

Evaluation of the Role of Mycotoxin Binders in Reducing Aflatoxicosis in Ducks: An Experimental Study Using Contaminated Feed

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Abstract: Aflatoxins are among the most hazardous mycotoxins affecting poultry production, causing severe economic losses and adverse health effects, particularly in highly susceptible species such as ducks. This study was conducted to evaluate the protective efficacy of dietary mycotoxin binders against aflatoxicosis induced by aflatoxin B1-contaminated feed in Pekin ducks. A total of 250 one-day-old Pekin ducklings were randomly allocated into five equal experimental groups (50 birds/group). Group 1 served as the negative control and received a basal uncontaminated diet, while Group 2 received feed contaminated with aflatoxin B1 at a concentration of 75 ppb without binder supplementation. Groups 3, 4, and 5 received feed contaminated with aflatoxin B1 at a concentration of 75 ppb and supplemented with mycotoxin binder at doses of 0.5, 1.0, and 1.5 kg/ton feed, respectively. The experiment lasted for 45 days under controlled management conditions. Growth performance parameters, including live body weight, body weight gain, feed intake, and feed conversion ratio (FCR), were evaluated weekly. Hematological indices, including red blood cell count (RBC), hemoglobin concentration (Hb), packed cell volume (PCV), white blood cell count (WBC), lymphocyte percentage, and heterophil percentage, were assessed on days 15 and 25. Serum biochemical analyses included alanine aminotransferase (ALT), aspartate aminotransferase (AST), blood urea nitrogen (BUN), creatinine, and C-reactive protein (CRP). The results demonstrated that aflatoxin exposure significantly reduced live body weight, body weight gain, feed intake, RBC count, Hb concentration, and PCV%, while significantly increasing FCR, WBC count, heterophil percentage, liver enzymes, renal function markers, and CRP levels compared with the negative control group ($P \leq 0.05$). Dietary supplementation with mycotoxin binders significantly ameliorated these adverse effects in a dose-dependent manner. The highest binder dose (1.5 kg/ton) showed the greatest protective efficacy, restoring most productive, hematological, and biochemical parameters near normal values. Correlation analysis revealed strong negative relationships between aflatoxin toxicity markers and productive performance, while positive correlations were observed between hematological recovery and growth indices. In conclusion, the use of mycotoxin binders can be considered an effective nutritional strategy for minimizing aflatoxin-related losses in duck production systems.

Keywords: Aflatoxin B1, Mycotoxin Binder, Growth Performance, Ducks.

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INTRODUCTION

Aflatoxins are toxic secondary metabolites primarily produced by *Aspergillus flavus* and *Aspergillus parasiticus* under favorable environmental conditions such as high temperature and humidity (Valchev *et al.*, 2022). Aflatoxins contaminated cereal (wheat, corn sorghum) and oil crops (sunflower, soybean, peanut and cotton flours) in any time, during pre-harvest, storage, or processing of food ingredients (Shlej *et al.*, 2015). Worldwide surveys indicate that 25–60% of poultry feed samples contain detectable aflatoxin levels, with tropical

and subtropical regions facing the highest prevalence due to warm, humid conditions favoring fungal proliferation (Eskola *et al.*, 2020; Syraji *et al.*, 2025). Among the different aflatoxins, aflatoxin B1 (AFB1) is considered the most toxic and prevalent form contaminating feed ingredients, particularly cereals and oilseed meals used in poultry diets (Omotayo *et al.*, 2019; Nazhand *et al.*, 2020). AFB1 contamination represents a serious threat to animal health, poultry productivity, food safety, and public health due to the transfer of toxin residues into edible tissues and eggs, where AFB1 is associated with

their teratogenic, mutagenic, carcinogenic and immunosuppressive effects (Awuchi *et al.*, 2022; Wang *et al.*, 2023). The sensitivity of animals to aflatoxins is species and age dependent. Among domestic fowl, ducklings, goslings and turkey poults are reported to be the most sensitive to aflatoxin-induced toxicity (Banerjee *et al.*, 2022). Ducks occupy a distinctive position, with the global duck population reaching approximately 1.15 billion birds in 2020, of which 89% were concentrated in Asia. China overwhelmingly dominates duck meat production, accounting for over 70% of the global total, followed by Myanmar and France as distant secondary producers (Patil *et al.*, 2021). Duck production is highly important in many countries, including Iraq, as ducks contribute significantly to meat and egg production. However, ducks are considered one of the most sensitive avian species to aflatoxins because of their limited hepatic detoxification capacity and increased susceptibility to liver damage (Wang *et al.*, 2023). AFB1 is a strong hepatotoxic and nephrotoxic agent in poultry farming, aflatoxins cause enormous losses by impeding growth performance of birds, increasing feed conversion rates (Ramadan and Al-Ameri, 2022), reducing meat production (Fan *et al.*, 2013), and changing relative weights of visceral organs. They cause immunosuppression and increased susceptibility to infectious diseases (Wang *et al.*, 2023; Alnuimy, 2024), higher mortality rates due to liver and kidney damage (Li *et al.*, 2022). In Iraq, climatic conditions characterized by high temperature and improper feed storage favor fungal growth and mycotoxin production. Several Iraqi studies have confirmed the occurrence of aflatoxins and other mycotoxins in poultry feeds collected from different Iraqi provinces (Alnaemi *et al.*, 2023; Almremdhly *et al.*, 2024). A recent study conducted in Nineveh Province detected significant contamination of poultry feeds with aflatoxin B1, ochratoxin A, and fumonisin B1, with some samples exceeding the permissible limits established by international standards (Alnaemi *et al.*, 2023). Another Iraqi investigation demonstrated the widespread presence of molds and mycotoxins in poultry feeds associated with cases of suspected mycotoxicosis in poultry farms. These findings indicate that mycotoxin contamination remains an important challenge for the Iraqi poultry industry.

Due to the harmful effects of aflatoxins and the difficulty of complete prevention of feed contamination, considerable attention has been directed toward strategies aimed at reducing toxin bioavailability and minimizing their toxic effects. Among these strategies, mycotoxin binders have gained substantial interest as practical and economical feed additives. Mycotoxin binders are substances added to poultry diets to absorb toxins within the gastrointestinal tract and reduce their absorption into the bloodstream. Common binders include bentonite, zeolite, hydrated sodium calcium aluminosilicates (HSCAS), activated charcoal, yeast cell wall components, and various organic and inorganic adsorbents.

Numerous studies have demonstrated that mycotoxin binders can improve growth performance, enhance immune response, reduce liver lesions, and decrease mortality associated with aflatoxicosis in poultry. The efficacy of these binders depends on several factors including toxin concentration, binder type, inclusion rate, intestinal pH, and the species of poultry involved (Sharma *et al.*, 2026). Iraqi researchers have recently investigated different detoxification approaches and feed additives capable of reducing mycotoxin toxicity in poultry feeds, including bentonite clay minerals and activated charcoal (Muhammad *et al.*, 2026). These studies reported promising protective effects against aflatoxin-induced performance deterioration in broiler chickens.

Despite the growing body of knowledge concerning aflatoxicosis in poultry, studies focusing specifically on ducks in Iraq remain limited. Ducks possess unique physiological and metabolic characteristics that may influence their response to aflatoxin exposure and detoxification strategies. Therefore, it is essential to evaluate the protective efficacy of commercially available and novel mycotoxin binders in ducks under controlled experimental conditions. This study aims to address this knowledge gap by systematically investigating the capacity of a selected mycotoxin binder to alleviate the adverse effects of aflatoxin B1-contaminated feed on growth performance, hematological and biochemical parameters, in Pekin ducks under Iraqi conditions. Such studies may contribute to improving duck health, enhancing production efficiency, and reducing economic losses in the Iraqi poultry sector.

MATERIALS AND METHODS

Experimental Animals

The experiment was conducted over a 45-day period, from November 14 to December 29, 2025. A total of 250 one-day-old Pekin ducklings, with an average initial weight of 45 ± 5 g, were purchased from the Babil Modern Hatchery for Ducks in Babil Province. From this stock, 250 ducklings were randomly selected and equally allocated into five experimental groups (50 ducklings per group). Each group was housed in sterile, separate pens (100×500 cm²) under standard hygienic management conditions. The control group (G1) was reared in a separate room to prevent any cross-contamination or stress from the treated groups.

The Experimental Groups

All birds in all groups of experimental animals were fed non-contaminated control feed for 7 acclimatization days. After this period, experimental feed was fed to the birds in accordance with the experimental design until the end of the 45-day extension period, as described below. During the experiment, the mycotoxin binder was homogeneously incorporated into

the diet for a consistent daily intake and a consistent interaction with aflatoxin.

Group 1 (G1) – Control: Fed an uncontaminated basal diet without any mycotoxin binder supplementation.

Group 2 (G2) – Aflatoxin-Only: Fed the basal diet artificially contaminated with aflatoxin B1 at a concentration of 75 ppb, without any binder.

Group 3 (G3) – Low-Dose Binder: Fed the aflatoxin-contaminated diet (75 ppb) supplemented with the mycotoxin binder at a dose of 0.5 g/kg of feed.

Group 4 (G4) – Medium-Dose Binder: Fed the aflatoxin-contaminated diet (75 ppb) supplemented with the binder at a dose of 1.0 g/kg of feed.

Group 5 (G5) – High-Dose Binder: Fed the aflatoxin-contaminated diet (75 ppb) supplemented with the binder at a dose of 1.5 g/kg of feed.

Preparation of Aflatoxin-Contaminated Feed

Prior to the experiment, the process of preparing the aflatoxin-contaminated feed was conducted. Commercial broiler feed was procured from the Al-Ghadir Feed Company on November 1, 2025. To confirm the initial mycotoxin load, a representative sample of the feed was analyzed using quantitative mycotoxin assay techniques, with a specific focus on aflatoxin B1. The analysis revealed a baseline aflatoxin contamination level of 3 ppb. To promote fungal growth and amplify aflatoxin production to the target experimental concentration of 75 ppb, the feed was subsequently treated. The feed was evenly moistened using a fine water spray to achieve a uniform moisture content conducive to fungal proliferation. The moistened feed was then stored in sealed containers under controlled conditions of high humidity and ambient temperature (approximately 25-30°C) for an incubation period of 14 days. Following the incubation period, on November 14, 2025, the feed was thoroughly homogenized. Subsequent quantitative analysis confirmed that the aflatoxin B1 concentration had reached the intended level of 75 parts per billion (ppb). This prepared, artificially contaminated feed batch was then used to formulate experimental diets for groups G2 through G5, as outlined in Section المصدر.

Growth Performance Parameters

Growth performance parameters (body weight, feed intake, weight gain and feed conversion ratio) were evaluated weekly by measuring the following parameters according to standard methodologies for meat-type ducks (Shu *et al.*, 2025).

Blood Sample Collection

Blood samples were collected at several time points, specifically on days 15 and 25 of the chickens' age. Ten chicks were randomly selected from each group for blood sampling (2 mL), which was aspirated from the jugular vein using disposable 3 mL syringes. This blood sample was divided into two parts: the first part was placed in a tube containing EDTA anticoagulant to perform hematological tests, while the other part was

placed in a gel tube (without anticoagulant) to separate the serum used for biochemical tests (AST/GOT), (ALT/GPT), CRP, serum urea, and serum creatinine by using apparatus SMT 100V / Seamaly 20 photometer using the original Seamaly Diagnostics kits.

Hematological Parameters

At each blood collection time point (days 15 and 25), the whole blood samples collected in EDTA tubes were used for the determination of hematological parameters. A complete blood count (CBC) was performed using a veterinary hematology analyzer within a few hours of collection. The parameters measured included red blood cell count (RBCs), hemoglobin concentration (Hb), packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), white blood cell count (WBCs), and the differential leukocyte count (Abang *et al.*, 2023).

Serum Biochemical Parameters

Serum samples collected at to evaluate liver and kidney function. The following parameters were measured spectrophotometrically according to the manufacturers' instructions for each specific kit (apparatus SMT 100V / Seamaly 20 photometer using the original Seamaly Diagnostics kits following standard protocols for duck studies (Cao *et al.*, 2024).

C-Reactive Protein (CRP) Determination

Serum CRP levels were measured using a latex agglutination slide test (CRP Latex Test Kit; Bio Research). The assay is based on the immunological reaction between CRP in the Duck serum and specific anti-duck CRP antibodies coated onto polystyrene latex particles (Zheng *et al.*, 2021).

Statistical Analysis: Statistical analysis of data was done by one-way ANOVA followed by Tukey-Kramer test ($P \leq 0.05$) (SAS, 2010).

RESULTS

Growth Performance Parameters

The results of live body weight demonstrated in table (1) showed no significant differences among all experimental groups at day 1 and day 7, indicating homogeneity of the experimental ducklings before aflatoxin exposure. However, after the administration of aflatoxin-contaminated feed, marked reductions in body weight were observed in the positive control group (AF group) compared with the negative control group throughout the experimental period. At day 42, the negative control group recorded the highest live body weight (1650.1 ± 9.06 g), whereas the aflatoxin-treated group showed a severe reduction reaching 825.9 ± 4.02 g. Supplementation with mycotoxin binders significantly improved body weight in treated groups in a dose-dependent manner. The group treated with 1.5 kg/ton

binder exhibited the greatest improvement (1203.5 ± 9.52 g), followed by the 0.5 kg/ton and 1 kg/ton groups. Statistical analysis revealed significant differences ($P \leq 0.05$) among the treated groups and the positive control group, particularly during the late growth stages (days 28–42). Similarly, weekly body weight gain in table (2) was significantly depressed in the aflatoxin-only group compared with the control negative group. The most pronounced decline was observed during days 35–42, where the AF group recorded only 180.8 g compared with 449.7 g in the negative control group. Birds supplemented with binders showed progressive recovery in weight gain, especially at the higher inclusion level (1.5 kg/ton), which achieved 413.5 g during the final week.

Feed intake data in table (3) revealed that aflatoxin exposure reduced feed consumption during

most experimental periods. Ducks in the AF group exhibited significantly lower feed intake compared with the negative control, particularly during days 14 and 42. In contrast, supplementation with binders improved feed intake values, with the 1.5 kg/ton group recording feed intake values approaching those of the negative control group.

Feed conversion ratio (FCR) in table (4) was markedly impaired in ducks fed aflatoxin-contaminated diets. The AF group demonstrated the poorest FCR values throughout the experiment, reaching 5.56 at day 42 compared with 2.43 in the negative control group. The best FCR among treated groups was observed in the 1.5 kg/ton binder group (2.66), which was close to the normal control value, indicating enhanced feed utilization and metabolic performance.

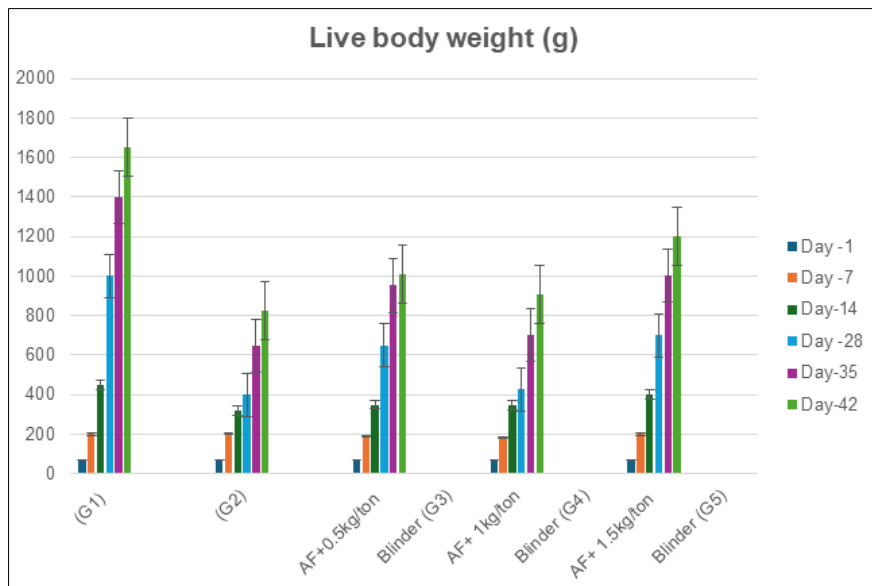


Figure 1: Live body weight (g)

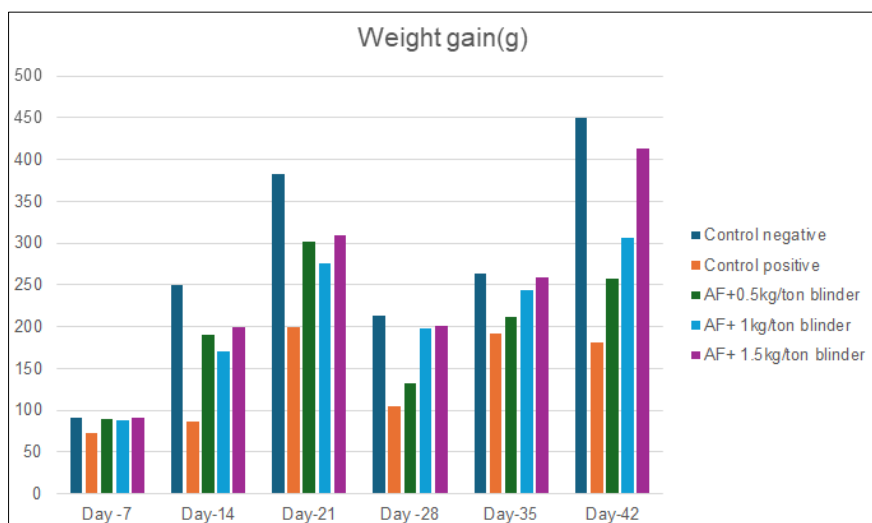


Figure 2

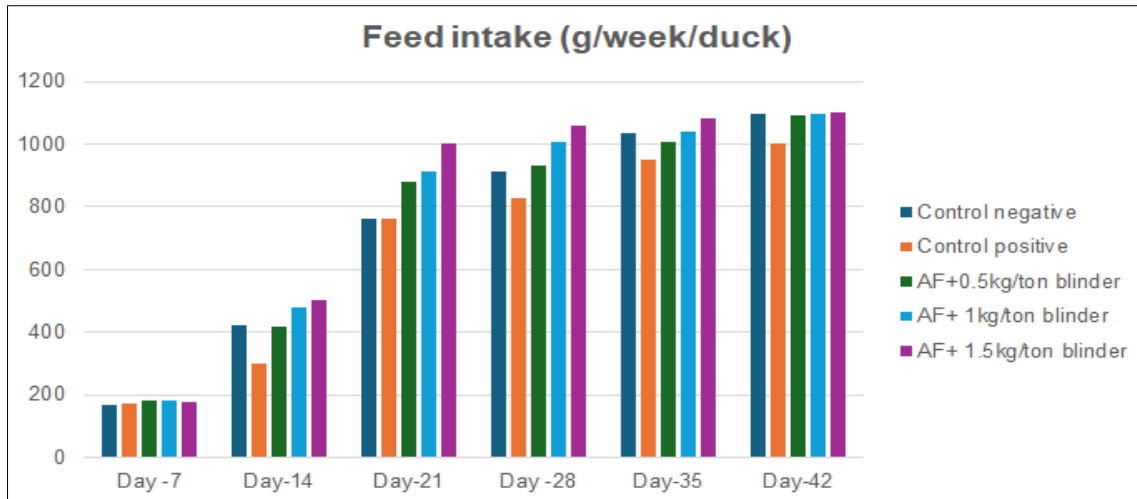


Figure 3

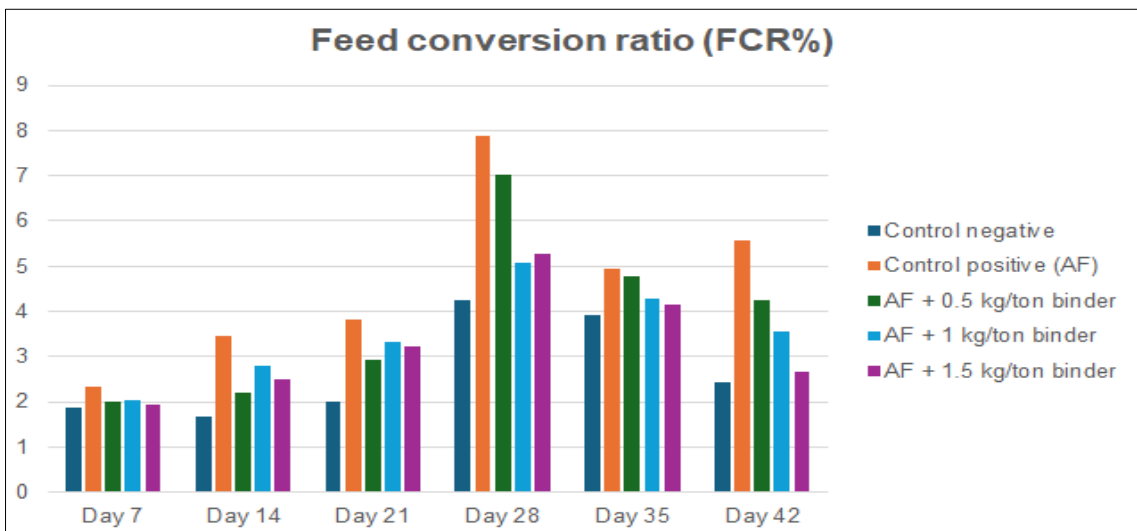


Figure 4

Hematological Analysis

The hematological findings in table (5) demonstrated that aflatoxin exposure significantly reduced erythrocytic indices. The AF group showed lower RBC count, hemoglobin concentration, and packed cell volume (PCV%) at both day 15 and day 25 compared with the negative control group. At day 25, RBC count declined to $2.33 \pm 0.01 \times 10^6/\mu\text{L}$ in the AF group compared with $3.30 \pm 0.03 \times 10^6/\mu\text{L}$ in the negative control group. Likewise, hemoglobin concentration and PCV values were markedly reduced, indicating anemia associated with aflatoxicosis.

Administration of mycotoxin binders ameliorated these hematological alterations. The highest binder dose (1.5 kg/ton) restored RBC, Hb, and PCV values near normal levels, suggesting improvement in erythropoietic activity and reduction of oxidative damage induced by aflatoxin. Statistical analysis

indicated significant differences ($P \leq 0.05$) between the AF group and binder-treated groups.

Leukocytic parameters illustrated in table (6) revealed an opposite trend. The AF group exhibited significant leukocytosis characterized by elevated WBC count and heterophil percentage, accompanied by decreased lymphocyte percentage. At day 25, WBC count reached $27.5 \pm 6.82 \times 10^3/\mu\text{L}$ in the AF group compared with $19.4 \pm 6.30 \times 10^3/\mu\text{L}$ in the negative control group. Heterophils increased markedly to $52.8 \pm 7.18\%$, whereas lymphocytes decreased to $42.1 \pm 6.33\%$. These findings indicate inflammatory and stress responses associated with aflatoxin toxicity.

Binder supplementation significantly improved leukocyte profiles, particularly at higher doses. The 1.5 kg/ton group demonstrated near-normal leukocyte values with reduced heterophilia and restored lymphocyte percentages, reflecting improved immune status and reduced systemic inflammation.

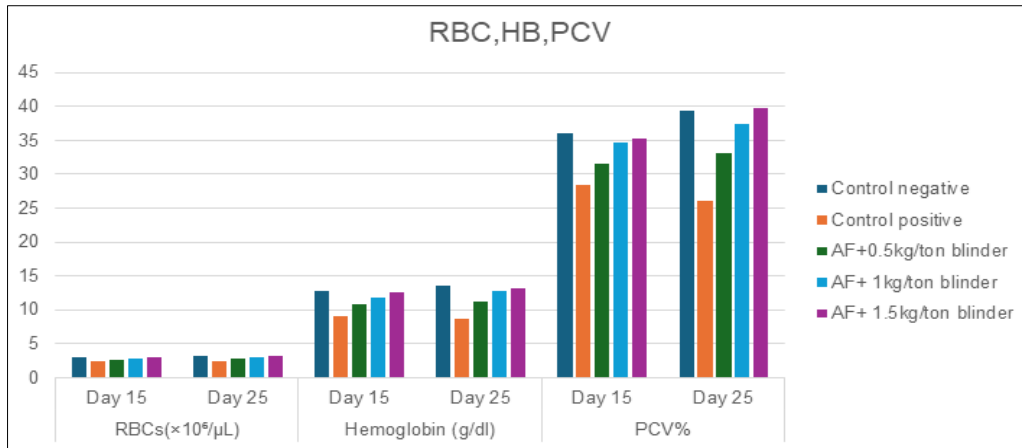


Figure 5

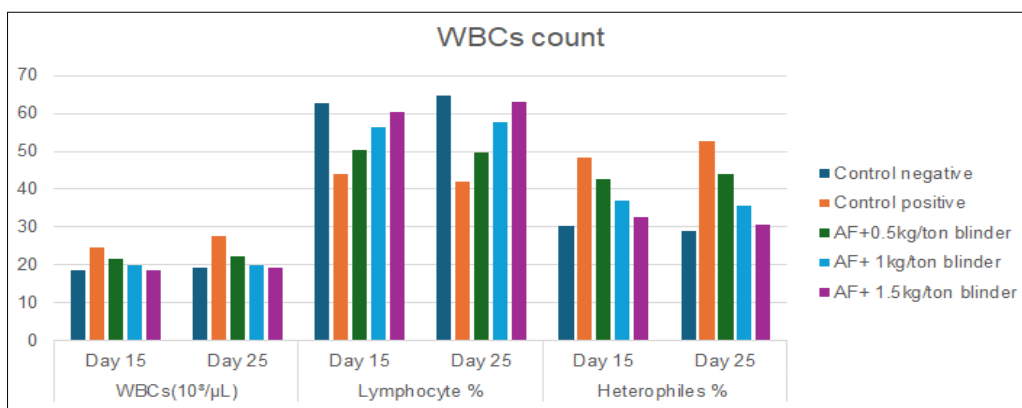


Figure 6

Biochemical Analysis

Biochemical analysis in table (7) revealed severe hepatic and renal impairment in ducks exposed to aflatoxin. Serum ALT and AST activities were significantly elevated in the AF group, reaching 95.6 ± 3.8 U/L and 544 ± 15.4 U/L, respectively, compared with 25.9 ± 1.2 U/L and 183 ± 6.1 U/L in the negative control group. Similarly, blood urea nitrogen (BUN), serum creatinine, and CRP concentrations were markedly

increased in the AF group, indicating kidney dysfunction and systemic inflammatory responses.

Supplementation with mycotoxin binders significantly reduced all biochemical abnormalities in a dose-dependent manner. Ducks receiving 1.5 kg/ton binder showed ALT, AST, BUN, creatinine, and CRP values approaching those of the control group. These results suggest substantial hepatoprotective, nephroprotective, and anti-inflammatory effects of the binder against aflatoxin toxicity.

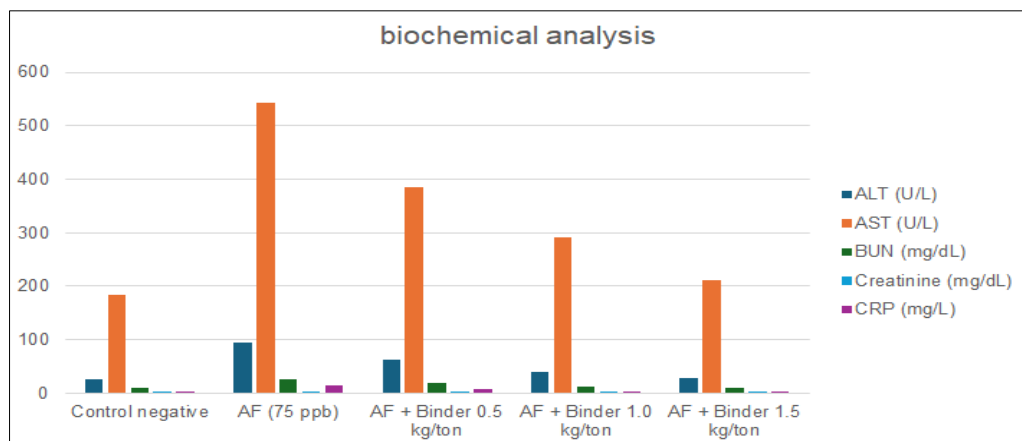


Figure 7

DISCUSSION

The present study demonstrated that dietary aflatoxin B1 exposure caused severe detrimental effects on growth performance, hematological profile, liver and kidney function, and inflammatory responses in Pekin ducks. Supplementation with mycotoxin binders significantly alleviated these adverse effects in a dose-dependent manner. The marked reduction in body weight and body weight gain observed in the aflatoxin-treated group may be attributed to impaired nutrient digestion, reduced feed efficiency, and hepatocellular dysfunction caused by aflatoxin toxicity (Yunus *et al.*, 2011; Almremdhly *et al.*, 2024). Aflatoxins interfere with protein synthesis, lipid metabolism, and enzymatic activity, ultimately leading to growth suppression. These findings are consistent with the results reported by Pasha *et al.*, (2007) who demonstrated that aflatoxin-contaminated diets significantly reduced growth performance and feed efficiency in poultry. Similar observations were also reported by Fan *et al.*, (2013) who linked aflatoxin exposure to reduced meat production and impaired nutrient utilization. The improvement in productive performance following binder supplementation suggests efficient adsorption of aflatoxin within the gastrointestinal tract, thereby reducing toxin bioavailability and tissue damage. The superior efficacy observed in the high-dose binder group may be related to increased binding capacity and enhanced prevention of toxin absorption. These findings agree with those of Kumari *et al.*, (2025) who reported that the efficacy of binders depends on dosage, physicochemical characteristics, and species-specific digestive physiology. The hematological alterations observed in the aflatoxin-treated ducks, particularly reductions in RBC count, hemoglobin concentration, and PCV%, may result from impaired hematopoiesis, oxidative stress, and suppression of erythrocyte production. Aflatoxins are known to induce lipid peroxidation and damage bone marrow tissues, leading to anemia. These findings are consistent with reports by Abang *et al.*, (2023) who demonstrated significant hematological depression in birds exposed to mycotoxins. Leukocytosis and heterophilia observed in the AF group likely reflect inflammatory stress and immune dysregulation induced by aflatoxin exposure. Increased heterophil counts are considered indicators of physiological stress and inflammatory reactions in birds. The restoration of leukocyte profiles following binder administration suggests improved immune competence and reduced systemic inflammation. Similar results were described by Indresh *et al.*, (2013) who reported that aflatoxins induce immunosuppression and inflammatory responses in poultry species. The substantial elevation of ALT and AST activities in the aflatoxin group confirms severe hepatocellular injury. Aflatoxin B1 primarily targets the liver because it undergoes bioactivation in hepatic microsomes, generating reactive metabolites capable of inducing oxidative damage and necrosis. Elevated AST and ALT levels have been widely used as

biomarkers of liver injury in aflatoxicosis studies. These findings are in agreement with Pappas *et al.*, (2014) who reported significant hepatic enzyme elevations in birds exposed to aflatoxins. The increased levels of BUN and creatinine observed in this study indicate renal dysfunction associated with aflatoxin toxicity. Kidney damage may result from oxidative stress, tubular degeneration, and impaired renal filtration. Similar findings were reported by Cao *et al.*, (2024) who observed nephrotoxic effects following prolonged mycotoxin exposure in ducks. Furthermore, the marked elevation of CRP concentrations in aflatoxin-treated ducks reflects acute inflammatory responses triggered by tissue injury and oxidative stress. CRP is considered an important biomarker of systemic inflammation and hepatic stress. The reduction of CRP levels following binder supplementation supports the anti-inflammatory role of mycotoxin binders through toxin sequestration and reduction of tissue damage (Hassan *et al.*, 2012). The correlation analysis further confirmed the interrelationship between productive performance and physiological disturbances induced by aflatoxin exposure (Saleemi *et al.*, 2015). Negative correlations between liver enzymes and body weight gain suggest that increasing hepatic injury directly impairs growth and metabolic efficiency. Conversely, positive correlations between hematological parameters and productive indices indicate that improved oxygen transport and immune competence contribute to enhanced growth performance.

CONCLUSION

In conclusion, dietary aflatoxin B1 exerts severe toxic effects on productive performance, hematological profile, liver function, kidney function, and inflammatory status in Pekin ducks. Supplementation with mycotoxin binders, particularly at 1.5 kg/ton feed, effectively reduced the harmful effects of aflatoxicosis and improved overall health and productivity. Therefore, the use of mycotoxin binders can be considered an effective nutritional strategy for minimizing aflatoxin-related losses in duck production systems.

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