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# Effect of Different Levels of Silymarin (Silybum marianum) on Growth Rate, Carcass Variables and Liver Morphology of Broiler Chickens Contaminated with Aflatoxin B<sub>1</sub>

Fani Makki O1., Afzali N1 & Omidi A1,2

<sup>1</sup>Department of Animal Sciences, Faculty of Agriculture, University of Birjand, Birjand, Iran. <sup>2</sup>Department of Animal Health Management, School of Veterinary Medicine, Shiraz University, Shiraz, Iran.

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# Abstract

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**Corresponding author:** Arash Omidi, Ph.D. aomidi@shirazu.ac.ir

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This experiment was conducted to evaluate the ability of Silybum marianum seeds (SMS) on performance, carcass variables, and liver morphology of the broiler chickens contaminated with aflatoxin B1 (AFB1). A total of 216 broiler chicks (Ross 308) were used. Birds were randomly assigned to nine treatment groups, with four replicates and six birds in each replicate. Chickens were reared on litter from 1 to 35 days of age. Treatments were (AFB1) in three levels (Zero, 250 and 500 ppb) and SMS in three levels (Zero, 0.5 and 1.0 percent) using factorial experiment based on completely randomized design. Feed intake at the end of the three weeks did not significantly change in comparison with the control group. With the increase in the level of (AFB<sub>1</sub>) in the diet, feed intake and body weight gain were decreased compared with the control group in week 4. Feed conversion ratio was not influenced by the treatments. In diets containing AFB1, breast muscle, carcass ratio, abdominal fat and bursal gland weight were significantly decreased (P<0.05), whereas the relative weight of the liver, pancreas, gizzard, proventriculus and full intestine were significantly increased (P<0.05). Feeding AFB1 alone did not affect thigh, back, neck, wings, heart, legs and spleen weights. Increasing the level of SMS in the diet alone or in combination with  $AFB_1$  resulted in significant changes in the weights of carcass and internal organs. Liver of birds fed diets containing AFB1 showed abnormal signs including enlargement, yellowish, friable and rounded shape. Liver of other treatments did not show any abnormal signs. In conclusion, these findings suggest that silymarin might be used in chickens to prevent the effects of AFB1 in contaminated feed

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#### Introduction

Aflatoxins, the toxic secondary metabolites of various *Aspergillus spp.*, are commonly encountered in a wide variety of tropical and subtropical feeds (Galvano *et al.*, 2005). These are difuranceoumarin compounds and mainly include aflatoxins B<sub>1</sub>, B<sub>2</sub>, G<sub>1</sub>, G<sub>2</sub>, and M<sub>1</sub>. The most toxic and commonly occurring aflatoxin in feed is aflatoxin B<sub>1</sub> (Yunus *et al.*, 2011). Aflatoxin B<sub>1</sub> biotransformed in the liver by monoxygenases and then transformed by cytochrome P<sub>450</sub> into aflatoxin 8,9 epoxide (Emerole *et al.*, 1979), a highly active electrophilic compound which is inactivated by conjugation with glutathione and excreted through the urine and bile (Essigmann *et al.*, 1982). Aflatoxicosis in chickens is characterized by high mortality, anorexia, decreased growth rates, negative feed conversions, fatty liver, decreased egg production, poor pigmentation, and increased susceptibility to some diseases such as infections (Arafa *et al.*, 1981; Doerr *et al.*, 1983).

The bioactive extract from Silybum marianum seed (SMS), silymarin, contains a mixture of flavonolignans and a residual fraction that has not been defined chemically in structural details (Skottova et al., 2003). Silymarin is used in humans for the treatment of numerous liver disorders characterized by degenerative necrosis and functional impairments (Luper, 1998). Its mechanism of action still seems to be poorly understood, but data in the literature suggest that it acts as an antioxidant, cell membrane stabilizer and permeability regulator, as well as a promoter of DNA, RNA, and protein synthesis (Magliulo et al., 1973). Kalorey et al. (2005), reported that SMS improved body weight and feed intake in the presence of aflatoxin B1 in feed, while it had no effect on the feed conversion ratio (Tedesco et al., 2004). Similarly, Gowda and Sastry (2000) confirmed a significant improvement of SMS on body weight gain and attributed its effects to antioxidant activity in the protein synthesis stimulation by the bird's enzymatic system. The higher weight gain was reported by Chakarverty and Parsad (1991), in SMS supplemented group. Kalorey et al. (2005) reported the protective role of SIMS against aflatoxicosis on the weight of bursa of fabricius. As evident from some researches, aflatoxins reduced lymphoid organs weight (thymus, bursa and spleen) in aflatoxicosis (Tedesco et al., 2004). Silybum marianum was more efficient to protect the spleen against adverse effects of aflatoxin as compared with the synthetic toxin binders (Kalorey et al., 2005).

The present study was conducted to evaluate the effects of *Silybum marianum* seeds on growth rate, carcass variables and liver morphology of broiler chickens contaminated with aflatoxin B<sub>1</sub>.

chickens (as rea-)			
Feed Stuffs (%)	Starter Period	Grower Period	Finisher Period
	(1-14 day)	(14-28 day)	(28-35)
Corn	54.46	50.42	45.96
Soybean meal (44% CP)	35.00	30.29	25.56
Wheat	-	10.00	20.00
Fish meal (60% CP)	3.07	2.04	1.06
Soybean oil	3.29	3.57	3.76
Dicalcium phosphate	1.73	1.47	1.49
Oyster shell	1.16	1.04	1.02
Mineral Premix <sup>2</sup>	0.25	0.25	0.25
Vitamin premix <sup>3</sup>	0.25	0.25	0.25
Salt	0.20	0.20	0.20
DL-methionine	0.35	0.28	0.24
L-lysine	0.24	0.19	0.21
Analyzed values			
ME (Kcal/Kg)	2980	3050	3100
CP (%)	22	20	18
Lys (%)	1.43	1.24	1.09
Met+Cys (%)	1.07	0.95	0.86
Thr (%)	0.31	0.28	0.26
Ca (%)	1.05	0.90	0.85
P (%)	0.52	0.45	0.42

Table 1. Composition of the basal starter, grower and finisher diets fed to broiler chickens (as fed<sup>1</sup>)

<sup>1</sup>For preparing Aflatoxin (AF) and *Silybum marianum* seed (SMS) diets, AF and SMS was added to the basal diets at 0, 250, 500 ppb and 0, 0.5, 1 % respectively.

<sup>2</sup>Provided at the following rates per kilogram of diet: Mn (from MnSO<sub>4</sub>-H<sub>2</sub>O), 0.63 mg; Zn (from ZnO), 0.52 mg; Fe (from FeSO<sub>4</sub>-7H<sub>2</sub>O), 22 mg; Cu (from CuSO<sub>4</sub>-5H<sub>2</sub>O), 3 mg; I (from Ca (IO<sub>3</sub>)<sub>2</sub>-H<sub>2</sub>O), 0.63 mg; Se, 0.08 mg (from sodium selenite).

<sup>3</sup>Provided at the following rates per kilogram of diet: 3400 IU vitamin A, 800 IU vitamin D<sub>3</sub>, 11 IU vitamin E, 0.74 mg vitamin B<sub>1</sub>, 4.3 mg vitamin B<sub>2</sub>, 0.4 mg vitamin B<sub>3</sub>, 1.6 mg vitamin B<sub>6</sub>, 0.41 mg vitamin B<sub>12</sub>, 1.8 mg vitamin K<sub>3</sub>, 0.6 mg folic acid, 1.8 mg H<sub>2</sub>, 200 mg Choline chloride.

#### Materials and Methods

## Experimental birds and diets

In this study, broiler chicks were fed for starter (1-14 days), grower (14-28 days) and finisher (28-35 days) periods. A basal diet without inclusion of either aflatoxin or binder was formulated to meet the nutrient requirements of broiler chicks based on Ross-308 strain recommendation (Table 1). The study was performed by 216 broiler chicks (Ross-308). They were randomly divided into 9 treatment groups; each had 4 replicates of 6 broiler chicks. Thirty six stainless steel cages (40×65×98cm) were used to house the chicks. The chicks had *ad libitum* access to the feed and water. The experiment lasted for 35 days. Treatments were (AFB<sub>1</sub>) in three levels (Zero, 250 and 500 ppb) and SMS in three levels (Zero, 0.5 and 1.0

percent). Powdered SMS and contaminated rice AFB<sub>1</sub> were added to basal diet to achieve the administered dose accurately. The experiment was approved by the animal welfare committee of the Agriculture Faculty of Birjand University, Birjand, Iran.

#### Measured variables

Feed intake (FI) and body weight gain (BWG) were measured every week for each replicate and feed conversion ratio (FCR) was calculated. Two birds from each treatment group were randomly selected at the end of study and killed by cervical dislocation. The relative weight of the carcass, thigh, breast, intestine, abdominal fat and internal organ weights were then recorded.

#### Statistical Analyses

The data were analyzed in a completely randomized design with a 3×3 factorial arrangement using SAS software (SAS Inst., Inc, Cary, NC, 2001). FI, BWG and FCR were analyzed using repeated measures design. The treatments mean were compared by Tukey–Kramer and least squares with their means ± standard errors were reported.

#### Results

The results of feed intake, body weight gain and feed conversion ratio during different stages of the study are presented in Tables 2, 3 and 4, respectively. The feed intake and body weight gain were severely depressed in birds receiving AFB1 contaminated feed without SMS as toxin binder. The results of this experiment showed that interaction between different levels of SMS and AFB<sub>1</sub> had an effective impact on increasing studied parameters from the fourth week onwards. Also, FI and BWG were higher in the treatment containing SMS alone compared with the groups contaminated with AFB<sub>1</sub>. Feed conversion ratio (FCR) was not influenced by treatments, however at main aflatoxin levels effect FCR increased significantly (P<0.05) during the third and fourth weeks. All levels of SMS had significantly beneficial effects on FCR (P<0.01). The percentages of thigh, back & neck, wings and legs relative to broiler body weight were not influenced by different levels of  $AFB_1$  and SMS (Table 5). The relative weights of the internal organs are shown in Table 6. In diet containing AFB<sub>1</sub>, abdominal fat and bursal gland weight were significantly decreased, whereas the relative weights of the liver, pancreas, gizzard, proventriculus, and full intestine were significantly increased (P<0.05). The liver morphology of the birds fed diets containing AFB<sub>1</sub> was enlarged, yellowish, friable and rounded (Figure 1).

Treat	ment	Weeks of experiment						
Aflatoxin	SMS	<b>W</b> 71, 1	M/L O	Wk 3	Wk 4	1471 - E		
(ppb)	(percent)	Wk 1	Wk 2	VVK S	VVK 4	Wk 5		
0	0	125.3	265.6	507.6	773.4a	851.8a		
250	0	123.6	240.2	500.3	596.6 <sup>b</sup>	721.4 <sup>b</sup>		
500	0	121.8	235.3	496.6	580.8 <sup>b</sup>	654.2c		
0	0.5	126.6	265.8	507.4	750.6a	851.8 <sup>a</sup>		
250	0.5	127.6	261.1	521.6	745.7 a	843.3ª		
500	0.5	126.2	260.9	502.7	739.4 <sup>a</sup>	830.3 <sup>a</sup>		
0	1.0	130.4	268.4	514.4	756.6 <sup>a</sup>	863.3ª		
250	1.0	128.7	264.7	515.8	734.9a	849.1ª		
500	1.0	128.1	257.6	519.9	730.4a	840.1ª		
SEM		1.56	5.47	8.12	18.92	14.93		
Main effec	t							
AFB <sub>1</sub> (ppb	)							
0	)	127.4	266.4 a	509.8 <sup>ь</sup>	760.2 a	855.6 a		
25	0	126.6	255.3 ь	512.5 ª	692.4 b	804.5 b		
50	0	125.4	251.2 ь	506.4 b	683.5 c	774.8 c		
SMS (Perce	ent)							
0		123.5	247.1 ь	501.5 a	650.2 b	742.4 c		
0.	5	126.8	262.6 a	510.5 ab	745.2 ª	841.7 <sup>b</sup>		
1.	0	129.1	263.5 a	516.7 ª	740.6 a	850.8 a		
Probabiliti	es							
AFB <sub>1</sub>		Ns	0.05	0.01	0.01	0.01		
SMS		Ns	0.01	0.05	0.01	0.01		
AFB <sub>1</sub> ×SMS	5	Ns	Ns	Ns	0.05	0.01		

Table 2. Effect of Aflatoxin B1 (AFB<sub>1</sub>) and *Silybum marianum* seeds (SMS) on Feed intake of broiler chickens (gr)

AFB<sub>1</sub>, Aflatoxin B<sub>1</sub>. SMS, *Silybum marianum* seeds. <sup>a-c</sup> Mean values within a column with no common superscript differ significantly from each other (P<0.05). Ns, not significant.



Liver of birds received diets with: T1) Control, T2) 250 ppb of  $AFB_1$ , T3) 500 ppb of  $AFB_1$ , T4) 0.5 percent of SMS, T5) 0.5 percent of SMS Plus 250 ppb  $AFB_1$ , T6) 0.5 percent of SMS Plus 500 ppb of  $AFB_1$ , T7) 1.0 percent of SMS, T8) 1.0 percent of SMS Plus 250 ppb  $AFB_1$  and T9) 1.0 percent of SMS Plus 500 ppb of  $AFB_1$ .

# Figure 1. Liver of birds fed diets containing AFB<sub>1</sub>were enlarged, yellowish, friable and rounded in comparison with the control group.

#### Discussion

The weight loss in the breast muscles and carcass ratio may be due to the high level of AFB<sub>1</sub> in the diet. In the first three weeks of the experimental period, there were no differences in FI among the treatments. After 3 weeks of treatment, the feed intake of the AFB<sub>1</sub> group was lower compared with the other groups (P<0.05). On the other hand, the presence of SMS in aflatoxin contaminated feed improved feed intake. The results of this study support the findings of Kalorey *et al.* (2005), who reported that SMS improved feed intake in the presence of AFB<sub>1</sub> in feed. Our results are in agreement with Tedesco *et al.* (2004), who observed improved feed

intake in SMS treated group as compared with the birds fed on AFB<sub>1</sub> contaminated feeds only.

In the first two weeks of the experimental period, there were no differences in BWG among the treatment groups. The mean body weight gain after the third week of the experimental period (Table 3) was lower in the AFB<sub>1</sub>-treated groups compared with the other groups (P<0.05). These results suggest that treatment with silymarin may be effective in counteracting the negative effects of AFB<sub>1</sub> intoxication on feed intake and BWG in growing broilers. Gowda and Sastry (2000) confirmed the improvement of SMS on body weight gain and attributed these effects to antioxidant activity that stimulated protein synthesis by the bird's enzymatic system. The higher weight gain was reported by Tedesco *et al.* (2004) in SMS supplemented group. However, Feed conversion ratio was not influenced by treatments (Tedesco *et al.*, 2004).

Table 3. Effect of Aflatoxin B<sub>1</sub> (AFB<sub>1</sub>) and *Silybum marianum* seeds (SMS) on Body weight gain of broiler chickenss(gr)

Treat	ment		W	eeks of experi	nent	
Aflatoxin	SMS	Wk 1	Wk 2	Wk 3	Wk 4	Wk 5
(ppb)	(Percent)	VVK 1	VVK Z	VVK S	VV K 4	VVK S
0	0	94.2	185.7	368.1ª	492.4ª	439.1ª
250	0	90.2	167.1	337.8 <sup>ab</sup>	353.1 <sup>b</sup>	354.9 <sup>b</sup>
500	0	87.6	161.1	323.5 <sup>b</sup>	349.9 <sup>b</sup>	302.8c
0	0.5	95.1	182.5	368.3ª	497.9ª	458.8a
250	0.5	93.8	181.9	366.1ª	490.6 <sup>a</sup>	450.7ª
500	0.5	93.5	182.5	366.3ª	480.2ª	443.5 <sup>a</sup>
0	1.0	94.5	189.1	369.1ª	507.8a	450.8a
250	1.0	95.3	183.3	369.4ª	477.3ª	452.8 <sup>a</sup>
500	1.0	93.5	176.3	364.4a	459.4ª	441.1ª
SEM		3.01	6.16	12.49	9.18	
Main effec	t					
AFB <sub>1</sub> (ppb	)					
(	)	94.65	185.7 a	368.5 ª	<b>499.4</b> a	499.6 a
25	50	93.11	177.4 ь	357.7 ь	440.3 b	419.4 ь
50	00	91.65	173.3 bc	351.4 <sup>bc</sup>	429.8 c	395.8 c
SMS (Perce	ent)					
(	)	90.61 <sup>ab</sup>	171.3 ь	343.1 ь	398.5 °	365.6 ь
0	.5	94.11ª	182.3 a	366.9 a	489.6 a	451.1 ª
1	.0	94.40a	182.9 a	367.6 ª	481.5 ab	448.2 a
Probabiliti	es					
AFB <sub>1</sub>		Ns	0.05	0.05	0.01	0.01
SMS		0.05	0.01	0.01	0.05	0.05
AFB <sub>1</sub> ×SMS	5	Ns	Ns	0.05	0.05	0.01

 $AFB_1$ , Aflatoxin  $B_1$ . SMS, *Silybum marianum* seeds. <sup>a-c</sup> Mean values within a column with no common superscript differ significantly from each other (P<0.05). Ns, not significant.

The results showed that increasing the level of  $AFB_1$  from 250 to 500 ppb in broiler diets, breast, carcass and thigh yields were decreased. In contrast, thigh, back & neck, legs and wings of broilers were not influenced by different levels of  $AFB_1$  and SMS. However, the highest breast weight muscle was observed in birds consumed 1.0 percent of SMS, compared with the groups received the different levels of  $AFB_1$  (P<0.05), (Chand *et al.*, 2011). Zahid and Durrani (2007) fed different levels of SMS to broilers and found significantly higher breast weights at the level of 15 g per kg of feed. The findings of the present study are supported by Chand *et al.* (2011), who reported reduced weight of the pancreas by feeding of  $AFB_1$ contaminated feeds to broilers. The results of the study were shown that with an increase in level of  $AFB_1$  from 250 to 500 ppb in poultry diets, relative weights of gizzard and full intestine weight were increased, and in contrast, the abdominal fat pad was decreased (P<0.05).

Table 4. Effect of Aflatoxin  $B_1$  (AFB<sub>1</sub>) and Silybum marianum seeds (SMS) on Feed conversion ratio of broiler chickens

Treat	ment		W	eeks of exper	iment	
Aflatoxin (ppb)	SMS (Percent)	Wk 1	Wk 2	Wk 3	Wk 4	Wk 5
0	0	1.33	1.43	1.38	1.57	1.93
250	0	1.37	1.43	1.48	1.68	1.96
500	0	1.39	1.46	1.56	1.65	2.03
0	0.5	1.33	1.45	1.40	1.50	1.85
250	0.5	1.36	1.43	1.43	1.51	1.87
500	0.5	1.34	1.42	1.38	1.53	1.87
0	1.0	1.37	1.41	1.39	1.52	1.91
250	1.0	1.35	1.44	1.41	1.53	1.87
500	1.0	1.37	1.46	1.44	1.52	1.91
SEM		0.04	0.03	0.04	0.04	0.05
Main effect AFB <sub>1</sub> (ppb)						
	)	1.34	1.43	1.39 <sup>b</sup>	1.53 <sup>b</sup>	1.89
2	50	1.36	1.43	1.44 <sup>ab</sup>	1.57 a	1.90
50	00	1.37	1.45	1.46 a	1.56 a	1.93
SMS (Perce	nt)					
. (	)	1.36	1.44	1.47 a	1.63 a	1.97 a
0	.5	1.34	1.44	1.40 <sup>b</sup>	1.51 b	1.86 <sup>b</sup>
1	.0	1.36	1.44	1.41 <sup>b</sup>	1.52 b	1.89 b
Probabilitie	S					
AFB <sub>1</sub>		Ns	Ns	0.05	0.01	Ns
SMS		Ns	Ns	0.01	0.01	0.01
AFB <sub>1</sub> ×SMS		Ns	Ns	Ns	Ns	Ns

AFB<sub>1</sub>, Aflatoxin B<sub>1</sub>. SMS, *Silybum marianum* seeds. <sup>a-c</sup> Mean values within a column with no common superscript differ significantly from each other (P<0.05). Ns, not significant.

Treat	ment	Со	mponent (	as percent	of live bod	y weight)	
Aflatoxin (ppb)	SMS (Percent)	Carcass	Thigh	Breast	Back & neck	Legs	Wings
0	0	71.22 a	22.34	22.21 ab	19.37	4.32	7.66
250	0	65.21 ab	20.34	20.31 bc	18.47	4.11	7.62
500	0	60.91 b	19.23	19.27 c	17.40	4.93	7.43
0	0.5	71.71 a	22.55	22.35 ab	19.34	4.26	7.83
250	0.5	71.84 a	22.87	22.21 ab	19.73	4.24	7.78
500	0.5	68.99 ab	22.44	21.35 <sup>abc</sup>	19.46	4.14	7.77
0	1.0	71.97 a	23.09	23.64 a	20.55	4.57	7.88
250	1.0	72.58 a	22.47	22.21 ab	19.60	4.35	7.82
500	1.0	67.56 <sup>ab</sup>	22.30	22.13 ab	19.36	4.15	7.81
SEM		2.71	1.20	0.80	0.99	0.24	0.72
Main effect AFB <sub>1</sub> (ppb)							
(		71.63 a	22.66	22.73 a	19.75	4.38	7.79
25	50	69.87 ab	21.92	21.58 <sup>ab</sup>	19.27	4.23	7.74
50	00	65.82 <sup>b</sup>	21.32	20.92 b	18.74	4.41	7.67
SMS (Perce	nt)						
(	)	65.77 <sup>b</sup>	20.66	20.59 b	18.41	4.45	7.57
0.	.5	70.84 a	22.62	21.97 ab	19.51	4.21	7.79
1.	.0	7.71 a	22.62	22.66 a	19.84	4.36	7.83
Probabilitie	S						
AFB <sub>1</sub>		0.05	Ns	0.05	Ns	Ns	Ns
SMS		0.01	Ns	0.05	Ns	Ns	Ns
AFB <sub>1</sub> ×SMS		0.05	Ns	0.05	Ns	Ns	Ns

Table 5. Effect of Aflatoxin B<sub>1</sub> (AFB<sub>1</sub>) and *Silybum marianum* seeds (SMS) on carcass component of broiler chickens (at day 35)

AFB<sub>1</sub>, Aflatoxin B<sub>1</sub>. SMS, *Silybum marianum* seeds. <sup>a-c</sup> Mean values within a column with no common superscript differ significantly from each other (P<0.05). Ns, not significant.

Bursal weight was significantly lower in AFB<sub>1</sub> group compared with the control, while no significant differences were observed in other groups. However, SMS supplemented feed restored the normal weight of bursa and liver completely. Spleen and heart weights did not significantly change by treatments (Table 6). The findings of the present study are supported by Tedesco *et al.* (2004), who reported reduced bursal weight in broilers by feeding aflatoxin contaminated feed. Kalorey *et al.* (2005) reported the protective role of SMS against aflatoxicosis on the weight of bursa of fabricius. Post-mortem lesions included pale, enlarged (swollen), yellow friable livers with pinpointed hemorrhages, swollen kidneys and atrophy of bursa and thymus in broiler suffering form aflatoxicosis. Heart showed hydropericardium, and intestines revealed hemorrhagic enteritis. These findings are conformed by those reported by Rahim *et al.* (1999). Gross pathological changes in the liver and other organs were in a milder degree in SMS fed birds as compared with the birds fed on contaminated feeds only and those raised on toxin binders.

Similar findings were recorded by Arshad *et al.* (1992) who conducted clinicopathological studies of experimentally induced aflatoxicosis in broiler chicks. Similar results were observed by Sabri *et al.* (1989) who studied the prevalence and pathology of mycontoxicosis in broiler chicks.

Treat	ment		1	nternal orga	ns weight (	as perce	nt of live b	ody we	eight)	
Aflatoxin (ppb)	SMS (Percent)	Liver	Spleen	Pancreas	Gizzard	Bursa	Peroven triculus	Heart	Full intestine	Abdominal fat
0	0	2.15 <sup>b</sup>	0.14	0.23 b	1.53 c	0.22 a	0.42 <sup>b</sup>	0.47	3.86 cd	1.92 bc
250	0	3.53 a	0.17	0.31 a	1.73 <sup>b</sup>	0.17 <sup>b</sup>	0.54 a	0.52	4.35 b	1.43 d
500	0	3.82 ª	0.21	0.32 a	1.82 a	0.15 <sup>b</sup>	0.56 a	0.53	4.54 a	0.95 e
0	0.5	2.16 <sup>b</sup>	0.13	0.23 b	1.43 d	0.24 a	0.41 <sup>b</sup>	0.48	3.82 cd	2.15 ab
250	0.5	2.41 <sup>b</sup>	0.16	0.22 b	1.45 d	0.23 a	0.42 <sup>b</sup>	0.48	3.74 d	1.84 bc
500	0.5	2.24 <sup>b</sup>	0.16	0.23 b	1.52 °	0.24 a	0.43 b	0.48	3.83 cd	1.72 cd
0	1.0	2.97 <sup>b</sup>	0.16	0.27 <sup>b</sup>	1.44 d	0.25 a	0.44 <sup>b</sup>	0.50	3.95 °	2.35 ª
250	1.0	2.32 ь	0.15	0.22 b	1.42 d	0.23 a	0.43 <sup>b</sup>	0.49	3.79 d	1.65 cd
500	1.0	2.24 <sup>b</sup>	0.14	0.26 b	1.53 °	0.21 a	0.45 <sup>b</sup>	0.50	3.83 cd	1.44 d
SEM		0.21	0.026	0.010	0.012	0.011	0.011	0.02	0.030	0.089
Main effect										
AFB <sub>1</sub> (ppb)										
u i	)	2.43 <sup>b</sup>	0.14	0.24 a	1.47 <sup>c</sup>	0.24 a	0.42 <sup>b</sup>	0.57	3.87 <sup>c</sup>	2.15 ª
25	50	2.75 a	0.16	0.25 ab	1.53 b	0.21 <sup>b</sup>	0.46 a	0.49	3.96 b	1.64 <sup>b</sup>
50	00	2.77 a	0.17	0.27 a	1.63 a	0.20 ь	0.48 a	0.50	4.07 a	1.37 °
SMS (Perce	nt)									
Ì (	) <sup>´</sup>	3.16 a	0.17	0.29 a	1.71 a	0.18 <sup>b</sup>	0.50 a	0.59	4.25 a	1.44 <sup>b</sup>
0.	5	2.27 °	0.15	0.23 b	1.47 <sup>b</sup>	0.23 a	042 b	0.48	3.79 b	0.90 c
1.	0	2.51 <sup>b</sup>	0.15	0.25 b	1.46 <sup>b</sup>	0.23 a	0.44 <sup>b</sup>	0.49	3.86 b	1.81 a
Probabilitie	es									
AFB <sub>1</sub>		0.05	Ns	0.05	0.01	0.01	0.01	Ns	0.01	0.01
SMS		0.01	Ns	0.01	0.01	0.05	0.01	Ns	0.05	0.05
AFB <sub>1</sub> ×SMS		0.01	Ns	0.01	0.01	0.01	0.01	Ns	0.01	0.01

Table 6. Effect of Aflatoxin B<sub>1</sub> (AFB<sub>1</sub>) and Silybum marianum seeds (SMS) on internal organs weight of broiler chickens (at day 35)

AFB<sub>1</sub>, Aflatoxin B<sub>1</sub>. SMS, *Silybum marianum* seeds. <sup>a-c</sup> Mean values within a column with no common superscript differ significantly from each other (P<0.05). Ns, not significant.

# Conclusions

The results of the present study suggest that treatment with silymarin phytosomes was helpful to reduce the toxicity of  $AFB_1$  on FI, BWG, internal organ weights and liver morphology in the broilers. The protective action of this herb was particularly evident on BWG and FI. These findings suggest that silymarin might be used in chickens to prevent the effects of  $AFB_1$  in contaminated feed.

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