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The Efficiency of Aloe vera in inhibiting the growth of Aspergillus flavus fungus and its protective effects on growth performance and serum biochemistry of broiler chickens fed by Aflatoxin B1

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Abstract: The study aimed to assess how well different Aloe vera gel extract (AVGE) concentrations inhibited the development of Aspergillus flavus in a potato dextrose agar (PDA) culture medium. According to the data, Aspergillus flavus growth in PDA culture medium was variably inhibited by Aloe vera gel extract at concentrations of 1, 2, 3, and 4%; these rates were 97, 100, 5.88, and 17.64%, respectively. The in vitro best concentration of AVGE was (2%). An in vivo study was conducted to investigate the protective effects of Aloe vera powder (AVP) on performance and serum biochemical parameters in broiler chickens fed on diets contaminated with aflatoxin B1 (AFB1). A total 450 broiler chicks (Ross-308) were randomly allocated to five treatments. 1) basal diet (control group), 2) contaminated diet with 1 mg AFB1/ kg diet (AFB1-diet), 3) AFB1-diet plus AVP 1g / kg diet, 4) AFB1-diet plus A 2 g / kg diet and 5) AFB1-diet plus AVP 3 g / kg diet. Each treatment consisted of six replicates of 15 birds. Growth performance was lower for groups fed 1 mg AFB1/ kg diet. At the same time, growth parameters were improved by incorporating 1 and 2 g/kg AVP in the AFB1 contaminated feed. Chicks fed AFB1 had significantly higher aspartate aminotransferase (AST), alanine aminotransferase (ALT), uric acid, and creatinine than other groups. In addition, Aloe vera powder improved biomarkers of the liver and kidney functions and reduced AFB1 residue in broilers' liver and muscle tissue. The results demonstrated that AVP at 2 g / kg diet could reduce the adverse effects of AFB1 exposure in broiler chicks.

Keywords: aflatoxin B1, Aloe vera, blood, chicks, performance

1. Introduction

Aflatoxins are secondary metabolites of mycotoxins generated from *Aspergillus flavus* and *Aspergillus parasiticus*. It is well-recognized to be highly poisonous and carcinogenic. Aflatoxin B1 (AFB1) is the most dangerous of the several toxic aflatoxins (B1, B2, G1, and G2, M1, M2). Additionally, it is regarded as a Group I carcinogen. Since AFB1 administration has adverse effects on people and many species of cattle, including hepatotoxic, teratogenic, mutagenic, and carcinogenic consequences, intake of contaminated food or feed by humans and animals is associated with significant economic losses (Zhang et al., 2016; Attia et al., 2013a & 2016). The reduction in body weight increase in broilers, which is likely brought on by changes in protein metabolism, is the most significant economic impact of aflatoxicosis (Shareef and Sito 2019). On the other hand, Aflatoxin negatively impacts liver tissue (Saleemi et al., 2020) and negatively impacts relative organ weight (Jahanian et al., 2016). Aflatoxin also affects biochemical and haematological parameters (Attia, 2023; Rashidi et al., 2020; Jahanian et al., 2019). Aflatoxin has been linked to harmful impacts on growth performance, carcass and meat qualities, and intestinal microbiota (Zaker-Esteghamati et al. (2020).

A. flavus is one of the most significant fungus-producing mycotoxins, particularly AFB1 (Hussein, 2008). It is also one of the most important contaminants of food and feed. The majority of these toxins are persistent and cannot be eliminated by heat, food processing, or conventional cooking

methods. Mycotoxins are secondary metabolites produced by some fungi naturally, and this fungi can grow on many food crops before and during harvest and storage. Most mycotoxins also have low molecular weights, ranging from 97 to 710 Dalton, making them resistant to harsh environmental conditions and preventing them from activating the immune system (Ismail, 2014).

Animals are exposed daily to toxins at low concentrations and thus experience prolonged shivering (Pitt, 2000; Attia et al., 2018). Several techniques are utilized to lower the mycotoxin content of chicken diets. A contaminated feed can be detoxified via physical, chemical, or biological methods. Cereals can be dried before being stored, which lowers the feed's high humidity content and effectively lowers the mild mycotoxin contamination (Bryden, 2012). However, chemical detoxification techniques are frequently expensive and ineffective. Additionally, in some nations, specific chemical techniques are prohibited (Park and Price, 2001).

Utilizing mineral adsorbents such as bentonites, zeolites, and aluminosilicates in the feed is the most popular method for defending animals against mycotoxicosis. The removal or reduction of mycotoxins in food and feed has been found to be possible by various microorganisms, including bacteria, yeast, and molds (Azam et al., 2021).

In addition, recent studies have looked into the natural inhibitions of various natural substances, such as medicinal plants, or fungi (Loi et al., 2020). Asphodelaceae (Liliaceae) is the family to which *Aloe vera* belongs. It has a light green color and is a succulent. The plant also has thick leaves with serrated edges that have thorns. The leaves consist of three layers, the inner is a pure gel composed of 99% water, 1% glucomannans, amino acids, glycosides, fats, vitamins, sterols, and the middle layer that contains latex is a yellow juice made up of anthraquinones and glycosides, whereas the third layer is the thick outer layer called the cortex and has two functions: protection and formation of proteins and carbohydrates, as well as the transport vessels inside it (Surjushe et al., 2008). It's interesting to note that *Aloe vera* is a short-stemmed succulent medicinal plant and that the amount of active ingredients in the gel varies depending on the growth conditions of the plant (Shireen et al., 2015). These ingredients include aloin, aluminan, elmodin, sterols, saponins, and more. In light of the aforementioned information, this study aimed to assess the effectiveness of *Aloe vera* supplements as dietary defenses agent against the toxicity of AFB1 in broiler chickens.

2. Materials and Methods

Preparation of *Aloe vera* gel extract

The fresh *Aloe vera* leaves were washed under running water for 5 minutes and rinsed with sterile deionized water, and the side spines and green outer cortex layer were removed with a clean knife. After that, the colorless gel was homogenized thoroughly in a blender, filtered through a Whatman No. 1 filter paper, and centrifuged at 500 rpm for 5 minutes. The filtrate was then used as an extract (Rasouli et al., 2019).

Preparation of *Aloe vera* powder

After being initially dried for three to four days, green *Aloe vera* leaves were then dried in an oven at 60 °C until the moisture content was below 10%. The leaves were then hand crushed to a fine consistency powder, it was put through a fine-mesh wire sieve.

Evaluation of different *Aloe vera* gel extract levels in inhibiting *Aspergillus flavus* grow on a potato dextrose agar culture medium

Tetracycline was added to potato dextrose agar (PDA) at a 250 mg/liter dosage to inhibit bacterial growth after PDA had been prepared and cooled to 45°C. The medium

was then poured into five 250 ml flasks, each holding 100 ml of the medium. *Aloe vera* gel extract was added to each flask in concentrations of 1, 2, 3, and 4% (ml/100 ml), with one flask remaining without gel extract as a control. All flasks were thoroughly mixed before being put into Petri plates with a 90 mm diameter and allowed to solidify.

Then, the Petri dishes were inoculated with a suspension of *Aspergillus flavus* spores isolate. The spore's suspension of the fungus isolate was prepared by adding 10 ml of sterile deionized water to a 10 mm diameter disc of 7 days old *A. flavus* colony. A drop of the fungus spore suspension was taken using the isolation needle, spread into the center of a Petri dish, and incubated at 25±2°C. The diameters of the developing colonies were measured every two days until the control dishes were full of fungal colonies, and the inhibition rate was calculated according to the following equation (Al-Hamiri and Hussein 2022):

% Inhibition = [(Average diameter of colony in control – average diameter of colony in the treatment) / Average diameter of colony in control] X 100

Evaluation of the effect of *Aloe vera* powder on broiler chick's performance:

Experimental design

A total of 450 one-day-old Ross 308 broiler chicks were obtained from a hatchery and raised for 35 days on a private farm in **Kafr El-Sheikh** city in 30 pens with 5 treatments and 6 replications. The Ross 308 strain catalogue (2018) served as a guide for the formulation of experimental diets. The chicks were kept in separate experimental rooms that were clean and well-ventilated. A floor-pen (0.1 m^2 per bird) was employed to house the birds. A sufficient number of feeders and water utensils were available in each room. Wood shavings served as the bedding for the birds. Ventilation was supplied by negative pressure with fans. Gas-fired brooders were used to generate heat. The experimental rooms' ambient temperature was kept at 32°C for the first week, and then dropped 3°C per week during the following weeks before being set at 22°C. Standard hygiene precautions against infectious diseases were used. The Newcastle disease vaccination program was carried out using the Hitchner B1 strain (Biove Egypt) at seven days and the LaSota vaccine (Intervet Company) at eighteen days in drinking water.

Additionally, birds were immunized against infectious bursal disease utilizing (Gumboro, 228, and Intervet Company) via drinking water at two weeks of age. The basal diet was tested for the presence of significant mycotoxins, including AFB1 and ochratoxin A, with ELISA. No mycotoxins were recorded. No drug administration was done in the experimental period (5 weeks). The chicks were randomly assigned to five treatment groups: 1) basal diet (control), 2) contaminated diet with 1 mg AFB1/ kg diet (AFB1-diet), 3) AFB1-diet plus AVP 1g / kg diet, 4) AFB1-diet plus A 2 g / kg diet and 5) AFB1-diet plus AVP 3 g / kg diet. In groups 2, 3, 4, and 5, pure crystalline AFB1 (Sigma-Aldrich) was added to the diets. AFB1 was first dissolved in chloroform (1 mg/10 mL), and then the solution was combined with a suitable amount of feed. The produced premix feed was left overnight at room temperature for solvent evaporation before being combined with the basal diet to provide the desired level of AFB1/kg of diet (1 mg of AFB1/kg of feed) (Denli et al., 2009).

Experimental procedure and sampling

Broilers and feed intake were weighed weekly from day 1 to 35 of the experiment. Feed conversion ratio was calculated as feed intake per unit body weight.

Table (1): Composition and calculated analysis of starter and finisher diets.

Ingredients	Starter-grower (1-21d)	Finisher (22-35d)
Yellow corn	54.40	62.00
Soybean meal, 44%	27.00	24.05
Corn Gluten meal, 60%	10.00	6.19
Soy bean oil	4.55	4.00
Limestone	1.10	1.00
Di-calcium phosphate	2.20	2.05
Vit& min. premix*	0.30	0.30
DL-Methionine	0.05	0.01
L-lysine (HCl)	0.15	0.15
Na Cl	0.25	0.25
Total	100	100
Calculated analysis: **		
CP, %	23.03	20.02
ME (Kcal/kg)	3204	3201
Calcium, %	1.05	0.97
Available phosphorus, %	0.45	0.42
Lysine, %	1.14	1.03
Methionine, %	0.52	0.41
TSAA, %	0.90	0.73

*Each 3kg contain Vit A 12000000IU, Vit D3 2000 000 IU, Vit E 10g, Vit K3 2g, Vit B1 1g, Vit B2 5g, Vit B6 1.5g, Vit B12 10mg, Nicotinic acid 30g, Pantothenic acid 10g, Folic acid 1g, Biotin 50mg, Choline chloride 250g, Iron 30g, Copper 10g, Zinc 50g, Manganese 60g, Iodine 1g, Selenium 0.1g, Cobalt 0.1g and carrier (CaCo3) to 3kg.

**Calculated values were according to NRC (1994) text book values for feedstuffs.

Serum biochemical parameters.

At the end of the experiment, blood samples (6 samples / treatment) were clotted at room temperature. Serum separation was performed by centrifuging the coagulated blood at 3000 rpm for 15 minutes. Serum was collected and stored at -20°C for determination of urea, creatinine, albumin, total protein, uric acid, and activity of AST, ALT, and ALP using commercial kits, spectrophotometrically.

Method for detection of aflatoxin residue in the liver and muscle tissue

After 35 days after the start of the experiment, the liver and muscle samples (6 samples / group) were collected. Until they were used, the samples were kept in a deep freezer. Thin-layer chromatography (TLC) was used to determine the AFB1 residue in the liver and muscle samples, according to Schuller and Van Egmond (1981).

Statistical analysis:

One-way variance analysis has been applied to the outcomes of all response variables (SAS, 2003) using the followingmodel:

$Yij = \mu + tj + eij$,

Where, Yij = any observation, μ = the general mean, tj = the effect of treatment,

eij = the experimental error.

The mean of the variables was compared using Duncan's Multiple Range Test (Duncan, 1955).

3. Results.

The efficiency of different concentrations of *Aloe vera* gel extract in inhibiting *Aspergillus flavus* growth on potato dextrose agar culture medium

The experiment's findings revealed that the growth of *A. flavus* was inhibited by 95.62, 100.00, 95.00, and 93.75%, respectively when *Aloe vera* gel extract was used in concentrations of 1, 2, 3, and 4% in the PDA culture medium. The most effective ratio for inhibiting the fungus totally was 2ml of extract/100ml PDA. It was apparent that the extract's ability to suppress the fungus was

less effective at greater concentrations of the extract.

Table 2. The effect of different Aloe vera gel extract concentrations on the inhibition rate of A. flavus grow on potato dextrose agar culture medium.

Concentrations of *Aloe vera* gel extract	Average colony diameters (mm)	Inhibition %
0 % *Aloe vera* (control)	8.00[a]	0.00[d]
1 % *Aloe vera*	0.35[c]	95.6[b]
2 % *Aloe vera*	0.00[d]	100[a]
3 % *Aloe vera*	0.40[bc]	95.0[b]
4 % *Aloe vera*	0.50[b]	93.7[c]
SEM	0.002	3.29
p-value	0.0001	0.0001

[abcd] Means within the same column with different letters are significantly other ($p < 0.05$). SEM: Standard error of the mean.

Growth parameters.

Table 3 displays how *Aloe vera* affected the chickens' growth parameters. Birds fed a meal containing AFB1 (AFB1-diet) experienced poorer growth metrics and feed intake than the control group ($p < 0.05$). The AFB1-diet group had the lowest weight, as seen in Table 3, while the control group had the greatest weight. In Table 3, the mortality percentage was greater in the AF-fed birds than in the AFB1-diet group ($p < 0.05$). In contrast, feeding on fodder contaminated with AFB1 and treated with *Aloe vera* powder at levels 1, 2, and 3g/kg diet resulted in a significant improvement in bird weight, feed intake, and FCR while lowering the mortality rate in comparison to the birds fed on fodders without any contamination (control group) (Table 3).

Table 3. Effect of dietary Aloe vera on the performance parameters for broiler chicks fed on contaminated diet with 1 mg AFB1 /kg diet during 1 to 35 day.

Items	Treatments					SEM	p-value
	Control group	AFB1-diet group	AFB1-diet + 1 g AVP	AFB1-diet +2 g AVP	AFB1-diet +3 g AVP		
IBW	40.8	41.0	40.6	40.8	41.0	1.16	0.585
FBW	2350[a]	1680[d]	2050[c]	2180[b]	2000[c]	3.25	0.0001
BWG	2309[a]	1639[d]	2009[bc]	2139[b]	1959[c]	2.36	0.004
TFI	3480[a]	2860[d]	3100[c]	3360[b]	3080[c]	2.52	0.002
FCR	1.51[c]	1.75[a]	1.54[c]	1.57[c]	1.57[b]	0.015	0.032
Mortality %	0.00[d]	12.2[a]	2.22[c]	0.00[d]	3.33[b]	0.08	0.006

[abcd]Means within the same row with different letters are significantly different ($p < 0.05$); SEM: Standard error of the mean; AVP: *Aloe vera* powder; IBW: initial body weight; FBW: Final body weight; BWG: body weight gain; TFI: total feed intake; FCR: feed conversion ratio.

Biochemical parameters

Table 4 illustrates how the inclusion of *Aloe vera* supplements to the aflatoxin-contaminated diet counteracted the harmful effects of aflatoxin B1 on some biochemical markers in broilers fed on for 35 days. Compared to the other groups, the AFB1-diet group's ALT, AST, and ALP activities increased significantly ($P \leq 0.05$). In contrast, there was a substantial ($P \leq 0.05$) decline in the activity of the liver enzymes in the T3, T4, and T5 groups as compared to the T1 group (control). In addition, when compared to the control group, there were substantial ($P \leq 0.05$) drops in all of the metrics for total protein, albumin, and globulin in the AFB1-diet group. On the other hand, compared to the AFB1-diet group, there were substantial ($P \leq 0.05$) increases in the total protein, globulin, and albumin in the T3, T4, and T5 groups. Regarding the renal function outcomes, the AFB1-diet group had

considerably ($P \leq 0.05$) higher urea and uric acid concentrations than the other groups.

Table 4. Effect of dietary Aloe vera on some blood constituents for broiler chicks fed on contaminated diet with 1 mg **AFB1 /kg diet during 1 to 35 day.**

Items	Treatments					SEM	p-value
	Control group	AFB1-diet group	AFB1-diet + 1 g AVP	AFB1-diet +2 g AVP	AFB1-diet +3 g AVP		
TP (g/dl)	6.15[a]	4.25[c]	5.17[b]	6.00[a]	5.00[b]	0.15	0.005
Alb. (g/dl)	3.36[a]	2.76[c]	3.10[b]	3.25[a]	3.00[b]	0.05	0.0001
Glo. (g/dl)	2.79[a]	1.49[c]	2.07[b]	2.75[a]	2.00[b]	0.08	0.002
AST (U/L)	21.0[d]	95.8[a]	40.0[b]	30.6[c]	46.0[b]	2.18	0.006
ALT (U/L)	24.0[c]	50.5[a]	38.0[b]	26.0[c]	30.8[b]	2.08	0.001
ALP (U/L)	902[c]	1205[a]	930b[c]	908[c]	946[b]	3.25	0.0001
Creatinine (mg/dl)	0.460[d]	1.058[a]	0.585[c]	0.492[d]	0.623[b]	0.004	0.045
Uric acid (mg/dl)	5.60c	7.68a	6.08b	5.80c	6.25[b]	0.55	0.038

[abcd] Means within the same row with different letters are significantly different ($p < 0.05$); SEM: Standard error of the mean; AVP: *Aloe vera* powder; TP: total protein; Alb.: Albumin; Glo: Globulin; AST = aspartate aminotransferase, and ALT = alanine aminotransferase. ALP= Alkaline Phosphatase.

Aflatoxin residue within the liver and muscle tissue of broilers

Table 5 shows the levels of AFB1 residues in each group's liver and muscle tissue at the end of the experiment. When compared to the other groups, the aflatoxin group (AFB1-diet) had significantly more AFB1 residue in both the liver and the muscle tissue at the end of the trial ($P < 0.05$). With the addition of *Aloe vera* specific to the aflatoxin group, this residue was significantly reduced ($P < 0.05$). The findings showed that, compared to the AFB1-diet group on day 35, the group treated with AF-*Aloe vera* 2 g/ kg diet had the lowest level of AFB1 (1.08 µg/kg). In contrast, the AFB1-diet group had the greatest level (5.26 µg/kg). On day 35, there were significant variations in the doses of 1, 2, and 3g ($p<0.05$). On day 35, those that received *Aloe vera* treatment along with meat showed no signs of AFB1 residue.

Table 5. Effect of dietary Aloe vera on AFB1 residues values for broiler chicks fed on a diet containing 1 mg aflatoxin B1 / kg diet at 1 to 35 days

Items	Treatments					SEM	p-value
	Control	AFB1-diet	AFB1-diet + 1 g AVP	AFB1-diet + 2 g AVP	AFB1-diet + 3 g AVP		
AFB1 residue (µg/kg) in the liver tissue at the end of the experiment	0.00[e]	5.26[a]	2.17[c]	1.08[d]	2.84[b]	0.04	0.0001
AFB1 residue (µg/kg) in the muscle tissue at the end of the experiment	0.00[b]	0.86[a]	0.00[b]	0.00[b]	0.00[b]	0.02	0.0001

[abcd] Means within the same row with different letters are significantly different ($p < 0.05$); SEM: Standard error of the mean; AVP: *Aloe vera* powder.

4. Discussion

According to several studies, *Aloe vera* has antifungal properties in in-vitro situations (Jasso de Rodrguez et al., 2005; Babaei et al., 2013). Also demonstrated was the ability of *Aloe vera* powder to function as a biosorbent and serve as a substitute for the removal of AFB1. In their Adsorption Experiment, Zavala-Franco et al. (2018) discovered that the control group, which was not supplemented with adsorbents, had an aflatoxin value of 27 ng/mL, indicating a lack

of AFB1 adsorption. In contrast, aloe powder demonstrated the highest biosorbent efficacy for aflatoxin removal; the biosorption uptake was 68.52%, statistically similar to those seen in zeolite (70.19%).

Al-Hamiri and Hussein (2022) found that 2% aloe vera gel extract inhibited *A. flavus* growth and prevented 86.5% of AFB1 synthesis in grains. The concentration of AFB1 dropped from 21.2 ppb in the control group to 2.86 ppb in the gel extract treatment group. Additionally, 2% *Aloe vera* extract decreased the concentration of AFB1 in rice grains that had been artificially inoculated with AFB1by 74.34%. AFB1 was lowered from a concentration of 22.88 ppb in the control treatment to 5.87 ppb in the grains treated with the extract. The study's findings demonstrated the high effectiveness of *Aloe vera* gel extract in preventing the growth of the *A. flavus* fungus, as it prevented 100% of growth in vitro when applied at a concentration of 2%, which reduced the generation of AFB1.

Growth Performance

According to the data on growth performance, the aflatoxicated group (AFB1-diet) had significantly lower mean values for final body weight, BWG, and feed intake. According to Dalvi and Ademoyero (1984), the impairment of metabolizing capability may cause AFB1's negative influence on growth parameters. These results are consistent with other research that found aflatoxin has a detrimental impact on growth performance (Wade and Sapcota, 2017; Subhani et al., 2018). Furthermore, Mahrose et al. (2021) reported the negative impacts of aflatoxin on the growth performance of Japanese quail. These findings conflict with those of dos Santos et al. (2021), who claimed that after 21 days of feeding, AFB1 had no adverse effects on the performance of the broilers.

Furthermore, according to Mahmood et al. (2011), broilers given either an aflatoxicated diet or an aflatoxin-free diet showed the same growth performance. This discrepancy may be explained by the length of the aflatoxin exposure (experimental period), which was 35 days in our study and 21 days in the dos Santos et al. (2021) trial. In addition to species and sex (Mahrose et al., 2021). Contrarily, the detrimental effects of aflatoxin B1 on the growth performance of broilers fed on the diet for 35 days were reduced when *Aloe vera* concentrations were added to the aflatoxin-contaminated diet.

The addition of *Aloe vera* to the diet dramatically increased growth parameters. *Aloe vera* was said to have a protective effect on rat models of experimental aflatoxicosis. This might be due to its antioxidant and anti-inflammatory qualities (Cui et al., 2017). Additionally, it was demonstrated that *Aloe vera* powder might function as a biosorbent and be utilized as a substitute for removing AFB1 (Zavala-Franco et al., 2018).

The results of the other studies that looked at *Aloe vera*'s potential to improve growth performance in broilers revealed that adding *Aloe vera* powder (0.1%, 0.3%, and 0.5%) to the feed of these broilers did not result in any appreciable differences in terms of body weight gain (Yim et al., 2011). However, special attention must be given to antibacterial activities and improvements in immune response as these two factors may help broilers perform better growth (Yang et al., 2009). Prior research has supported these two properties of *Aloe vera* (antibacterial effect and improvements in immune response). The antibacterial qualities of *Aloe vera* enhance intestinal microflora and decrease pathogens, altering the intestinal shape and enhancing development efficiency. *Aloe vera*, on the other hand, indirectly influences growth performance by boosting

body resistance and strengthening the immunological response.

Aloe vera's enhanced growth-promoting effects have been seen in feed intake, weight gain, and feed efficiency (Table 3). *Aloe vera* can potentially be a growth booster in broiler chicks, and Singh et al. (2013) claim that its effects are similar to those of antibiotic growth promoters. Mmereole (2011) examined the growth-promoting effects of *Aloe vera* and terramysin in broiler chickens and found that the former treatment resulted in significantly faster weight gain. Raziq et al. (2012) found that broilers dosed with water-based infusions of polyherbal plants, including *Aloe vera* (20 mL/L of drinking water), had improved feed efficiency and weight growth. According to Bernard et al. (2016) and Nalge et al. (2017), *Aloe vera* gel extract in drinking water is more efficient than antibiotic growth boosters in improving broiler performance without hurting the birds' overall health. *Aloe vera* has been shown to have many beneficial effects, including increasing feed intake, endogenous digestive enzyme secretion, antioxidation status, and antibacterial capabilities (Toghyani et al., 2011 and Elwan et al., 2019).

Aloe vera supplementation in broilers reduced the mortality rates compared to the positive control group, according to Eevuri and Putturu's (2013) research. According to Yadav et al. (2017), *Aloe vera*'s antibacterial, antioxidant, immune-modulating, antiviral, and anti-inflammatory characteristics may be to blame for the lower mortality rate.

Biochemical parameters

The liver is the target organ for aflatoxin detoxification since reactive 8,9-epoxide is created when aflatoxin is activated. Proteins and DNA can attach to this active form. As a result, aflatoxin-related liver damage is shown by the measurement of the liver enzyme (Denli and Okan, 2006).

The levels of ALT and AST in serum are typically regarded as crucial indicators of the liver's health. These enzymes' serum activity will decrease when the liver is functioning normally. According to Table 4, supplementing with *Aloe vera* decreased AST and ALT levels in the serum, proving that the supplement has no harmful effects on the liver. Accordingly, the ALT, AST, and ALP activities in the aflatoxin group significantly increased in the current trial, as reported by earlier research (Hussain et al., 2016 and Xu et al., 2017), and this is proof that liver damage from toxicity exists. However, these findings vary from those of the earlier study (Zhao et al., 2021), which shows that AFB1 does not affect serum ALT and AST activity. Furthermore, the addition of *Aloe vera* concentrations to the aflatoxin-contaminated meal reduced the harmful effects of aflatoxin B1 on the liver enzymes in broilers fed for 35 days, in line with several studies (Son et al., 2013 and Rahman et al., 2020) where the structural integrity of the cell membrane was maintained while the levels of the liver enzymes dropped.

Table 4 demonstrates how adding *Aloe vera* powder to a diet containing AFB1 dramatically lessened the negative impact of AFB1 on some biochemical markers. The raised activity of AST, ALT, and ALP was improved and restored by adding *Aloe vera* powder to the tainted food. However, there was no appreciable difference between this treatment and the control treatment. The present study's findings indicated that including *Aloe vera* in the diet protects against aflatoxin exposure. The adverse effects of AFB1 exposure appear to have been lessened by adding *Aloe vera* powder to the diet, especially the 2g /kg diet. The antioxidant properties of *Aloe vera* may be responsible for the less severe lesions in chickens given aflatoxin and *Aloe vera* powder. *Aloe vera* polysaccharides have a

potent antioxidative and radical-scavenging ability, according to Cui et al. (2017), which helps to lessen the harm caused by oxidative stress caused by AFB1.

The serum levels of the enzymes alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (ALP), indicators of antioxidant status and liver injury in Japanese quail, did not differ noticeably at 35 days of age (Tariq et al., 2015). According to Fallah (2015), broiler chickens given *Aloe vera* gel (1.5 or 3.0%) in drinking water at 42 days of age showed lower blood activity of ALP, AST, and ALT compared to a control. According to Sinha et al. (2017), adding *Aloe vera* leaves alcoholic extract (2, 5 or 7 g/kg food) to the diet of Jabalpur color birds (32 weeks of age) reduced lipid peroxidation, increased antioxidant status, and protected the liver and kidney. *Aloe vera* is a natural antioxidant that works well to stop pro-oxidants harm to cells and membranes. It can be used as an excellent substitute for synthetic antioxidants. The body's defenses against oxidative stress are strengthened by encouraging the activity of endogenous antioxidant enzymes such as catalase, glucose-6-phosphate dehydrogenase, and superoxide dismutase (Sinha et al. 2017). *Aloe vera* includes a variety of active substances, including glucomannans, acemannans, carotene, and vitamin B12, that contribute to raising the total leucocyte count (Singh et al., 2013).

Aflatoxins cause a wide variety of metabolic losses, and alterations in blood biochemicals are an indication of liver injury and a disruption in the metabolic process (Barati et al., 2018). Researchers Abd El-Ghany et al. (2013), Gholami-Ahangaran et al. (2016), and Naseem et al. (2018) found that the aflatoxicated group's total protein, albumin, and globulin levels were significantly lower than those of the group that just received a basal diet. This is a sign of decreased protein synthesis and the toxic effects of aflatoxin B1 on the liver and kidneys (Abd El-Ghany et al. 2013). As demonstrated in our study, the harmful effects of aflatoxin B1 on the hepatic function in broilers fed on for 35 days, where the total serum protein, albumin, and globulin concentrations were elevated, were reduced by the addition of *Aloe vera* concentrations to contaminated diet with aflatoxin. Regarding renal function, several earlier research (Valchev et al., 2014; Rotimi et al., 2018; and Karamkhani et al., 2020) demonstrated that consuming aflatoxin increased kidney function, including urea, creatinine, and uric acid. In contrast, our results showed that the concentrations of creatinine and uric acid were significantly increased in T2 compared with the other groups. Total blood protein, on the other hand, is crucial for the movement of electrolytes, hormones, enzymes, and vitamins.

Furthermore, albumin is included in this total protein, accounting for a sizeable amount of it. On the other hand, the indicators of protein metabolism are creatinine and uric acid, which form new tissues and support renal function (Mahrose et al., 2021). These details supported our findings that aflatoxin damaged the liver, lowering total protein levels and raising creatinine levels, which in turn impaired growth capacity.

Aflatoxin B1 residue in the liver and meat tissues.

Animal products like meat, eggs, and milk are contaminated when AB1-infected feed is used. It has been demonstrated in previous studies and the current study. Consequently, AFB1 can enter the body of a human through the consumption of infected foods (Fakhri et al., 2019). According to Ferreira et al. (2019), the hazardous characteristics of AFB1 are caused by the bioavailability of AFB1-8.9 epoxy to cellular macromolecules, particularly nucleic acids,

nucleoproteins, and mitochondria, which has cytotoxic effects over both short and extended periods.

The prevalence of aflatoxins in chicken meat and eggs has been documented in earlier investigations. From 115 chicken meat and 80 egg samples, the findings showed that 35% of the chicken and 28% of the egg samples were AFs-contaminated (Iqbal et al., 2014). The Food and Drug Administration (FDA) has established standards for the maximum total AF level authorized in poultry feed, which is 20 ppb in maize and peanut products for chicks and 100 ppb in feed for adult hens (FDA, 2009) (Al-Ruwaili et al., 2018). Five weeks were used to complete this experimental model. The findings showed that compared to chicks only fed with Aflatoxin B1, the level of AFB1 residues in meat samples from the chicks treated with AFB1 with *Aloe vera* powder were significantly reduced or undetectable. Although no research has been done on the effects of *Aloe vera* on AFB1 residues in chicken meat, a study that was done on the results of *Aloe vera* on AFB1 in another animal model indicated that *Aloe vera* powder was very efficient in lowering the toxicity of aflatoxin B1 (Cui et al., 2017). By using lactic acid bacteria in the drinking water of broiler chickens, for example, the toxicity of Aflatoxin B1 in chickens has been reduced, with reductions in AFB1 of 55.46% (1.19 to 0.43 ng/g), and 37.68% (0.69 to 0.53 ng/g), in the leg and breast, respectively, at the end of week 6. AFB1 levels were decreased in the gizzard, kidney, and liver by 60.68% (1.88 to 0.74 ng/g), 60.64% (2.06 to 0.81 ng/g), and 52.73% (1.10 to 0.52 ng/g), respectively. The effects of zeolite on AFB1 residues in laying ducks were discovered in a different investigation. According to Sumantri et al. (2018), zeolite incorporation did not significantly ($P>0.05$) lower the levels of AFB1 and AFM1 in beef, liver, or eggs. Prior research using carvacrol and trans-cinnamaldehyde in poultry feed for controlling *A. flavus* and *parasiticus* growth and Aflatoxin production had positive results in reducing *A. flavus* and *parasiticus* growth and AFB1 production (Wang et al., 2019). Although the addition of AF-binding adsorbent in the feed is used to protect poultry from the harmful effects of AF, it has resulted in decreased mineral and nutrient intake in hens (Edrington et al., 1997) and reduced nutrient consumption in poultry (Chung et al., 1990). As a result, the methods for controlling AF are insufficient, necessitating the development of effective and crucial food safety techniques (Teniola et al., 2005).

5. Conclusion

The results of this investigation verified the remarkable effectiveness of *Aloe vera* gel extract in preventing the development of *A. flavus*, as it prevented 100% of the growth in vitro when administered at 2% concentration, and then the effectiveness of *Aloe vera* gel extract in lowering AFB1 production. This study highlights the new significance of supplementing the aflatoxin-contaminated diet (1 mg AFB1/kg diet) with *Aloe vera* concentrations. It significantly reduced the damaging effects of aflatoxin B1 on the growth performance, blood and serum parameters, and aflatoxin residue in the liver and meat tissue of broilers fed on for 35 days. *Aloe vera* supplementation at 2 g / kg in the diet may lessen aflatoxin B1's harmful effects. However, the outcome of the aflatoxin residue in the liver and muscle tissue is quite significant for public health. *Aloe vera* powder concentrations, notably 2 g, produced the best results when added

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فاعلية الصبار في تثبيط نمو فطر اسبرجلس فلافس وتأثيراته الوقائية على أداء النمو والكيمياء الحيوية لدجاج التسمين المغذى على افلاتوكسين ب١

رضا على حسن ، ابتهال عبد المنعم حسن ، زينب محمد فاروق ، بهاء أبو شحيمه ، مايكل جورجى ، شريف محمد زايد ، محمود الجبالي ، عادل محمد نصر ، السيد أبو الحسن

معهد بحوث الإنتاج الحيواني ، مركز البحوث الزراعية ، الدقي ، الجيزة ، مصر.

مستخلص. هدفت الدراسة إلى تقييم مدى نجاح تراكيز مختلفة من مستخلص جل الصبار (AVGE) في تثبيط نمو فطراسبرجلس فلافس على بيئه أجار دكستروز البطاطس (PDA). وفقًا للبيانات ، تم تثبيط نمو فطراسبرجلس فلافس على بيئه PDA بشكل متفاوت بواسطة مستخلص هلام الصبار بتركيزات ١ و ٢ و ٣ و ٤٪ ؛ كانت هذه المعدلات ٩٧ و ١٠٠ و ٥,٨٨ و ١٧,٦٤٪ على التوالي. كان أفضل تركيز من AVGE في المختبر (٢٪). أجريت دراسة على كتاكيت التسمين للتحقق من التأثيرات الوقائية لمسحوق الصبار (AVP) على الأداء الأنتاجى والمعلمات البيوكيميائية في مصل دجاج التسمين المغذى على علائق ملوثة بالأفلاتوكسينB1 . تم توزيع إجمالي ٤٥٠ كتكوت تسمين (Ross-308) بشكل عشوائي على خمسة معاملات. ١) النظام الغذائي الأساسي (المجموعة الضابطة) ، ٢) عليقه ملوثه مع ١ مجم AFB1 / كجم علف ،3)عليقه ملوثه بالإضافة إلى ١ جم مسحوق صبار/كجم عليقه ، ٤) عليقه ملوثه بالإضافة إلى 2 جم مسحوق صبار/كجم عليقه و ٥) عليقه ملوثه بالإضافة إلى 3 جم مسحوق صبار/كجم عليقه. يتكون كل معامله من ستة مكررات من ١٥ طائر. كان أداء النمو أقل بالنسبة للمجموعات التي تتغذى على ١ مجم افلاتوكسين B1 / كجم علف. في الوقت نفسه ، تم تحسين معايير النمو من خلال دمج ١ و ٢ جم / كجم من مسحوق الصبار في العلف الملوث. كانت الكتاكيت التي تم تغذيتها على عليقه ملوثه تحتوي على نسبة أعلى من انزيمات الكبد ، حمض البوليك ، والكرياتينين مقارنة بالمجموعات الأخرى. بالإضافة إلى ذلك ، فإن مسحوق الصبار يحسن المؤشرات الحيوية لوظائف الكبد والكلى ويقلل من بقايا AFB1 في الكبد والأنسجة العضلية للطيور. أظهرت النتائج أن مسحوق الصبار عند ٢ جم / كجم من النظام الغذائي يمكن أن يقلل من الآثار الضارة للتسمم الفطرىAFB1على كتاكيت التسمين.