumonisins=50. The diet micotoxyne concentration was calculated based on the percentages of corn silage and concentrate. The average concentrations of the experimental diet are presented in Table 1.

Analyses

At the initial (D0), intermediate (D21 and D42), and final (D63) feedlot periods, blood samples were collected by coccygeal venipuncture into vacuum tubes for immune function assessment, including measurement of leukocyte oxidative metabolism, leukogram, and globulin levels. Inflammatory status was evaluated using rectal temperature measurements. Indicators of respiratory disease were assessed based on the presence of mucopurulent nasal discharge and orbital temperature. Hepatic status indicators were determined by measuring serum gamma-glutamyl transferase (GGT), aspartate aminotransferase (AST), total protein, and albumin levels. Performance indicators included: BW; average DM intake (DMI) expressed in kg animal^{-1} day^{-1}; average DMI as a percentage of BW (DMI, %BW); average daily gain (ADG, kg day^{-1}); and feed conversion ratio (FCR, kg^{-1}). Macroscopic and microscopic evaluation of the lungs, liver, and rumen was performed during slaughter at 64 day.

Leukocyte oxidative metabolism was evaluated in heparinized blood samples (4 mL) using a quantitative nitroblue tetrazolium (NBT) assay stimulated with PMA (Zdepski et al., 2023). Blood samples were collected in tubes containing 4 mL sodium ethylenediaminetetraacetic acid (EDTA) and leukograms were obtained using an automatic cell counter (SDH-3 VET; Labtest, São Paulo, Brazil). Cellular populations were classified on stained blood smears based on the morphological and staining characteristics observed under a light optical microscope.

Hepatic enzyme activity, and protein and albumin concentrations were analyzed in blood samples collected in vacuum tubes without anticoagulants (8 mL). Serum globulin concentrations were calculated by subtracting serum albumin from total serum protein. The blood was centrifuged (2,800 × g for 15 min), and the supernatant serum was separated, aliquoted, and stored frozen at -20 °C until analysis. Total protein, albumin, GGT, and AST were then measured using biochemical analyses with specific commercial kits (Labtest®, Lagoa Santa, MG, Brazil).

On the day of slaughter, the livers were evaluated macroscopically for congestion, steatosis, and presence of hepatic abscesses. Subsequently, hepatic tissue fragments (2 cm²) were preferentially collected from the transition zone between the normal tissue and lesion areas. These fragments were subjected to histopathological analysis to classify the degree of hepatic steatosis, degeneration, and necrosis, using a scoring system ranging from 1 to 3, with a score of 3 indicating the most severe tissue damage.

The evaluated respiratory disease indicators were nasal discharge and orbital temperature. The assessments were conducted over eight days in each experimental period (four days before and four days after collect days), always before morning feeding. Nasal secretions were scored as follows: 1, serous secretion; 2, mucous secretion; 3, small mucopurulent secretion; and 4, large mucopurulent secretion. The median score was calculated as described by Zdepski et al. (2023). The animals' orbital temperatures were measured using infrared thermometry (Fluke®, Whan, China), and the mean values were calculated for each experimental time point. On the day of slaughter, the animals' lungs were evaluated based on the macroscopic characteristics of lobar consolidation using the following scoring system: 1, absence of lesion; 2, lesion involving 10–50% of the affected lobe; 3, lesion involving 50–75% of the lobe; and 4, lesion involving more than 75% of the lobe, according to the criteria described by Zdepski et al. (2023). Subsequently, pulmonary fragments were collected, preferably from the transitional area between the normal tissue and areas of consolidation (when present). Approximately 2 cm² of each fragment was fixed in 10% formalin and processed for the preparation of histopathological slides stained with hematoxylin and eosin (HE). Under light microscopy (400x), alveolar or interstitial inflammatory infiltrates were classified according to the following scores: 1, absence of infiltrate; 2, infiltrate affecting 10%–50% of the analyzed fragment; 3, infiltrate affecting 50%–75%; and 4, infiltrate affecting more than 75%. Data are expressed as the frequency of lesions.

On the day of slaughter, fragments were collected from the ventral sac of the rumen (5 cm²) and preserved in 10% formalin until tissue fixation. The samples were then embedded in paraffin, sectioned, and stained with Harris HE. The microscopic variables measured included papillary thickness, stratum corneum thickness, connective tissue width, and non-keratinized epithelium of 10 papillae per animal. Measurements were performed using the Leica Qwin image analyzer, coupled to a Leica optical microscope at 40× magnification, and utilizing the ImageJ 1.43 software (ImageJ, National Institutes of Health, Bethesda), as described by Zdepski et al. (2023). Inflammation scores were determined by neutrophilic infiltration in the ruminal epithelium using scores of 1–3 (3 scores indicated the highest accumulation of inflammatory cells).

Animal performance was evaluated after a ten-hour solid feed withdrawal. The evaluated parameters included BW, average DMI expressed in kg animal^{-1} day^{-1} (DMI, kg day^{-1}), average DMI expressed as a percentage of BW (DMI, % BW), ADG (kg day^{-1}), and FC (kg^{-1}).

The DMI (kg day^{-1}) was measured by calculating the difference between the amount of feed offered daily and the amount of feed refused from the previous day. The DMI as a percentage of BW (% BW) was calculated by the ratio of DMI to the mean BW during the period, multiplied by 100 ($DMI\%BW = DMI \div BW * 100$). The ADG was determined by the difference between final BW (BWf) and initial BW (BWi) divided by the number of days in the evaluation period ($ADG = BWf - BWi \div 21$). The FC was calculated by the ratio between DMI and ADG ($FC = DMI \div ADG$).

Statistical analysis

For health analysis, 12 replicates were used, and each animal was considered an independent experimental unit. For the performance evaluation, six replicates were considered, with each pen serving as an experimental unit. The UNIVARIATE procedure was used to detect outliers. Subsequently, ordinal data that met the assumptions of normality (Kolmogorov–Smirnov test) and homogeneity of variances (Bartlett's test).

The analysis of each variable followed the statistical model: $Yijk = \mu + \alpha i + \varepsilon ij$; where

Yijk represents the dependent variables, μ is the overall mean of all observations, αi is the fixed effect of the treatment of order "i" (with 1=control diet without mycotoxin adsorbent; 2=diet with MA; 3=diet with IA), and εij corresponds to the random residual error. The parametric data were analyzed using the GLM PRO MIXED procedure to evaluate the interaction treatment X day effect (Trtxday) (Statistical Analysis System Institute [SAS Institute], 1993). Tukey's test was applied when an interaction treatment X time effect was observed ($p \leq 0.05$), and data were expressed as mean and standard deviation. Data that did not meet the assumptions of normality or homogeneity (no parametrics) were evaluated using Friedman test and Dunn's test as pos test, and the day interaction was analised by kruskul wallis. These results were expressed as medians and 5- 95% confidence intervals (CIs). Differences were considered statistically significant when $p \leq 0.05$, and considered as trends when $0.05 < p \leq 0.10$

Results and Discussion

Mycotoxin-containing diets are challenging for animals, particularly in terms of immunity, liver health, and performance. Mixed adsorbent used in this study mitigated these effects, and inorganic adsorvent had an intermediate effect, basically on livhepatic profile.

The experimental diet used (Table 1) showed higher levels of mycotoxin contamination than those reported in the literature (Biscoto et al., 2022; Custódio et al., 2019), but were still within the limits established by the European Union for AFLA and exceeded the limits for DON, ZEA and FM. Batista et al. (2023) found that a diet containing fewer mycotoxins compared to the present study (TMR: FM 4,544 ppb, AFLA 12.2 ppb, trichothecenes 1,423 ppb, and ZEA 100 ppb), administered for a 29-day feedlot period, was already sufficient to impair animal performance and immunity, although it did not affect liver health. Based on Batista et al. (2023) experiment, the diet contaminated by mycotoxins used in their study reduced the intake as well as digestibility of neutral detergent fiber and altered the profile of short-chain fatty acids, increasing isobutyric acid production, which, in turn, reduced ruminal pH. Moreover, it increased ketogenesis and decreased animal immunity, as evidenced by reductions in IgA, lymphocyte count, and neutrophil-to-lymphocyte ratio. In the present study, diet-induced effects were similar to those reported by Batista et al. (2023). This reinforces the need for Brazilian legislation to reconsider mycotoxin limits.

Immunity

There was an interaction treatment day for leukocyte counts and lymphocyte populations, which were higher in MA than C (p Trtxd =0.05 and p Trtxd =0.03), and an interaction treatment day for albumin:globulin ratio (A:G), which was higher in MA and IA than C (Trtxd =0.03, Table 2). Time affects all variables (oxidative metabolism, leukocytes, neutrophils, lymphocytes, serum globulins, and the albumin:globulin ratio). They were higher at D0 than at other times (p day < 0,05) (Table 2). Most animals exhibited leukocytosis, characterized by neutrophilia and lymphocytosis, at the first collection (D0, P>0,05) (Table 3), possibly due to stress induced by the collection procedure, a phenomenon reported by Virmond et al. (2020) and Zdepski et al. (2023). This stress was more intense during the initial stages of adaptation to the sampling procedure, likely due to increased adrenaline release (Tizard, 2014).

Table 2
Immunological variables of steers fed with a diet containing mycotoxins according to the adsorbent used, during 64 days of feedlot

Parameters	Treatments			P Value			Reference values
	C	MA	IA	Trt	Day	Trtxday	
Oxidative Metabolism[1] (d.o.)	0,34 0,31-0,36	0,32 0,31-0,36	0,32 0,31-0,36	0.5	0.02	0,39	
Leukocytes[1] (x10 cel mm^{-3})	10,2b 9,9-12,9	12,4a 12,0-14,9	11,05ab 10,5-12,8	0.04	0.0001	0,05	4,0-12,0[3]
Neutrophils[1] (x10 cel mm^{-3})	1,73 1,5-2,7	1,92 1,5-2,8	2,04 1,8-2,4	0.10	0.0001	0,09	0,6-4,0[3]
Lymphocytes[1] (x10 cel mm^{-3})	7,64b 7,4-9,7	9,05[a] 8,7-10,8	7,97ab 7,1-8,7	0,03	0.0001	0,03	2,5-7,5[3]
Globulin (g dL^{-1})	6,19 1,6	5,78 1,7	5,80 1,7	0,19	0,0001	0,19	2,1-9,0[4]
Albumin: Globulin[1]	0,46b 0,19	0,61a 0,34	0,62a 0,34	0,02	0,0001	0,03	0,6-0,9[4]

C- Control, MA- mixed adsorbent, IA- inorganic adsorbent. o.d- optical density. [1]Median and 5% and 95% CI (confidence interval) were evaluated by Friedman test (treatment X day interaction: P value Trtxday), Kruskal wallis (p value day) and Dunn's pos tests (Trt: p value treatment) (. [2]Means and SD (standard deviation) were evaluated by GLM Pro mixed model (P value Trtxday and p value day) and Tukey's test (p value treatmen- Trt) P. Different lowercase letters (a, b) in the same line indicate difference between treatments p value ≥0,05, reference values [3]Radostits et al. (2000), 4Tothova et al. (2016).

Table 3
Leukocytes, neutrophils and lymphocytes (median and 5 and 95% confidence interval) of feedlot steers fed with diets containing mycotoxins according to adsorbent treatment.

Parameters		Treatments			P trt
		C	MA	IA	
Leukocytes ($\times 10^3$ cells$\cdot$mm^{-3})	D0	18,51 (12,9-21,5)A	19,55 (16,321,8)A	15,15 (12,317,8)A	0,15
	D21	9,52 (8,3-10,7)B	10,12 (8,6-11,1)B	9,00 (8,1-11,6)B	0,81
	D42	8,70 (7,7-10,7)B	12,50 (9,9-13,7)B	10,80 (8,6-12,4)B	0,11
	D63	9,85 (7,9-11,2)B	11,20 (10,2-15,4B	9,90 (8,1-12,0)B	0,21
	P day	0,004	0,002	0,008	
Neutrophils ($\times 10^3$ cells$\cdot$mm^{-3})	D0	2,80 (1,7-3,4)	3,29 (2,6-4,3)A	2,37 (1,5-2,9)	0,17
	D21	1,05 (0,9-1,5)	1,16 (0,9-1,7)B	1,93 (1,3-2,5)	0,19
	D42	1,77(1,2-2,1)	1,92 (1,4-3,3)AB	1,75 (1,3-2,6)	0,49
	D63	1,65 (0,9-2,1)b	1,97 (1,4-3,1)aAB	2,56 (1,8-3,0)a	0,02
	P day	0,06	0.01	0.09	
Lymphocytes ($\times 10^3$ cells$\cdot$mm^{-3})	D0	13,36 (9,3-16,1)A	13,91 (11,4-15,7)A	10,66 (8,8-13,2)A	0,38
	D21	7,67 (6,5-8,7)B	8,31 (6,8-9,2)B	6,43 (5,7-8,6)B	0,51
	D42	6,26 (5,3-8,1B	8,60 (7,0-9,8)B	8,12 (6,4-9,8)AB	0,15
	D63	6,56 (5,4-8,0)bB	8,29 (7,5-11,4)aB	7,33 (6,1-9,4)abAB	0,05
	P day	0,0009	0,003	0,04	
Albumin: Globulin ratio	D0	0,36 (0,2)	0,35(0,1)A	0,38(0,1)A	0,91
	D21	0,48(0,2)	0,51(0,2) AB	0,60(0,42)AB	0,60
	D42	0,46(0,2)	0,64 (0,4)B	0,75(0,3)B	0,08
	D63	0,55(0,1)a	0,90 (0,3)bB	0,75 (0,3)abB	0,01
	P day	0,09	0,01	0,01	

C- control, MA- mixed adsorbent, IA- inorganic adsorben P value: Trt- treatment effect (kruskall walis and Dunn pos test), P day-time effect (Friedman and Dunn pos test), 1Different lowercase letters (ab) in the same row indicate treatment effect and different uppercase letters (AB) in the same colun indicate time effect on the same treatment test, p value <0.05.

At subsequent time points, the animals became more adapted to the experimental conditions, and thus, most no longer exhibited leukocytosis but rather relatively mild lymphocytosis (Table 3). Although C showed a trend toward lower neutrophil counts compared to MA and IA (p Trtxd =0.09), particularly on D63 for neutrophils (p Trt=0.02) and at same day compared to MA for lymphocytes (pTrt= 0.05).

Roberts et al. (2021) observed a significant reduction in the proportion of lymphocytes and their CD4+ and CD8+ subpopulations in feedlot steers that were fed a mycotoxin-contaminated diet. Pestka et al. (2004) found that exposure to high doses of DON promotes leukocyte apoptosis, resulting in immunosuppression. This effect is time-dependent, explaining the reduction in circulating neutrophils and lymphocytes over time, which is more pronounced on D63 in C.

Besides, both the additives attenuated this effect, probably by minimizing the absorption of dietary mycotoxins. The effect of MA on immunity was more pronounced because it contains yeast extract, which acts as an immunostimulant, enhancing lymphocyte replication, immunoglobulin production, and phagocyte activation (Virmond et al., 2020).

Oxidative metabolism, also known as the leukocyte respiratory burst, is a process conducted by phagocytes that produce reactive oxygen species (ROS), which are oxidant compounds responsible for the elimination of internalized pathogens, during phagocytosis. A reduction in phagocytic activity indicates immunosuppression and increased susceptibility to disease (Tizard, 2014). Therefore, mycotoxins were expected to interfere with this parameter, as they can inhibit intracellular ROS production and decrease cellular antioxidant capacity (Mavrommatis et al., 2021).

The absence of changes in leukocyte oxidative metabolism among the treatments suggests the 63-day intoxication may have been insufficient to impair this physiological function. Studies involving immunostimulants in feedlot cattle have shown an increase in this parameter between 40 and 72 days of use (Zdepski et al., 2023). Additionally, mycotoxin adsorbents based on bentonite or mixed formulations increased the oxidative metabolism of leukocytes in dairy cows after 16–24 days of use (Oliveira et al., 2021; Di Domenico et al., 2022), indicating that the composition of the adsorbents, challenge, and duration of mycotoxicosis can influence this parameter.

Globulins are serum proteins composed of alpha, beta, and gamma fractions. The gamma fraction is the most abundant, and its increase generally indicates activation of the immune system, whereas its decrease indicates immunosuppression (Tothova et al., 2016). Although immunoglobulin detection using ELISA is more sensitive, a substantial increase in immunoglobulin levels may lead to elevated globulin levels (Alberghina et al., 2011). Because the average globulin concentrations in all treatments were within the reference range and were not influenced by the treatments, it can be inferred that the mycotoxins present in the diet did not affect serum globulin.

The A:G ratio is inversely proportional to the production of acute-phase proteins, which are early markers of inflammation (Tothova et al., 2016). The A:G ratio was lower in C than in the reference parameter, indicating the depletion of albumin in globulin synthesis (Alberghina et al., 2011; Tothova et al., 2016). Furthermore, the A:G ratio in MA was higher than C at D63 (p Trt=0.03) and a trend toward a higher A:G ratio in IA than C at D42 (p Trt=0.08, Table 3), within physiological parameters, suggesting the effectiveness of these treatments in mitigating inflammatory processes.

Hepatic profile

There was an interaction treatment day only for serum AST, being higher in C than in animals treated with the adsorbents (p TrtXday=0.02; Table 4), at D 42 (p Trt=

0.03, Table 5). Serum AST, GGT, protein, and albumin levels were influenced by day, being higher at D21 than on the other days (p day < 0,05; Table 5). When each treatment was analysed by day, serum AST was higher at D63 than at D21 for all groups (p day < 0,05, Table 4).

Table 4
Indicators of liver lesion of steers fed with a diet containing mycotoxins according to the adsorbent used, during 64 days of feedlot

Parameters	Treatments			P value			Reference parameters
	C	MA	IA	Trt	Day	Trtxday	
AST (UI L^{-1})	73,00a 71,5-90,8	68,00b 62,3-75,0	68,00b 63,3-76,0	0,02	0,0001	0,02	78-1321
GGT (UI L^{-1})	22,00 23,2-29,2	22,00 22,5-28,6	28,57 25,9-31,2	0,12	0,0001	0,14	0-482
TSP (g dL^{-1})	8,45 8,3-9,6	7,80 7,7-8,9	8,15 7,8-9,1	0,13	0,0001	0,44	5,7-8,11
ALB(g dL^{-1})	2,75 2,6-3,0	2,86 2,7-3,2	3,05 2,7-3,2	0,37	0,001	0,86	2,1-3,61
Liver lesion escore							
BS (escore)	1,5 1,3-2,3	2,0 1,3-2,3	1,0 1,0-2,1	0,68			
LD (escore)	2,0 1,3-2,5	1,0 1,3-2,4	1,0 1,01-2,1	0,06			
LN (escore)	1,5 1,2-2,2	1,5 1,2-2,4	1,0 1,1-2,2	0,88			

AST- aspartate aminotransferase, GGT- gamma glutamyl transferase, TSP- total serum protein, ALB- albumin, BS- biliary stasis, LD- liver degeneration, LN- liver necrosis, C- control, MA- mixed adsorbent, IA- inorganic adsorbent.Median and 5% and 95% CI (confidence interval) were evaluated by Friedman test (treatment X day interaction: P value Trtxday) Kruskal wallis (p value day) and Dunn's pos tests (p value treatment:Trt) . Different lowercase letters in the same row indicate time interaction, p<0.05. [1] Kaneko et al. (2008), [2] Thrall (2007).

Table 5
Serum AST ((median and 5 and 95% confidence interval) of steers fed with a diet containing mycotoxins according to adsorbent treatment

Parameters		Treatments			P trt
		C	MA	IA	
AST (UI L^{-1})	D0	73AB 54,5-116,4	73AB 58,9-90,4	68AB 61,7-94,0	0,81
	D21	54,5A 45,6-81,1	52,0A 45,3-82,9	52,0B 42,9-61	0,50
	D42	80,5aAB 68,1-104,5	68,0bAB 56,7-72,7	68,0bAB 58,7-70,6	0,03
	D63	83,0B 74,7-96,9	70,5B 57,9-105,2	75,5A 67,6-90,5	0,30
	P day	0,02	0,03	0,002	

C- control, MA- mixed adsorbent, IA- inorganic adsorben P value: Trt- treatment effect (kruskall walis and Dunn pos test), P day-time effect (Friedman and Dunn pos test), 1Different lowercase letters (ab) in the same row indicate treatment effect and different uppercase letters (AB) in the same colun indicate time effect on the same treatment test, p value <0.05.

Regarding macroscopic liver lesions, hepatic steatosis was the most common, with no treatment effect observed. The most prevalent microscopic findings were bile stasis and liver necrosis, both of which were unaffected by treatment. Only the liver degeneration score was influenced by treatment, showing a trend toward lower severity in animals treated with adsorbents compared to controls (p Trt=0.06, Table 4).

Mycotoxins such as FB1 compromise cellular antioxidant systems and interfere with the intracellular signaling of target cells, primarily due to increased ROS production and intensified lipid peroxidation, particualrly in the liver (Weinert, 2020). This suggests that the adsorbents mitigated liver conditions in animals. Additionally, silymarin in MA may have contributed to protection. The polyphenolic fraction of silymarin interferes with lipid metabolism, preventing fat accumulation and, consequently, steatosis, which can lead to degeneration or necrosis, depending on severity. Furthermore, its anti-inflammatory action helps protect the liver by reducing the production of inflammatory cytokines, thereby preventing apoptosis and tissue damage (Pinheiro et al., 2021).

Based on these results, hepatic fat infiltration (steatosis) led to moderate degeneration (score=2) and mild hepatocellular necrosis (score=1.5) (Table 4). The trend of the treated animals to present lower severity of hepatic degeneration can justify the lower levels of serum AST compared to C. AST is released into the bloodstream when there is damage to the hepatocyte membrane caused by necrosis or hepatic degeneration, which is more pronounced in the case of necrosis (Kaneko et al., 2008). Because the hepatic necrosis score was low (mean score of 1.5) and the treatment only influenced the hepatic degeneration score, it was inferred that the rupture of hepatocytes was not significant (Kaneko et al., 2008).

Nonetheless, the greater intensity of hepatic degeneration in group C was reflected in a trend toward more pronounced hepatic degeneration compared with the animals treated with adsorbents.

FB was present in higher concentrations in the experimental diets. It causes severe liver and gastrointestinal lesions in a dose-dependent manner (Upadhaya et al., 2010). Jennings et al. (2020) observed only alterations in hepatocytes with no visible macroscopic hepatic lesions in their studies with feedlot cattle exposed to different FB doses. If the animals remained in the feedlot for a longer period, hepatic alterations would have been more pronounced, and the effects of the adsorbents would have been more significant.

Albumin and serum proteins are markers of liver function, and their synthesis is reduced when approximately 70% of the liver becomes insufficient. Although the albumin levels were within the reference range, some animals showed elevated serum protein concentrations. This may have occurred due to an immune response, with increased immunoglobulin production in an attempt to combat infectious conditions, such as bovine respiratory disease complex (BRD), or due to dehydration associated with subacute ruminal acidosis syndrome (SARA), both of which are common disorders in feedlot systems (Sato, 2015).

An increase in serum GGT level indicates cholestasis (Kaneko et al., 2008). There is wide variability in the literature regarding the normal activity range of this enzyme in bovine species. Although Kaneko et al. (2008) reported lower activity levels, Thrall et al. (2015) reported higher values, consistent with those found in previous studies (Oliveira et al., 2021; Batista et al., 2023). Given that the hepatic steatosis score ranged from mild to moderate (scores of 1 to 2), this alteration was not highly expressive, which explains the lack of difference between the treatments for this variable.

Pulmonary status

Mycotoxins in the diet lead to immunosuppression, increasing the occurrence of BRD in feedlot cattle (Timsit et al., 2011). The orbital temperature was influenced only by day, being higher at D63 than at D21 (p=0.001), when the ambient temperature was higher than on the other days of the experiment. Most animals exhibited small amounts of mucopurulent nasal discharge (score 2), which was not influenced by day or treatment (p Trtxday=0.12, Table 6). These methods of pneumonia detection serve as alternatives to pulmonary auscultation and can be performed without restraining the animal; however, they are low-sensitivity tests, and therefore direct lung inspection remains essential (Zdepski et al., 2023).

Table 6
Indicators of respiratory disease of steers fed with a diet containing mycotoxins according to the adsorbent used, during 64 days of feedlot

Parameters	Treatments			P value		
	C	MA	IA	Trt	Day	Trtxday
Orbital Temperature ºC[1]	28,90 0,80	29,20 0,90	29,21 1,0	0,47	0,001	0,97
Nasal Score[2]	2,00 2,0-2,0	2,00 1,9-2,3	2,00 2,0-2,0	0,12	0,36	0,12
Pulmonary score						
Macroscopic Score[2]	1,50 1,5-2,0	2,50 1,5-2,7	1,50 1,2-2,6	0,32		
Neutrophil Infiltrate Score[2]	1,00 1,0-2,2	1,45 1,4-2,5	1,23 1,2-2,4	0,31		

C- Control, MA- mixed adsorbent, IA- inorganic adsorbent. [1]Means and SD (standard deviation) were evaluated by proc Mixed model (P value Trtxday and p value day) and Tukey's test (p value treatmen- Trt) [2]Median and 5% and 95% CI (confidence interval) were evaluated by Friedman test (treatment X day interaction: P value Trtxday), Kruskal wallis (p value day) and Dunn's pos tests (Trt: p value treatment).

In the pulmonary analysis, most animals (58.34%; 21/36) showed no macroscopic lung alterations, regardless of treatment. In animals with macroscopic pulmonary lesions, there was no treatment influence on neutrophil infiltrate score (p Trt=0.31, Table 6)

The lack of treatment effects on respiratory indicators may be because the diet was not sufficiently challenging for pulmonary immunity, as the feedlot period was short (only 64 days). Notably, mycotoxin-induced immunosuppression is a time-dependent phenomenon (Luna-López et al., 2025).

Animal performance

There was a tendency for the daily weight gain of the animals treated with the adsorbent to increase in relation to the C group (pTrt=0.09, Table 7). Custódio et al. (2019) also found that the use of adsorbents promoted partial recovery of performance in cattle fed a diet with lower levels of mycotoxin contamination, results similar to those of our study. Our treatments did not influence DMI, expressed as kg per day or per 100 kg of BW (DMI, % BW), indicating that neither dietary contamination nor adsorbent use affected feed intake (Table 7).

Table 7
Average daily weight gain (ADG), dry matter intake expressed in kg day^{-1} (DMI, kg day^{-1}) or per 100 kg of body weight (DMI, % BW), and feed conversion ratio (FC) of steers fed with a diet containing mycotoxins according to the adsorbent used, during 64 days of feedlot

Period	Treatments			Mean	CV	P Trt
	C	MA	IA			
ADG, kg day^{-1}: 0 a 64 days	1,198	1,362	1,266	1,276	9,34	0,09
DMI, kg day^{-1}: 0 a 64 days	9,80	9,89	10,33	10,01	8,25	0,50
DMI, % BW: 0 a 64 days	1,82	1,86	1,93	1,87	6,94	0,34
FC: 0 a 64 days	7,94a	7,37b	8,47a	7,93	11,07	0,03

C- Control, MA- mixed adsorbent, IA- inorganic adsorbent. 1Different lowercase letters (ab) in the same row indicate treatment effect (p valeuTrt <0.05), GLM Pro mixed model and Tukey pos test .CV: Coefficient of variation.

Regarding FC, the MA treatment showed better results than the other treatments (p Trt=0.03). Mixed mycotoxin adsorbents adsorbed DON, ZEA, and FB1, whereas inorganic adsorbents were more effective against AFLA. Because the diet used in this study was predominantly contaminated with FB1, it is reasonable to assume that the MA treatment was more effective due to its greater affinity for this specific mycotoxin. Furthermore, naturally occurring mycotoxins in the diet impair protein synthesis and muscle development in feedlot cattle (Batista et al., 2023).

Ruminal inflammation was mild, with no significant difference between the groups (p=0.49, Table 8). However, the thicknesses of the stratum corneum (pTrt=0.03) and connective tissue (p Trt =0.03) were greater in the MA group than those in the other groups. The size and shape of the ruminal papillae adapt to dietary changes and are responsible for the increase in the absorptive area in the rumen (Oliveira et al., 2019). These morphological changes suggest a greater capacity for epithelial adaptation and a potential increase in the absorptive surface, which may have contributed to the improvement in the FC observed in the MA group. Thus, the results indicate that despite the dietary challenge posed by mycotoxins, the use of MA promoted beneficial structural changes in the ruminal epithelium, thereby favoring the performance of feedlot cattle.

Table 8
Microscopic evaluations of ruminal papillae of steers fed with a diet containing mycotoxins according to the adsorbent used, during 64 days of feedlot

Parameters	C	MA	IA	P Trt
Total Width[1] (μm)	282,33 77,4	275,42 43,7	300,42 67,9	0,62
Unstratified Epithelium[1] (μm)	14,54 3,7	14,57 2,1	14,66 2,6	0,99
Stratum Corneum[1] (μm)	10,31b 1,9	12,66[a] 2,6	10,46b 1,9	0,03
Connective Tissue[1] (μm)	257,48ab 66,0	275,30[a] 48,1	248,19b 90,3	0,03
Inflammation Score[2]	1 1,1-1,9	2 1,2-2,0	1,5 1,1-2,0	0,77

C- Control, MA- mixed adsorbent, IA- inorganic adsorbent. [1] Values expressed as mean and standard deviation (SD), Proc mixed test and Tukey pos test, [2]Values expressed as median and 5% and 95% confidence interval (CI), Kruskall walis test and Dunn pos test, [1]Different lowercase letters (ab) in the same row indicate treatment effect (Ptrt) p<0.05.

Conclusion

Mixed adsorbent produced a better attenuation of the adverse effects of mycotoxins, thereby enhancing the performance and health of feedlot steers. The inorganic adsorbent showed an intermediate effect, attenuating liver disease primarily.

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References

Alberghina, D., Giannetto, C., Vazzana, I., Ferrantelli, V., & Piccione, G. (2011). Reference intervals for total protein concentration, serum protein fractions, and albumin/globulin ratios in clinically healthy dairy cows. *Journal of Veterinary Diagnostic Investigation, 23*(1), 111-114. doi: 10.1177/104063871102300119

Batista, L. H. C., Siqueira, G. R., Costa e Silva, L. F., Koontz, A., Granja-Salcedo, Y. T., Ferreira, I. M., Cidrini, I. A., Oliveira, K. A., Queiroz, A. C. M. de, Rodrigues, A. N., & Resende, F. D. de. (2023). Effects of feeding mycotoxin-contaminated diets and the use of an adsorbent on the intake, digestibility, rumen fermentation, nitrogen balance and physiological parameters in the blood of Nellore steers. *Animal Feed Science and Technology, 307*(1), 115835. doi: 10.1016/j.anifeedsci.2023.115835

Biscoto, G. L., Salvato, L. A., Alvarenga, É. R., Dias, R. R. S., Pinheiro, G. R. G., Rodrigues, M. P., Pinto, P. N., Freitas, R. P., & Keller, K. M. (2022). Mycotoxins in cattle feed and feed ingredients in Brazil: a five-year survey. *Toxins, 14*(8), 1-16. doi: 10.3390/toxins14080552

Chhaya, R. S., O'Brien, J., & Cummins, E. (2022). Feed to fork risk assessment of mycotoxins under climate change influences - recent developments. *Trends in Food Science & Technology, 126*(1), 126-141.doi: 10.1016/j.tifs.2021.07.040

Custódio, L., Figueira, D. N., Gloria, E. M. D., Holder, V. B., Yiannikouris, A., Pettigrew, J. E., Kuritza, L. N., Resende, D., & Siqueira, G. R. (2017). Survey of mycotoxin contamination in feedlot diets in Brazil. *Journal of Animal Science, 95*(4), 19-19. doi: 10.2527/asasann.2017.038

Custódio, L., Prados, L. F., Yiannikouris, A., Holder, V., Pettigrew, J., Kuritza, L., Resende, F. D. de, & Siqueira, G. R. (2019). Mycotoxin contamination of diets for beef cattle finishing in feedlot. *Revista Brasileira de Zootecnia, 48*(1), 1-12. doi: 10.1590/rbz4820190079

Debevere, S., Cools, A., De Baere, S., Haesaert, G., Rychlik, M., Croubels, S., & Fievez, V. (2020). In vitro rumen simulations show a reduced disappearance of deoxynivalenol, nivalenol and enniatin B at conditions of rumen acidosis and lower microbial activity. *Toxins, 12*(2), 1-23. doi: 10.3390/toxins12020101

Di Domenico, D., Oliveira, M. S., Garbossa, G., Thomaz, G. R., Carolina, A., Depaoli, C. R., Ornaghi, M. G., Stuani, O. F., & Bertagnon, H. G. (2022). Influence of bentonite adsorbents associated with hepatic antioxidants on the health and production of dairy cows fed diets containing naturally produced mycotoxins. *Acta Veterinaria Brasilica, 16*(2), 126-133. doi: 10.21708/avb.2022.16.2.10490

Elliott, C. T., Connolly, L., & Kolawole, O. (2019). Potential adverse effects on animal health and performance caused by the addition of mineral adsorbents to feeds to reduce mycotoxin exposure. *Mycotoxin Research, 36*(1), 115-126. doi: 10.1007/s12550-019-00375-7

Jennings, J. S., Ensley, S. M., Smith, W. N., Husz, T. C., & Lawrence, T. E. (2020). Impact of increasing levels of fumonisin on performance, liver toxicity, and tissue histopathology of finishing beef steers. *Journal of Animal Science, 98*(12), 1-8. doi: 10.1093/jas/skaa390

Kaneko, J. J., Harvey, J. W., & Bruss, M. L. (Eds.). (2008). Clinical biochemistry of domestic animals (6th ed.). Elsevier Academic Press. https://doi.org/10.1016/B978-0-12-370491-7.X0001-3

Kihal, A., Rodríguez-Prado, M., Godoy, C., Cristofol, C., & Calsamiglia, S. (2020). In vitro assessment of the capacity of certain mycotoxin binders to adsorb some amino acids and water-soluble vitamins, *Journal of Dairy Science, 103*(4), 3125-3132. doi: 10.3168/jds.2019-17561

Luna-López, M. C., Valdivia-Flores, A. G., Rangel-Muñoz, E. J., Hernández-Valdivia, E., Quezada-Tristán, T., Jaramillo-Juárez, F., & Ortiz-Martínez, R. (2025). Time-dependent changes in performance, biochemistry, and histology in dairy calves with acute aflatoxicosis. *Veterinary Sciences, 12*(3), 1-14. doi: 10.3390/vetsci12030273

Mavrommatis, A., Giamouri, E., Tavrizelou, S., Zacharioudaki, M., Danezis, G., Simitzis, P. E., Zoidis, E., Tsiplakou, E., Pappas, A. C., Georgiou, C. A., & Feggeros, K. (2021). Impact of mycotoxins on animals' oxidative status. *Antioxidants, 10*(2), 1-26. doi: 10.3390/antiox10020214

Ministério da Agricultura, Pecuária e Abastecimento. (2025, 15 de maio). Portaria nº 1281, de 15 de maio de 2025. Diário Oficial da União. https://www.gov.br/agricultura/pt-br/assuntos/insumos-agropecuarios/insumos-pecuarios/alimentacao-animal/PORTARIASDA_MAPAN1.281DE15DEMAIODE2025PORTARIASDA_MAPAN1.281DE15DEMAIODE2025DOUImprensaNacional.pdf,

Ogunade, I. M., Martinez-Tuppia, C., Queiroz, O. C. M., Jiang, Y., Drouin, P., Wu, F., Vyas, D., & Adesogan, A. T. (2018). Silage review: mycotoxins in silage: occurrence, effects, prevention, and mitigation. *Journal of Dairy Science, 101*(5), 4034-4059. doi: 10.3168/jds.2017-13788

Oliveira, M. S., Di Domenico, D., Thomaz, G. R., Garbossa, G., Depaol, C. R., Abreu, A. C. A., & Bertagnon, H. G. (2021). Effect of bentonite on the health and dairy production of cows submitted to a diet naturally contaminated by mycotoxins. *Semina: Ciências Agrárias, 42*(6), 3337-3354. doi: 10.5433/1679-0359.2021v42n6p3337

Oliveira, V. da S., Santos, A. C. P. dos, & Valença, R. de L. (2019). Desenvolvimento e fisiologia do trato digestivo de ruminantes. *Ciência Animal, 29*(3), 114-132. Sem doi: Disponível em https://revistas.uece.br/index.php/cienciaanimal/article/view/10085/8374,

Pestka, J. J., Zhou, H.-R., Moon, Y., & Chung, Y. J. (2004). Cellular and molecular mechanisms for immune modulation by deoxynivalenol and other trichothecenes: unraveling a paradox. *Toxicology Letters, 153*(1), 61-73. doi: 10.1016/j.toxlet.2004.04.023

Pinheiro, J. K., Henriques, L. T., Gonzaga, S., Neto, Signoretti, R. D., Oliveira, J. S. de, Pinheiro, J. K., & Alves, R. T., Jr. (2021). Uso da homeopatia no gado de leite como modulador da função hepática: uma revisão. *CES Medicina Veterinaria Y Zootecnia, 16*(1), 29-46. doi: 10.21615/cesmvz.16.1.2

Radostits, O. M., Blood, D. C., & Gay, C. C. (2000). Veterinary medicine: a textbook of the diseases of cattle, sheep, pigs, goats and horses (9th ed.). W.B. Saunders.

Roberts, H. L., Bionaz, M., Jiang, D., Doupovec, B., Faas, J., Estill, C. T., Schatzmayr, D., & Duringer, J. M. (2021). Effects of deoxynivalenol and fumonisins fed in combination to beef cattle: immunotoxicity and gene expression. *Toxins, 13*(10), 1-14. doi: 10.3390/toxins13100714

Sato, S. (2015). Pathophysiological evaluation of subacute ruminal acidosis (SARA) by continuous ruminal pH monitoring. *Animal Science Journal, 87*(2), 168-177. doi: 10.1111/asj.12415

Statistical Analysis System Institute (1993). SAS language reference. Version 6. Cary, NC: SAS Institute Inc.

Thrall, M. A., Baker, D. C., Campbell, T. W., Denicola, D., Fettman, M. J., Lassen, E. D., Rebar, A., Weiser, G., & Fagliari, J. J. (2007). *Hematologia e bioquímica clínica veterinária.* Editora Roca

Timsit, E., Bareille, N., Seegers, H., Lehebel, A., & Assié, S. (2011). Visually undetected fever episodes in newly received beef bulls at a fattening operation: occurrence, duration, and impact on performance1,2. *Journal of Animal Science, 89*(12), 4272-4280. doi: 10.2527/jas.2011-3892

Tizard, I. R. (2014). Imunologia veterinária (9a ed.). GEN Guanabara Koogan.

Tothova, C., Nagy, O., & Kovac, G. (2016). Serum proteins and their diagnostic utility in veterinary medicine: a review. *Veterinární Medicína, 61*(9), 475-496. doi: 10.17221/19/2016-vetmed

Upadhaya, S. D., Park, M. A., & Ha, J. K. (2010). Mycotoxins and their biotransformation in the rumen: a review. *Asian-Australasian Journal of Animal Sciences, 23*(9), 1250-1260. doi: 10.5713/ajas.2010.r.06

Virmond, M. P., Rossi, P. S., Antunes, A. V., Mattei, R. I., Schllemer, N. R., Thomaz, G. R., Garbossa, G., Neumann, M., & Bertagnon, H. G. (2020). Fresh yeast additives improve immune parameters and reduce respiratory disease in heifers finished in feedlots. *Semina: Ciências Agrárias, 41*(6 Supl.2), 3177-3188. doi: 10.5433/1679-0359.2020v41n6Supl2p3177

Weinert, N. C. (2020). *Modelo hepático ex vivo em suínos: efeito do acetaminofeno, da fumonisina e do ácido fítico.* Tese de doutorado, Universidade Estadual de Londrina, Londrina, PR, Brasil. https://repositorio.uel.br/srv-c0003-s01/api/core/bitstreams/add803a3-ce06-47ee-9903-36d1240d767e/content

Xu, R., Kiarie, E. G., Yiannikouris, A., Sun, L., & Karrow, N. A. (2022). Nutritional impact of mycotoxins in food animal production and strategies for mitigation. *Journal of Animal Science and Biotechnology, 13*(1), 1-19. doi: 10.1186/s40104-022-00714-2

Zanella, J. B., Rommel, A. A., Turmina, A., Kailer, E. K., Rosin, M. R., Schlichmann, F., Franciscon, H., Cattelam, P. M. M., Silveira, M. F., & Cattelam, J. (2022). Fermentative profile of corn silages with the inclusion of chemical additives or bacterial inoculant. *Arquivo Brasileiro de Medicina Veterinária e Zootecnia, 74*(6), 1143-1150. doi: 10.1590/1678-4162-12760

Zdepski, B. F., Peres, J. A., Milla, B., Oliveira, S. N., Depaoli, C. R., Abreu, A. C. A., Garbossa, G., Manzano, R. P., & Bertagnon, H. G. (2023). Effect of Saccharomyces cerevisiae in a culture or lysate on innate immunity and ruminal and intestinal morphology of steers finished in feedlot. *Semina: Ciências Agrárias, 44*(4), 1321-1340. https://doi.org/10.5433/1679-0359.2023v44n4p1321