SVU-International Journal of Agricultural Sciences
Volume 4 Issue (4) pp.: 121-135, 2022
Print ISSN 2636-3801 | Online ISSN 2636-381X
Doi: 10.21608/svuijas.2022.183363.1260
RESEARCH ARTICLE

The effect of dietary aflatoxin B1, thyme oil, and their combination on sustainability of meat production of broiler chickens

Fawaz, M.A., H.A. Hassan and A.A.A. Abdel-Wareth *

Department of Animal and Poultry Production, Faculty of Agriculture, South Valley University, 83523 Qena, Egypt.

Abstract

This study was conducted to investigate the effect of diet including aflatoxin B1, thyme oil, and their combination on productive performance, nutrient digestibility, and carcass criteria of broiler chickens. A total of 192 one-day-old, unsexed broiler chickens (Ross 308) were divided into four treatment diets. Each treatment included 6 replicates (8 birds per each). During the period from 11-20 days of age, the birds were fed a basal diet without any supplementation (Negative control group; NC), a basal diet supplemented with aflatoxin B1 at 40 µg/kg in (positive control; PC), positive control diet supplemented with thyme oil at 200 mg/kg (Treatment 1; T1) and negative control diet supplemented with thyme oil at 200 mg/kg (Treatment 2; T2). The results indicated that supplementation of a combination of aflatoxin B1 at 40 µg/kg to broilers diet significantly ($P<0.05$) reduced body weight gain during the period of from 21-30 d and 1-38 days of age compared to other treatments. Thyme oil supplementation dramatically improved body weight, body weight gain and feed conversion ratio compared to other treatments. Regarding feed intake, nutrient digestibility, and carcass parameters, there were no appreciable variations between treatments. It could be concluded that thyme oil can reduce the negative impact of aflatoxin B1 in broiler diets.

Keywords: Aflatoxins; Broilers; Sustainability; Phytogenic.

1. Introduction

Animals suffer severe health issues and economic losses as a result of food contamination with various mycotoxin components (Agag 2004; Limaye *et al.*, 2018). Due to the mycotoxin's retention in broiler meat, this issue will also affect others who eat these meats (Alam *et al.*, 2020; Wild and Gong, 2010). A decrease in growth rate is one of aflatoxicosis in broiler chickens' detrimental impacts on the economy (Denli *et al.*, 2009). Aflatoxin A, ochratoxin A, T-2 toxin, nivalenol, zearalenone, and Deoxynivalenol are among the mycotoxins that are most frequently found in food (Huwig *et al.*, 2001; Devegowda *et al.*,1998). A set of chemically identical and dangerous substances known as aflatoxins (aflatoxin B1, B2, G1, and G2) are produced by fungi of the Aspergillus species (Huff *et al.*, 1986).

The two most significant toxigenic fungus involved in the synthesis of aflatoxin are Aspergillus flavus and *Aspergillus parasiticus* (Dutta and Das, 2001). When exposed to a suitable environment, such as one with the right temperature, humidity, CO_2, and oxygen levels in the feed, these toxic fungi create aflatoxin (Abidin *et al.*, 2011). The most physiologically active substance is aflatoxin B1 (Busby and Wogan., 1981). It is important to note that animals exposed to low dietary levels of aflatoxin may experience liver damage, worse reproductive success, and immune system inhibition (Agag,

.*Corresponding author: Ahmed A.A. Abdel-Wareth
Email: a.wareth@agr.svu.edu.eg
Received: December 20, 2022; Accepted: December 30, 2022; Published online: December 30, 2022.

2004; Denli *et al.,* 2009). In broilers, aflatoxicosis can result in a number of symptoms, including enlarged livers, pancreas, and spleens (Daghir, 2008).

Thyme (*Thymus vulgaris* L.), an aromatic plant species of the Lamiaceae family that is used as a culinary spice and in herbal medicine to boost immunity as well as for its antioxidant, antigenotoxic, and antibacterial properties (Mimica-Dukic *et al.,* 2004; De Martino *et al.,* 2009). Pathogenic bacteria can be controlled, digestive enzyme activity can be stimulated, nitrogen absorption can be improved, and excreta odour and ammonia content can be controlled using volatile essential oils (Hippenstiel *et al.,* 2011; Sethiya 2016; Abdel-Wareth, 2016). Essential oils contain varied chemical compositions and concentrations of molecules with biological activity (Simitzis, 2017). Studies have also revealed that thymol and carvacrol, whose percentages are 28.53% and 25.06%, respectively, are the compounds from thyme oil with the greatest biological activity (Abbasi *et al.,* 2020). Additionally, there is some evidence to suggest that the antioxidant properties of thymol and carvacrol (Deighton *et al.,*1993; Aljabeili *et al.,* 2018). This substance contains phenolic, a chemical frequently used as an antibacterial (El-Ghousein and Al-Beitawi, 2009; Toghyani *et al.,* 2010). In addition to those studies, 0.1 and 0.5% thyme significantly (p 0.05) decreased the amount of *E. coli* in the hens' faeces (Bölükbaşi and Erhan, 2007). However, when broilers were fed diets supplemented with 0.1% thyme extract, the quantity of lactic acid bacteria in the ileum considerably increased (Rahimi *et al.,* 2011; Sigolo *et al.,* 2021). The purpose of this study was to evaluate that feeding broiler chickens a diet contaminated with aflatoxin B1, thyme oil, and their combination affected the productivity, nutrient digestibility, and serum metabolic profile of the birds.

2. Materials and Methods

2.1. Experimental animals and design, and feed preparation

This experiment has been carried out in the Experimental Poultry Farm, Department of Animal and Poultry Production, Faculty of Agriculture, South Valley University, Qena, Egypt. The experiment were conducted in accordance with guidelines approved by the Animal Health and Care Committee of South Valley University, Egypt where is a prevailing tropical climate.

A total of 192 one-day-old, unsexed broiler chickens (Ross 308) were divided into four treatments. Each treatment included 6 replicates (8 birds per each). The birds were housed in metabolic wire cages. Chickens had free access to feed and water during the experimental period. The basal diet was formulated according to NRC (1994) to meet the nutrient requirements (Table 1). During the period from 11-20 days of age, the birds were fed basal diet without any supplementation (Negative control group; NC), the birds were fed negative control diet supplemented with aflatoxin B1 at 40 µg/kg in (positive control; PC), the birds were fed positive control diet supplemented with of thyme oil at 200 mg/kg (Treatment 1; T1) and the birds were fed negative control diet supplemented with thyme oil at 200 mg/kg (Treatment 2; T2). All birds were received negative control out of this period (starter diet during 11-20 day of age) and (grower diet during 21-38 day of age) and offered the respective diets for ad libitum consumption and had free access to water for the entire period. The experimental period lasted 38 days.

Aflatoxin B1 from Aspergillus flavus (Purity of aflatoxin B1 ≥ 98%, catalog no. A606874-0005, Sangon Biotech Shanghai Co., Ltd.).

2.2. Productive performance parameters

Feed intake (FI) and body weight (BW) were recorded during the experimental periods. To

determine growth performance (i.e., BW gain) and feed conversion ratio (FCR). Mortality was recorded as it occurred during the entire experimental period. FCR was estimated using the formula:

$$\text{Feed conversion ratio (FCR)} = \frac{\text{Avrage daily feed intake}}{\text{Avrage daily body weight gain}}$$

$$\text{Avrage daily body weight gain (ADWG)} = \frac{\text{Final body weight} - \text{initial body weight}}{\text{period by days}}$$

Table 1. Ingredients and chemical composition of diets

Ingredients, g/kg	Starter diet	Grower diet
Maize, ground	276	300
Sorghum, ground	276	300
Soybean meal (44% CP)	285	250
Corn gluten meal (60% CP)	95.0	60.0
Vit & Min. Premix[a]	3.00	3.00
Sunflower oil	30.0	55.2
Dicalcium phosphate	20.0	18.0
Limestone	10.0	10.00
Salt	3.80	3.80
DL-methionine	0.40	---
L-lysine HCl	1.00	---
Total	1000	1000
Analysis chemical composition, g/kg		
Dry matter	925	924
Crude protein	233	216
Ether extract	53.7	57.5
Crude fibre	25.8	37.8
Ash	67.4	61.8
Ca	13.22	12.84
P	7.05	7.21
GE, MJ/kg	18.55	19.18

[a]Supplied vitamin-mineral premix contains per kg: 2400.000 IU vitamin A; 1000.000 IU vitamin D; 800 mg vitamin K;16.000 IU vitamin E; 650 mg vitamin B1; 1.600 mg vitamin B2; 1.000 mg vitamin B6; 6 mg vitamin B12; 8.000 mg niacin; 400 mg folic acid; 3.000 mg pantothenic acid; 40 mg biotin; 3.000 mg antioxidant; 80 mg cobalt; 2.000 mg copper; 400 mg iodine; 1.200 mg iron; 18.000 mg manganese; 60 mg selenium; 14.000 mg zinc.

2.3. Digestibility trial

Excreta were collected twice a day during the last of each trial period. At the end of the experimental periods, total excreta from each bird were quantified. All excreta were kept in a freezer at a constant temperature of -20 °C until preparation for chemical analysis. Before chemical analysis the excreta were homogenized. Moreover, excreta were oven dried and afterwards, ground finely using a 1 mm sieve with a centrifugal mill. fecal samples were analyzed for determination dry matter by oven drying (930.15), Ash by incineration (942.05), Ether extract by Soxhlet fat analysis (954.02), Crud protein by Kjeldahl (984.13), as described by the AOAC International (2006). The nutrients digestibility was estimated using the formula:

$$\text{Apparent didestibility} = \frac{(\text{Nutrient ingested} - \text{Nutrient excreted in feces})}{\text{Nutrient ingested}} \times 100$$

2.4. Carcass criteria

At 38 day of age, birds were starved overnight with access to water. Twelve birds per treatment (two birds per replicate) were randomly selected, weighted, and sacrificed and plucked. After removal of the head, neck, viscera, shanks, spleen, digestive tract, liver, heart, gizzard and abdominal fat, the rest of the body was weighed to determine the dressed weight. Liver, heart, empty gizzard, spleen, cecum, and abdominal fat from each bird were weighted and calculated as a percentage of live body weight. Dressing percentage was calculated using the formula:

$$\text{Dressing percentage \%} = \frac{\text{hot carcass weight}}{\text{Live body weight}} \times 100$$

2.5. Chemical analysis

The diet and faces were analyzed for dry matter by oven drying (Method Nr.: 930.15), Ash by incineration (Method Nr.: 942.05), Protein by Kjeldahl (Method Nr.: 984.13), and Ether extract by Soxhlet fat analysis (Method Nr.: 920.39), Crude Fiber was determined by the Weende method as described by the AOAC International (2006). Gross energy was determined by Parr adiabatic bomb (Moline, IL, USA).

2.6. Statistical analysis

The statistical analysis was performed separately for each trial using a completely randomized design and the general linear models (GLM) procedure of SAS 9.2 (SAS Institute, 2009). The individual broiler bird was the experimental unit for all analysis. Data were analyzed by one-way ANOVA. Duncan multiple range tests were used to compare means. Significance was declared at $P<0.05$, and a tendency toward significance was declared at $0.05<P<0.10$. P-values less than 0.001 are expressed as "<0.001" rather than the actual value.

3. Results

3.1. Productive performance

Table 2 show the impact of food components, such as aflatoxin B1, thyme oil, and their combination, on the productive performance of broilers. Aflatoxin B1 supplementation at doses of 40 g/kg decreased body weight increase in broilers from (21 to 30 d) of age considerably compared to other treatments. Additionally, broilers fed diets enriched with aflatoxin B1 at 40 g/kg for the periods of (21-30 d), (1-30 d) days of age showed a significantly higher feed conversion ratio than other treatments. In comparison to PC, broilers' feed conversion ratio improved significantly when their diets contained 200 mg/kg of thyme essential oil alone or in conjunction with aflatoxin B1. Throughout the experimentation periods, supplementation in treatments had no impact on feed intake.

3.2. Nutrient digestibility

The effects of dietary supplementation with aflatoxin B1 at 40 µg/kg, thyme oil at 200 mg/kg and their mixture during the periof from 11-20 days of age on nutrient digestibility of broilers are present in (table 3). Aflatoxin B1, thyme oil, and their combination as dietary supplements had no effect on the broiler chickens' digestibility of DM, CP, and EE.

Table 2. The effect of diet including aflatoxin B1, thyme oil, and their combination on productive performance, of broiler chickens.

Items	Treatments				SEM	P-Value
	NC	PC	T1	T2		
Body Weight, g						
1 day	45.6	39.4	42.5	43.4	0.88	0.062
10 days	308	297	292	303	4.88	0.086
20 days	905	797.3	875	870	16.44	0.095
30 days	1878	1714	1987	1885	45.36	0.179
38 days	2593[b]	2376[c]	2697[a]	2583[b]	37.66	0.001
Body weight gain, g						
1-10 days	262	238	250	260	4.02	0.092
11-20 days	597	520	583	567	13.08	0.176
21-30 days	873[b]	817[b]	918[aa]	915[a]	22.19	0.044
31-38 days	815	762	810	798	17.00	0.428
1-38 days	2547[b]	2337[c]	2655[a]	2540[b]	37.03	0.004
Feed intake, g						
1-10 days	329	337	333	334	2.82	0.842
11-20 days	734	677	723	788	11.64	0.269
21-30 days	1433	1404	1401	1447	24.30	0.059
31-38 days	1167	1121	1132	1110	20.50	0.790
1-38 days	3661	3538	3588	3677	35.76	0.837
Feed conversion ratio						
1-10 days	1.252	1.317	1.333	1.285	0.028	0.181
11-20 days	1.229	1.353	1.239	1.389	0.016	0.235
21-30 days	1.641[b]	1.719[a]	1.526[b]	1.581[b]	0.018	0.044
31-38 days	1.432	1.470	1.398	1.390	0.041	0.670
1- 38 days	1.437[b]	1.516[a]	1.352[c]	1.448[b]	0.017	0.002

[a-c] Means not sharing a common superscript in a row are significantly different (P<0.05)
SEM; Standard error of the means.

Table 3. The effect of diet including aflatoxin B1, thyme oil, and their combination on nutrient digestibility of broiler chickens.

Items	Treatments				SEM	P-Value
	NC	PC	T1	T2		
Dry Matter %	82.06	77.52	78.44	79.57	0.966	0.424
Crude Protein %	88.59	86.53	86.48	85.27	0.846	0.639
Ether Extract %	83.49	82.16	81.60	81.62	3.586	0.932

[a-c] Means not sharing a common superscript in a row are significantly different (P<0.05)
SEM; Standard error of the means.

3.3. Carcass criteria

The results of carcass criteria as affected by feeding of aflatoxin B1, thyme oil, and their combination in broiler chickens are given in (Table 4). Aflatoxin B1 at 40 g/kg, thyme oil at 200 mg/kg, and their combination were added to the feed of broilers between the ages of 11 and 20 days without changing their relative weights for dressing, liver, spleen, gizzard, heart, pancreas, or abdominal fat ($p>0.05$). Aflatoxin B1 was not found in the meat samples of broilers that were fed a contaminated meal containing thyme oil at 200 mg/kg, aflatoxin B1 at 40 g/kg, and their combination between 11 and 21 days of age.

Table 4. The effect of diet including aflatoxin B1, thyme oil, and their combination on carcass criteria of broiler chickens.

Items	Treatments (T)				SEM	p Value
	NC	PC	T1	T2		
LBW g	2447	2355	2630	2345.0	83.93	0.667
Dressing%	76.48	77.66	75.98	75.36	0.557	0.574
Liver%	2.188	1.753	1.761	1.582	0.104	0.203
Spleen%	0.112	0.103	0.122	0.1370	0.007	0.421
Gizzard%	1.099	1.027	1.520	1.224	0.078	0.098
Heart%	0.478	0.542	0.472	0.472	0.021	0.670
Pancreas %	0.233	0.164	0.174	0.215	0.015	0.354
Small intestine W%	3.497	3.109	3.431	2.829	0.185	0.618
Small intestine L	170.0	150.0	186.6	167.0	5.395	0.096
Cecum W %	0.728	0.450	0.622	0.773	0.057	0.198
Cecum L	17.33	16.00	17.00	17.66	0.603	0.833
Abdominal fat %	0.706	0.722	0.901	1.273	0.114	0.285

[a-c] Means not sharing a common superscript in a row are significantly different (P<0.05)
SEM; Standard error of the means.

4. Discussion

The impact of broiler food contaminated with high or low levels of aflatoxins on the health and sustainability of production, however, has not been thoroughly explored in the literature. Although most of the experiments produced modestly beneficial benefits, substantial outcomes were infrequent. In the current study, Aflatoxin B1 supplementation at doses of 40 g/kg decreased body weight increase in broilers from (21 to 30 d) of age considerably compared to other treatments. Dietary contamination with aflatoxin B1 levels 100, 200 and 400 ng/g had been significantly ($p<0.05$) reduced feed consumption of broilers compared to the control group (Alam *et al*., 2020). Likewise, Nazarizadeh *et al*. (2019) observed a significant ($p<0.05$) decrease in feed intake of broilers treated with dietary 0,5 g/kg aflatoxin B1 during the period from 1 to 20 days of age, compared to the control group. Also, feed intake was significantly lower in broilers fed contaminated diet with aflatoxin B1 at 0.25 mg/kg than control group (Alharthi *et al*., 2022). Additionally, Khaleghipour *et al*. (2019) reported that supplementation of 2.2 mg/kg aflatoxin B1 to broiler Japanese quail diet during the period from 7 to 35 days of age had been significantly ($p<0.05$) reduced feed intake compared to the control group. Furthermore, Santurio (1999) found that broiler fed diet supplemented with aflatoxin at 3 mg/kg was significantly ($p<0.05$) reduced feed intake compared to non-treated broilers. Moreover, feed

intake was reduced ($p<0.05$) in broilers consumed contaminated feed with 0.8 mg/kg compared to the control diet (Tedesco *et al.,* 2004). Thus, Nazarizadeh and Pourreza (2019) indicated that average daily feed intake was significantly lower in broilers fed a diet supplemented with aflatoxin B1 at 2 and 4 µg/g than the control group. Additionally, Liu *et al*. (2018a) observed that feed intake was reduced ($P<0.05$) in broiler fed diet added with 40 µg/kg of aflatoxin B1 from 1 to 42 days of age compared with the control group. Also, Supplementation of aflatoxin B1 at 0.5 mg/kg to broilers diet from 1 to 42 days of age was significantly ($P<0.05$) reduced feed intake compared to the negative control (Saei *et al.,* 2013). Thus, Liu *et al*. (2018b) found a significant decreased in feed intake of broilers fed contaminated diet with aflatoxin at 2 µg/g compared to control group. Feed intake was significantly ($P<0.05$) decreased in broilers fed contaminated diet with aflatoxin B1 at 2 mg/kg when compared to control group (Yarru *et al*., 2009). In addition, Raju and Devegowda (2000) noted that feed intake was significantly decreased ($P<0.01$) in broilers consumed diet added with aflatoxin B1 at 0.3 mg/kg alone or in combination with ochratoxin A when compared to the control group. However, feed intake did not affect in broilers fed diet supplemented with aflatoxin B1 at 0.5 mg/kg (Rashidi *et al*., 2020). Also, Denli *et al*. (2009) noted that the dietary supplementation of aflatoxin B1 at 1mg/kg had a non-significant effect on feed intake of broiler chickens. In addition, Chen *et al*. (2016) who found that feed intake did not affect in broiler chickens fed 1.5 mg/kg aflatoxin B1 from 1 to 20 days of age. Likewise, Cao and Wang, (2014) stated that supplementation of aflatoxin B1 at 0.4 mg/kg of broilers diet did not affect feed intake. Thus, feed intake was not affected in broilers fed diet supplemented with 2 ppm aflatoxin B1 (Solis-Cruz *et al*., 2019). Average daily body weight gain was significantly decreased ($p<0.05$) in broilers consumed 0.5g/kg aflatoxin B1 compared to control diet (Nazarizadeh *et al*., 2019). Also, Bhatti *et al*. (2016) summarized that a significant decrease ($p<0.05$) had been observed in body weight gain of broiler fed dietary added with 0.1, 0.2 and 0.6 mg/kg aflatoxin B1 compared to control group. In addition, Khaleghipour *et al*. (2019) observed a significant ($p<0.05$) reduce in body weight gain of broiler Japanese quail when fed diet supplemented with 2.2 mg/kg aflatoxin B1 during the period from 7 to 35 days of age compared to non-treated broilers. Thus, Huff, *et al*. (1986) found that supplementation of aflatoxin at 2.5 µg/g diet had been significantly ($p<0.05$) reduced body weight gain of broilers compared to control group. Likewise, Alam *et al*. (2020) who indicated that body weight gain was significantly ($p<0.05$) decreased in broilers consumed dietary added with aflatoxin B1 at 200 and 400 ng/g compared to non-treated broilers. Additionally, daily body weight gain was significantly lower ($p<0.05$) in broilers fed contaminated diet with aflatoxin B1 at 0.25 mg/kg than control group (Alharthi *et al.,* 2022). Also, Santurio (1999) reported that supplementation of aflatoxin at 3 mg/kg to broilers diet had been significantly ($p<0.05$) decreased body weight gain compared to control group. Thus, daily body weight gain was significantly ($p<0.05$) reduced when broilers fed diet added with aflatoxin B1 at 0.5 mg/kg compared with control group (Rashidi *et al*., 2020). Also, Tessari *et al*. (2006) Indicated that body weight gain was significantly ($p<0.05$) lower in broiler chickens receiving a diet added with aflatoxin B1 at 50 and 200 µg/kg of feed. Thus, Denli *et al*. (2009) reported that body weight gain was significantly ($p<0.05$) decreased by supplementation of aflatoxin B1 at 1mg/kg to broilers diet. In addition, Nazarizadeh and Pourreza (2019) who found that body weight gain was lower in broilers fed diet added with aflatoxin B1 at 2 and 4 µg/g than control group. Thus, there were a significant ($P<0.01$) decreased in body weight gain of broilers fed diet added with aflatoxin B1 at 0.3 mg/kg alone or in combination with ochratoxin A when compared

to control group (Raju and Devegowda, 2000). Thus, body weight gain was significantly ($P<0.05$) reduced in broilers fed diet supplemented with 1 and 2 mg/kg (Yarru *et al.,* 2009).

Additionally, Saleh et al. (2014) added different level of thyme oil at 100,200 and 300 mg/kg and observed a significant ($P<0.05$) increase in growth performance when broilers consumed 100 and 200 mg/kg compared with control group. Thus, Addition of thyme powder at 5g/kg significantly increased feed intake and body weight gain in broiler when compared to control group (Fallah and Mirzaet, 2016). Likewise, Bölükbaşi *et al.* (2006) stated that supplemented thyme oil at 100 and 200 mg/kg and indicated a significant increase in body weight and feed intake of broiler compared to control group. Also, Al-Kassie (2009) Found a significant increased ($p<0.05$) in feed intake of broiler consumed 200 ppm from thyme essential oil for 6 weeks compared to control group. In addition, Feed intake was significantly higher in broilers fed diet added with thyme oil at 0.5 and 1g/kg compared to control group (Pournazari *et al.*, 2017).

On other hand, dietary including thyme essential oil at 100 mg/kg did not affect growth performance of broilers (Moustafa *et al.,* 2020). Thus, Hashemipour *et al.* (2013) reported that 60, 100 and 200 mg/kg thymol and carvacrol in broiler diet linearly ($P< 0.001$; quad $p<0.003$) increased FCR in broiler compared to control group. Likewise, Additionally, Demir *et al.* (2008) found that feed intake did not change in broiler fed diet supplemented with 1g thyme powder 1g /kg compared with control group. Likewise, Wade *et al.,* (2018) who noted that supplementation of different level of thyme oil at 100,200 and 300 mg/kg to broiler diet did not affect feed intake. In addition, high level of thyme oil 1.5 and 2g/kg cannot influence feed intake (Attia *et al.,* 2017). Thus, for 42 day feeding, non-significant effect was observed in daily feed intake of broiler fed thyme powder at 5 and 10 g/kg (Toghyani *et al.,* 2010). Feed intake did not affect in broiler consumed 0.3 and 0.6% of thyme extract (Amouzmehr *et al.,* 2012). Dietary including thyme powder at 1g/kg did not changed growth performance value in broiler (Sarica *et al.,* 2005). Feed intake was non-significant increase in broiler fed diet supplemented with thyme extract levels 0.2, 0.4 and 0.6% compared to control group (Pourmahmoud *et al.,* 2013). Likewise, Tekeli *et al.* (2006) who added thyme oil to broiler diet at 120 mg/kg and noted that feed intake was non-different effect. Non- significant different was noted in feed intake when broilers consumed 0.05 and 0.1% thyme essential oil for 28 day (Placha *et al.,* 2019). Thus, Fallah and Mirzaet (2016) who noted that broilers feeding thyme powder at 5g /kg diet had non-significant different in final body weight compared to control group. In addition, Gradual addition of thyme oil from 0.05 up to 0.35 mg/kg broilers diet did not affect on feed intake (Zhu *et al.,* 2014). For example, Moustafa *et al.* (2020) reported that supplementation of thyme oil at 100 mg/kg had been improved feed conversion ratio of broilers compared to control group. Additionally, Ragaa *et al.* (2016) observed a significant improved in FCR when broiler consumed diet supplemented with thyme powder at 1g/kg compared to control group. Likewise, Zhu *et al.* (2014) reported that feed conversion ratio was significantly decreased in broilers fed gradual level of thyme oil from 0.1 up to 0.35 mg/kg compared to control group. Also, El-Ghousein and Al-Beitawi (2009) who found that supplementation of crushed thyme level 0.5, 1, 1.5 and 2% significantly ($p<0.05$) decreased feed conversion ratio compared to control group. (Al-Kassie, 2009) supplemented of thyme essential oil at 100 and 200 ppm to broiler diet and observed a significant ($p<o.o5$) decrease in feed conversion ratio compared to control group.

In the current study, Aflatoxin B1, thyme oil, and their combination as dietary supplements had no effect on the broiler chickens' nutrient digestibility. Furthermore, Matur *et al.* (2010) who added 100 g/kg of supplements to the diet of

Ross 308 female chickens saw a substantial rise in the pancreatic enzymes chymotrypsin and -amylase activity while observing a decline in the activity of lipase when compared to the control group. On the other hand, dietary including on 40 µg/kg of aflatoxin B1 (from 19 to 21 days of age) have been significantly ($p<0.05$) decreased dry matter, crud protein and gross energy digestibility of broilers compared with control group (Liu *et al*., 2018a). Supplementation of aflatoxin B1 at 2.5, 3.13 and 3.91 mg/kg to White Leghorn female chicks diet have been significantly ($p<0.05$) reduced retention of dry matter, ether extract, crud protein and calcium compared to control treatment (Pandey and Chauhan, 2007). Protein utilization and metabolizable energy were significantly depressed when laying hens consumed contaminated diet with aflatoxin B1 at 1 or 2 mg/kg compared to control group (Verma *et al*., 2007). However, Denli *et al*. (2009) noted that crude protein and gross energy digestibility were not affected in broilers fed diet contaminated with 1mg/kg aflatoxin B1. On the other hand, an increase in dry matter and organic matter digestibility, as well as nitrogen metabolisability, was not significant from 7 to 28 days of age when thyme oil was included in the diet at a dose of 120 mg/kg (Cross *et al.*, 2007). Additionally, Hashemipour *et al.* (2013) found that adding 60, 100, and 200 mg/kg thymol and carvacrol to the food of broilers linearly enhanced (p 0 05) the activity of digestive enzymes (trypsin, protease, and lipase) from 1 to 24 day of age, but there were no effects at 42 day of age.

In the present study, Aflatoxin B1 at 40 g/kg, thyme oil at 200 mg/kg, and their combination were added to the feed of broilers between the ages of 11 and 20 days without changing their relative weights for dressing, liver, spleen, gizzard, heart, pancreas, or abdominal fat. To date, various aspects of the aflatoxicosis in poultry farm including effects on broiler performance and carcass criteria have been the subjects of several comprehensive reviews. Internal parts of carcass (gizzard, liver, and pancreas weight) have been indicated to decreased in broilers fed 0.5g/kg of aflatoxin B1 compared with control diet (Nazarizadeh *et al*., 2019). Likewise, Alam *et al*. (2020) who found that dressing percentage of carcass was significantly ($p<0.05$) reduced in broilers fed diet contaminated with 200 and 400 ng/g aflatoxin B1compared to control group. Furthermore, Tessari *et al*. (2006) found that relative weights of the heart was significantly ($p<0.05$) higher in broiler chickens feeding a diet supplemented with aflatoxin B1 at 50 and 200 µg/kg of feed however, liver and spleen weight were not affected. relative weight of liver was significantly higher in broilers fed contaminated feed with 1g/kg aflatoxin B1 however, spleen weight was not affected (Denli *et al*., 2009). The relative weights of the spleen, liver and kidney were significantly ($p<0.05$) increased in broiler fed diet added with aflatoxin at 2.5 µg/g compared to control group (Huff *et al.*, 1986). Furthermore, weight of liver was significantly ($p<0.05$) higher in broilers fed diet contaminated with aflatoxin at 3 mg/kg than control group however, heart and pancreas weight were not affected (Santurio, 1999). Likewise, Raju and Devegowda (2000) indicated that liver and kidney weight were significantly increased in broilers fed diet supplemented with aflatoxin B1 at 0.3 mg/kg compared to control group. Thus, Denli *et al*. (2004) noted that liver weight was ($p<0.05$) higher in broilers consumed 200,300 ng/kg aflatoxin B1 compared to control group. Also, Khaleghipour *et al*. (2019) found that liver and spleen percentage were significantly ($p<0.05$) reduced in broiler Japanese quail fed 2.2 mg/kg aflatoxin B1 during the period from 7 to 35 days of age compared to control group. Liver weight was significantly ($p<0.05$) increased in broiler chickens received contaminated diet with 0.5 mg/kg aflatoxin B1 from 1 to 42 days of age however spleen, Abdominal fat and pancreas weights were non-affected compared to control group (Saei *et al.*, 2013). Likewise, Solis-Cruz *et al*. (2019) who discovered that the relative weight

of liver and spleen were significantly ($p<0.05$) higher in broilers fed contaminated diet with 2ppm aflatoxin B1 during the period from 1 to 21 days of age than control group. Hot carcass and Liver weight were significantly decreased in broiler fed diet added with thyme oil at 100 mg/kg compared to control group (Bölükbaşi *et al.*, 2006). Liver, spleen, heart, gizzard and Pancreas weight were non-significant affected in broiler consumed thyme powder at 1g/kg compared to control group (Sarica *et al.,* 2005). Same result was indicated. Pourmahmoud *et al.* (2013) who found that internal parts (Liver, spleen, heart, gizzard Pancreas and abdominal fat) were non-significant affected when broiler fed thyme extract at 0.2, 0.4 and 0.6%. Thus, Tekeli *et al.* (2006) supplemented thyme oil at 120 mg/kg to broiler diet and noted that hot carcass and abdominal fat were not affected. Non- significant differences was observed in Pancreas, liver, bile, spleen and gizzard percentage in broiler consumed dietary added with thyme oil at 0.5 and 1g/kg compared to control group (Pournazari *et al.*, 2017). It has been found that hot carcass, liver and heart were non-significant affected in broiler fed 300mg/kg thyme oil (Sariözkan *et al.,* 2020). Relative weight of internal parts of broilers (spleen, Pancreas, Cecum, liver and heart) weight were non-significant affect when broilers consumed thyme powder at 5g/ litter drink water however, gizzard weight had a significant increase compared to control group (Sadeghi *et al.*, 2012).

5. Conclusion

It could be concluded that thyme oil can reduce the negative impact of aflatoxin B1 in broiler diets. To assess the ideal aflatoxin B1 dosage, the precise mechanism of action, and its effects on the sustainability of broiler meat production and residues of aflatoxin B1 in broiler meat, additional research under more standardized conditions is still required.

Authors' Contributions

All authors are contributed in this research.

Funding

There is no funding for this research.

Institutional Review Board Statement

All Institutional Review Board Statements are confirmed and approved.

Data Availability Statement

Data presented in this study are available on fair request from the respective author.

Ethics Approval and Consent to Participate

Not applicable

Consent for Publication

Not applicable.

Conflicts of Interest

The authors disclosed no conflict of interest starting from the conduct of the study, data analysis, and writing until the publication of this research work

6. References

Abbasi, M.A., Ghazanfari, S., Sharifi, S.D., Ahmadi Gavlighi. (2020). 'Influence of dietary plant fats and antioxidant supplementations on performance, apparent metabolizable energy and protein digestibility, lipid oxidation and fatty acid composition of meat in broiler chicken', *Veterinary Medicine and Science,* 6(1), pp. 54-68.

Abdel-Wareth, A.A.A. (2016). 'Effect of dietary supplementation of thymol, synbiotic and their combination on performance, egg quality and serum metabolic profile of Hy-Line Brown hens', *British poultry science*, 57, pp. 114-122.

Abidin, Z., Khatoon, A., Numan, M. (2011) 'Mycotoxins in broilers: pathological alterations induced by aflatoxins and ochratoxins, diagnosis and determination, treatment and control of mycotoxicosis', *World's poultry science journal,* 67, pp. 485-496.

Agag, B. (2004). 'Mycotoxins in foods and feeds: 1-aflatoxins', *Ass. Univ. Bull. Environ. Res,,* 7(1), pp. 173-205.

Alam, S., Khan, N.A., Muhammad, A., Jan, I., Hashmi, M.S., Khan, A., Khan, M.O. (2020). 'carryover of aflatoxin b1 from feed to broilers'tissues and its effect on chicken

performance', *fresenius environmental bulletin,* 29, pp. 214-221.

Alharthi, A.S., Al Sulaiman, A.R., Aljumaah, R.S., Alabdullatif, A.A., Elolimy, A.A., Alqhtani, A.H., Abudabos, A.M. (2022). 'Protective Effect of Date Pits on Growth Performance, Carcass Traits, Blood Indices, Intestinal Morphology, Nutrient Digestibility, and Hepatic Aflatoxin Residues of Aflatoxin B1-Exposed Broilers', *Agriculture*, 12(4), pp. 476.

Aljabeili, H.S., Barakat, H., Abdel-Rahman, H.A. (2018). 'Chemical composition, antibacterial and antioxidant activities of Thyme essential oil (*Thymus vulgaris*)', *Food and Nutrition Sciences,* 9, pp. 433.

Al-Kassie, G.A. (2009). 'Influence of two plant extracts derived from thyme and cinnamon on broiler performance', *Pakistan Veterinary Journal*, 29, pp. 169-73.

Amouzmehr, A., Dastar, B., Nejad, J.G., Sung, K.-I., Lohakare, J., Forghani, F. (2012). 'Effects of garlic and thyme extracts on growth performance and carcass characteristics of broiler chicks', *Journal of Animal Science and Technology,* 54, pp. 185-190.

AOAC. (2006). *'Official methods of analysis',* 17th edition', Assoc. Off. Anal. Chem, Arlington, Virginia, USA.

Attia, Y.A., Bakhashwain, A.A., Bertu, N.K. (2017). 'Thyme oil (Thyme vulgaris L.) as a natural growth promoter for broiler chickens reared under hot climate', *Italian Journal of Animal Science,* 16(2), pp. 275-282.

Bhatti, S.A., Khan, M.Z., Saleemi, M.K., Saqib. M. (2016). 'Aflatoxicosis and ochratoxicosis in broiler chicks and their amelioration with locally available bentonite clay', *Pak. Vet. J.,* 36, pp. 68-72.

Bird, F.H. (1978). 'The effect of aflatoxin B1 on the utilization of cholecalciferol by chicks', *Poultry Science,* 57, pp. 1293–1296.

Bölükbaşi, Ş.C., Erhan M.K. (2007). 'Effect of Dietary Thyme (*Thymus vulgaris*) on Laying Hens Performance and *Escherichia coli* (*E. coli*) Concentration in Feces', *International Journal of Natural & Engineering Sciences 1.*

Bölükbaşi, Ş.C., Erhan, M.K., Özkan, A. (2006). 'Effect of dietary thyme oil and vitamin E on growth, lipid oxidation, meat fatty acid composition', *South African Journal of Animal Science*, 36(3), pp. 189-196.

Busby, W.F., Jr., Wogan, G.N. (1981). 'Aflatoxins. Pages 4—27 in I. Mycotoxins and n-nitrosocompounds: Environmental risks', *Vol. 2, R. C. Shank, ed. CR Press Inc., Boca Raton, FL.*

Cao, J., Wang. W. (2014). 'Effects of astaxanthin and esterified glucomannan on hematological and serum parameters, and liver pathological changes in broilers fed aflatoxin-B 1-contaminated feed', *Animal Science Journal*, 85, pp. 150-157.

Chen, X., Naehrer, K., Applegate, T. (2016). 'Interactive effects of dietary protein concentration and aflatoxin B1 on performance, nutrient digestibility, and gut health in broiler chicks', *Poultry science*, 95, pp. 1312-1325.

Cross, D., McDevitt, R., Hillman, K., Acamovic, T. (2007). 'The effect of herbs and their associated essential oils on performance, dietary digestibility and gut microflora in chickens from 7 to 28 days of age', *British poultry science*, 48, pp. 496-506.

Daghir, N.J. (2008). *'Poultry production in hot climates',* Cabi, chapter 8, mycotoxins in poultry feed, pp. 197- 226.

De Martino, L., De Feo, V., Fratianni, F., Nazzaro, F. (2009). 'Chemistry, antioxidant, antibacterial and antifungal activities of volatile oils and their components', *Nat. Prod. Commun.,* 4, pp. 1741–50.

Deighton, N., Glidewell, S.M., Deans, S.G., Goodman, B.A. (1993). 'Identification by EPR spectroscopy of carvacrol and thymol as

the major sources of free radicals in the oxidation of plant essential oils', *Journal of the Science of Food and Agriculture,* 63, pp. 221-225.

Demir, E., Kilinc, K., Yildirim, Y., Dincer, F., Eseceli, H. (2008). 'Comparative effects of mint, sage, thyme and flavomycin in wheat-based broiler diets', *Archiva Zootechnica,* 11(3), pp. 54-63.

Denli, M., Blandon, J., Guynot, M., Salado, S., Perez, J. (2009). 'Effects of dietary AflaDetox on performance, serum biochemistry, histopathological changes, and aflatoxin residues in broilers exposed to aflatoxin B1', *Poultry Science,* 88, pp. 1444-1451.

Denli, M., Okan, F., Doran, F. (2004). 'Effect of conjugated linoleic acid (CLA) on the performance and serum variables of broiler chickens intoxicated with aflatoxin B1', *South African Journal of Animal Science,* 34, pp. 97-103.

Devegowda, G., Raju, M.V.L.N., Afzali, N., Swamm, H.V.L.N. (1998). 'Mycotoxin picture worldwide: novel solutions for their counteractions', *Feed Compounder*, 18(6), pp. 22–27.

Dutta, T., Das, P. (2001). 'Isolation of aflatoxigenic strains of Aspergillus and detection of aflatoxin B 1 from feeds in India', *Mycopathologia,* 151, pp. 29-33.

El-Ghousein, S.S., Al-Beitawi, N.A. (2009). 'The effect of feeding of crushed thyme (Thymus valgaris L) on growth, blood constituents, gastrointestinal tract and carcass characteristics of broiler chickens', *The Journal of Poultry Science,* 46, pp.100-104.

Fallah, R., Mirzaei, E. (2016). 'Effect of Dietary inclusion of turmeric and thyme powders on performance, blood parameters and immune system of broiler chickens', *J. Livestock Sci.,* 7, pp.180–186.

Hashemipour, H., Kermanshahi, H., Golian, A., Veldkamp, T. (2013). 'Effect of thymol and carvacrol feed supplementation on performance, antioxidant enzyme activities, fatty acid composition, digestive enzyme activities, and immune response in broiler chickens', *Poultry science*, 92, pp. 2059-69.

Hippenstiel, F., Abdel-Wareth, A.A.A., Kehraus, S., Südekum, K. (2011). 'Effects of selected herbs and essential oils, and their active components on feed intake and performance of broilers-a review', *Arch. Geflügelk,* 75, pp. 226-234.

Huff, W. E., Kubena, L. F., Harvey, R. B., Hagler Jr, W. M., Swanson, S. P., Phillips, T. D., Creger, C. R. (1986). "Individual and combined effects of aflatoxin and deoxynivalenol (DON, vomitoxin) in broiler chickens" *Poultry Science,* 65(7), pp. 1291-1298.

Huwig, A., Freimund, S., Käppeli, O. Dutler, H. (2001). 'Mycotoxin detoxication of animal feed by different adsorbents" *Toxicology letters*, 122, pp. 179-188.

Khaleghipour, B., Khosravinia, H., Toghiyani, M., Azarfar, A. (2019). 'Effects of silymarin on productive performance, liver function and serum biochemical profile in broiler Japanese quail challenged with dietary aflatoxins', *Italian Journal of Animal Science*, 18 (1), pp. 564-573.

Limaye, a., yu, r.-c., chou, c.-c., liu, j.-r., cheng, k.-c. (2018). 'Protective and detoxifying effects conferred by dietary selenium and curcumin against AFB1-mediated toxicity in livestock: a review', *Toxins,* 10(1), 25.

Liu, N., Ding, K., Wang, J., Deng, Q., Gu, K., Wang, J. (2018a). 'Effects of lactic acid bacteria and smectite after aflatoxin B1 challenge on the growth performance, nutrient digestibility and blood parameters of broilers', *Journal of animal physiology and animal nutrition*, 102, pp. 953-961.

Liu, N., Wang, J. Q., Liu, Z. Y., Chen, Y. K., Wang, J. P. (2018b). 'Effect of cysteamine hydrochloride supplementation on the growth performance, enterotoxic status, and glutathione turnover of broilers fed aflatoxin

B1 contaminated diets', *Poultry science*, 97(10), pp. 3594-3600.

Matur, E., Ergul, E., Akyazi, I., Eraslan, E., Cirakli, Z. (2010). 'The effects of Saccharomyces cerevisiae extract on the weight of some organs, liver, and pancreatic digestive enzyme activity in breeder hens fed diets contaminated with aflatoxins', *Poultry science*, 89, pp.2213-2220.

Mimica-Dukic, N., Bozin, B., Sokovic, M., Simin, N. (2004). 'Antimicrobial and antioxidant activities of *Melissa officinalis* L. (Lamiaceae) essential oil', *J. Agric. Food Chem.,* 52, pp. 2485– 9.

Moustafa, N., Aziza, A., Orma, O., Ibrahim, T. (2020). 'Effect of supplementation of broiler diets with essential oils on growth performance, antioxidant status, and general health', *Mansoura Veterinary Medical Journal*, 21, pp. 14-20.

Nazarizadeh, H., Mohammad Hosseini, S., Pourreza, J. (2019). 'Effect of plant extracts derived from thyme and chamomile on the growth performance, gut morphology and immune system of broilers fed aflatoxin B1 and ochratoxin A contaminated diets', *Italian Journal of Animal Science*, 18, pp. 1073-1081.

Nazarizadeh, H., Pourreza, J. (2019). 'Evaluation of three mycotoxin binders to prevent the adverse effects of aflatoxin B1 in growing broilers', *Journal of Applied Animal Research*, 47, pp. 135-139.

NRC [National Research Council]. (1994). *'Nutrient requirements of poultry',* 9th revised ed., National Academie Press, Washington, D. C., 155 pp.

Osborne, D., Hamilton, P. (1981). 'Decreased pancreatic digestive enzymes during aflatoxicosis', *Poultry Science,* 60, pp. 1818-1821.

Osborne, D.J., Wyatt, R.D., Hamilton, P.B. (1975). 'Fat digestion during aflatoxicosis in broiler chickens', *Poultry Science,* 54, pp. 1802-1802 (Abstract).

Pandey, I., Chauhan, S. (2007). 'Studies on production performance and toxin residues in tissues and eggs of layer chickens fed on diets with various concentrations of aflatoxin AFB1', *British Poultry Science,* 48, pp. 713-723.

Placha, I., Ocelova, V., Chizzola, R., Battelli, G., Gai, F., Bacova, K., Faix, S., (2019). 'Effect of thymol on the broiler chicken antioxidative defence system after sustained dietary thyme oil application', *British poultry science*, 60, pp. 589-596.

Pourmahmoud, B., Aghazadeh, A.M., Sis, N.M. (2013). 'The effect of thyme extract on growth performance, digestive organ weights and serum lipoproteins of broilers fed wheat-based diets', *Italian Journal of Animal Science,* 12:e53.

Pournazari, M., AA-Qotbi, A., Seidavi, A., Corazzin, M. (2017). 'Prebiotics, probiotics and thyme (Thymus vulgaris) for broilers: Performance, carcass traits and blood variables', *Revista Colombiana de Ciencias Pecuarias,* 30, pp. 3-10.

Ragaa, N.M., Korany, R.M., Mohamed, F. (2016). 'Effect of thyme and/or formic acid dietary supplementation on broiler performance and immunity', *Agriculture and Agricultural Science Procedia,* 10, pp. 270-279.

Rahimi, S., Teymouri, Z.Z., Karimi, T.M., Omidbaigi, R., Rokni, H. (2011). 'Effect of the three herbal extracts on growth performance, immune system, blood factors and intestinal selected bacterial population in broiler chickens', *Journal of Agricultural Science and Technology,* 13(4), 527-539

Raju, M., Devegowda, G. (2000). 'Influence of esterified-glucomannan on performance and organ morphology, serum biochemistry and haematology in broilers exposed to individual and combined mycotoxicosis (aflatoxin, ochratoxin and T-2 toxin)', *British poultry science,* 41, pp. 640-650.

Rashidi, N., Khatibjoo, A., Taherpour, K., Akbari-Gharaei, M., Shirzadi, H. (2020). 'Effects of licorice extract, probiotic, toxin binder and poultry litter biochar on performance, immune function, blood indices and liver histopathology of broilers exposed to aflatoxin-B1', *Poultry Science*, 99(11), pp. 5896-5906.

SAS, Institute. (2009). *'User's Guide: Statistics',* Version 9.2. SAS Institute, Inc., Cary, NC, USA.

Sadeghi, G., Karimi, A., Padidar Jahromi, S., Azizi, T., Daneshmand, A. (2012). 'Effects of cinnamon, thyme and turmeric infusions on the performance and immune response in of 1-to 21-day-old male broilers', *Brazilian Journal of Poultry Science*, 14, pp. 15-20.

Saei, M.M., Sadeghi, A.A., Ahmadvand, H. (2013). 'The effect of Myrtus communis oil extract on growth performance, serum biochemistry and humoral immune responses in broiler chicks fed diet containing aflatoxin B1', *Archives Animal Breeding*, 56, pp. 842-850.

Saleh, N., Allam, T., El-Latif, A., Ghazy, E. (2014). 'The effects of dietary supplementation of different levels of thyme (*Thymus vulgaris*) and ginger (Zingiber officinale) essential oils on performance, hematological, biochemical and immunological parameters of broiler chickens', *Global Vet.*, 12, pp. 736-744.

Santurio, J. (1999). 'Effect of sodium bentonite on the performance and blood variables of broiler chickens intoxicated with aflatoxins', *British Poultry Science*, 40, pp. 115-119.

Sarica, S., Ciftci, A., Demir, E., Kilinc, K., Yildirim, Y. (2005). 'Use of an antibiotic growth promoter and two herbal natural feed additives with and without exogenous enzymes in wheat based broiler diets', *South African Journal of Animal Science*, 35, pp. 61-72.

Sariözkan, S., Güçlü, B.K., Konca, Y., Aktuğ, E., Kaliber, M., Beyzi, S.B., Şentürk, M. (2020). 'The effects of thyme essential oil and vitamin combinations on performance, carcass quality and oxidation parameters in broilers exposed to heat stress', *Ankara Üniversitesi Veteriner Fakültesi Dergisi,* 67, pp. 357-64.

Sethiya, N K. (2016). 'Review on natural growth promoters available for improving gut health of poultry: an alternative to antibiotic growth promoters', *Asian J. Poult. Sci.,* 10, pp.1–29.

Sigolo, S., Milis, C., Dousti, M., Jahandideh, E., Jalali, A., Mirzaei, N., Rasouli, B., Seidavi, A., Gallo, A., Ferronato, G. (2021). 'Effects of different plant extracts at various dietary levels on growth performance, carcass traits, blood serum parameters, immune response and ileal microflora of Ross broiler chickens', *Italian Journal of Animal Science,* 20, pp. 359-371.

Simitzis, P. E. (2017).'Enrichment of animal diets with essential oils-agreat perspective on improving animal performanceand quality characteristics of the derived products', *Medicines,* 4, pp. 35-52.

Solis-Cruz, B., Hernandez-Patlan, D., Petrone, V.M., Pontin, K.P., Latorre, J.D., Beyssac, E., Hernandez-Velasco, X., Merino-Guzman, R., Arreguin, M.A., Hargis, B.M. (2019). 'Evaluation of a Bacillus-based direct-fed microbial on aflatoxin B1 toxic effects, performance, immunologic status, and serum biochemical parameters in broiler chickens', *Avian diseases*, 63, pp. 659-669.

Tedesco, D., Steidler, S., Galletti, S., Tameni, M., Sonzogni, O., Ravarotto, L. (2004). 'Efficacy of silymarin-phospholipid complex in reducing the toxicity of aflatoxin B1 in broiler chicks', *Poultry science*, 83, pp. 1839-1843.

Tekeli, A., Celik, L., Kutlu, H., Gorgulu, M. (2006). *'Effect of dietary supplemental plant extracts on performance, carcass characteristics, digestive system development, intestinal microflora and some blood parameters of broiler chicks',* In

Proceedings of 12th European Poultry Conference, pp. 10-14.

Tessari, E., Oliveira, C., Cardoso, A., Ledoux, D., Rottinghaus, G. (2006). 'Effects of aflatoxin B1 and fumonisin B1 on body weight, antibody titres and histology of broiler chicks', *British poultry scien*ce, 47, pp. 357-364.

Toghyani, M., Tohidi, M., Gheisari, A.A., Tabeidian, S.A. (2010). 'Performance, immunity, serum biochemical and hematological parameters in broiler chicks fed dietary thyme as alternative for an antibiotic growth promoter', *African Journal of Biotechnology*, 9, pp. 6819-6825.

Verma, J., Johri, T.S., Swain, B.K. (2007). 'Effect of aflatoxin, ochratoxin and their combination on protein and energy utilisation in white leghorn laying hens', *Journal of the Science of Food and Agriculture*, 87, pp. 760-764.

Wild, C.P., Gong, Y.Y. (2010). 'Mycotoxin and human diseases: a largely ignored global health issue', *Carcinogenesis*, 31, pp. 71–82.

Yarru, L.P., Settivari, R.S., Antoniou, E., Ledoux, D.R., Rottinghaus, G.E. (2009). 'Toxicological and gene expression analysis of the impact of aflatoxin B1 on hepatic function of male broiler chicks', *Poultry Science*, 88(2), pp. 360-371.

Zhu, X., Liu, W., Yuan, S., Chen, H. (2014). 'The effect of different dietary levels of thyme essential oil on serum biochemical indices in Mahua broiler chickens', *Italian Journal of Animal Science*, 13, pp. 3238.