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Biological Control of Aflatoxin on Egg Production Performance of Laying Hens

Olayinka Abosede Ojo[1*] and Omololu Olusanya[2]

[1]Department of Animal Production, Fisheries and Aquaculture, Kwara State University, Nigeria.
[2]Department of Animal Science, Animal Physiology Unit, University of Ibadan, Nigeria.

Authors' contributions

This work was carried out in collaboration between both authors. Author OAO designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author OO managed the analyses of the study and managed the literature searches. Both authors read and approved the final manuscript.

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ABSTRACT

The deleterious effects of aflatoxin have been well documented in the literature on livestock and animal products. Although toxin binders and some methods of mitigating aflatoxin has been proven to illicit a positive response, but the effectiveness of biocontrol method of mitigation on the laying performance, egg production and characteristics have not been fully documented. Therefore, effect of aflatoxin bio-control method (Aflasafe) on growth indices, egg production and characteristics of laying chicken (LC) were investigated in this study. 700 point-of-lay Bovan Nera (LC) were randomly allotted to four dietary treatments: Aflasafe maize-based diet (AMBD), Farm Feed with toxin binder (FF+toxin binder), aflatoxin-contaminated diet with toxin binder (ACDTB) and aflatoxin-contaminated diet without toxin binder (ACDWTB). The contaminated diets contained 306.3ppb aflatoxin and the experimental design was completely randomised with 4 treatments (n=175) and 5 replicates (n=35) per treatment for 14 weeks. Daily feed intake (DFI), Average egg weight (AEW), Average egg width (AEWd), Albumen length (AL) were determined using standard procedures. The DFI (g) of LC on ACDTB (113.54) and ACDWTB (115.13) was significantly lower than AMBD (124.66) and FF+toxin binder (129.06). The AMBD (1.45 g) significantly enhanced the AYD than

**Corresponding author: Email: ykmelodya1@yahoo.com;*

other treatments. Aflasafe maize-based diet enhanced, egg characteristics in laying chicken. It also improved egg quality in laying chickens without adverse effect on the production. The use of aflasafe maize grain in poultry diet is recommended.

Keywords: Aflatoxin-contaminated diet; aflasafe; egg characteristics; aflatoxicosis; laying chickens; mycotoxin; aflatoxin.

1. INTRODUCTION

Aflatoxin have been given a relatively extensive concentration as a result of the carcinogenicity and hepatotoxicity effects it poses to human and animal health. Among the known AF, aflatoxin B1 (AFB1) is most commonly encountered and considered the most toxic [1]. In animals, contrary effects of Aflatoxin also include depression in growth performance, egg production, hatchability, including increased susceptibility to diseases. Young animals are easily affected by aflatoxin than older ones. Although, there is significant difference in specie susceptibility, the relative importance of the response is influenced by the sex, age weight, diet, exposure to infectious agents and the presence of other mycotoxins and pharmacological active substances [2]. Aflatoxin causes a variety of symptoms depending on the animal, dose, length of exposure, species, breed and diet or nutritional status [3]. In layers recently, the influence of aflatoxicosis on reproductive performance of poultry breeding stock has become a question of economic concern. Aflatoxin affects layers and result into depressed egg production, poor egg quality and increased mortality of exposed layers. It negatively influences egg quality by decreasing shell thickness, egg weight, egg energy and cessation of egg production [4]. The negative touch of aflatoxin on layers can be induced when feed contains 1-2 mg/kg tottal AFB [5,6]. In addition, aflatoxin in layers feed can give rise to an aflatoxin residue in the eggs (feed to egg aflatoxin transmission ratio was approximately 5000:1) it is consequently very important to reduce minimally or eliminate aflatoxin concentrations in feeds for layers [7-9] observed lowered egg production and hatchability in Japanese quail fed with aflatoxin-contaminated diet. Although a lot of research has been done to evaluate the possible solution to the aflatoxicosis challenge in poultry, this involve the use of binders, and mechanical control methods, but these however, has their limitations. Aflasafe is produced from a novel bio-control method involving the transfer of toxigenic strains of *Aspergillus flavus* on agricultural fields with atoxigenic strains. This works by interference with the contamination process by both physically excluding the toxigenic strain during infection and by competing for nutrients required for aflatoxin biosynthesis by the toxigenic strains, thereby reducing the overall toxigenicity of *A. flavus* population. This research was therefore designed to investigate the efficacy of Aflasafe ^{TM} as a biological control of aflatoxin on the egg production performance of layers.

2. MATERIALS AND METHODS

This study was carried out at God's Grace Farm, located in Lagun Town, along Ibadan-Iwo road, Oyo State, while the aflatoxin-contaminated maize grain and aflasafe maize grains used for this experiment were obtained from the plant pathology unit, International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria. Other ingredients used for the feeds formulated were purchased from God's Grace feed mill located at Lagun Town, along Ibadan-Iwo road, where the study was conducted. Maize grain which was used as the aflatoxin carrier was inoculated with toxigenic strain of *Aspergillus flavus* of Nigerian origin. The culturing and inoculation was done at the plant pathology unit, International Institute of Tropical Agriculture, Nigeria using 5% V8 juice and 2% agar, having a PH of 5.2 and a spore load of 2.475×10^6 per ml. The total aflatoxins were quantified to be 306.3ppb aflatoxin using scanning densitometer, CAMAG TLC scanner 3 with –CATS 1,4,2 software (Camag AG, Muttenz, Switzerland) [10].

2.1 Experimental Birds and Management

The study was carried out using a total of 700 30-week old Bovan Nera black chickens with a mean body weight of 2±0.35kg. There were four (4) treatments with five replicates per treatment. Chickens were randomly allotted into groups with 175 birds per treatment and 35 birds/replicate. Experiment lasted for 14 weeks. The chickens were housed in battery cages having linear feed troughs and nipple drinkers for running water. All the birds were fed basal diets for 2 weeks. Provision of experimental diets and fresh

water were done *ad libitum*. At the end of the acclimatization period, the chickens were weighed and randomly allotted into four treatments with 175 layers per treatment (5 replicates with 35 birds per replicate) in a completely randomized design. The experiment lasted for 14 weeks. Data collection was done weekly throughout the duration of the study.

2.2 Experimental Diets

Four (4) experimental diets were formulated based on the nutrient requirement of the chickens. The experimental diet is presented in Table 1. The diet was subjected to chemical analyses to obtain its proximate composition. The diets were as outlined below;

Treatment 1 (T1) – Aflasafe maize-based diet
Treatment 2 (T2) – Farm feed with toxin binders (Control diet)
Treatment 3 (T3) – Aflatoxin contaminated diet with toxin binder
Treatment 4 (T4) – Aflatoxin- contaminated diet without toxin binder.

Each group was provided with a known weight of diet daily. Daily feed intake was obtained by deducting the leftover feed and waste from the total feed supplied per day. The average weekly feed consumption/bird was calculated by dividing the group feed intake by the number of chickens present in the replicate per week. The feed conversion ratio was defined as the amount of feed required for producing a given kg of flesh or a given kg of egg. It was calculated as presented below;

$$\text{Feed conversion ratio (FCR)} = \frac{\text{Feed intake (kg)}}{\text{Weight gain (kg)}}$$

$$\text{Feed efficiency ratio (FER)} = \frac{\text{Weight gain (kg)}}{\text{Feed intake (kg)}}$$

The hen-day production (HDP) is the percentage lay on daily basis. This was calculated using the formula presented below [11].

$$\text{HDP (\%)} = \frac{\text{Total eggs laid on daily basis}}{\text{Number of hens available in the flock on that day}} \text{X } 100$$

The percentage egg production in this study was estimated using the formula below:

$$\text{Egg production \%} = \frac{\text{Total number of eggs produced in one treatment}}{\text{Total number of eggs produced in the experiment}} \text{X } 100$$

The egg production capacity of each hen was recorded as the total sum of eggs laid per bird for a period of one week. This record was taken weekly throughout the experimental period as presented below:

Egg production capacity/hen = Number of egg produced per hen on weekly basis

The egg quality characteristics measured were the external qualities which include; egg weight (g), egg length (mm), egg width (mm), shell weight (g), shell thickness (mm) and internal qualities such as albumen weight (g), albumen width (mm), albumen height (mm), yolk height, yolk weight, yolk width (Anjum, 2013).The weight of each egg was taken using electronic digital balance with a range from 0.01 to 200 g (Kern Model No. 440-33N, Germany), while length and width of the egg was measured with sensitive electronic vernier caliper. The shell weight was measured using sensitive measuring scale, while the shell thickness was measured using the micrometer screw gauge. Measurements of the internal components were obtained by carefully making an opening around the sharp end of the egg, large enough to allow passage of both the albumen and the yolk through it without mixing their contents together. The yolk was then carefully separated from the albumen and placed in a petri dish for weighing, while the albumen was placed in another petri dish and weighed. Values obtained were recorded in grammes (g) as stated below;

Weight of albumin/ yolk (Wt_2) = Wt –Wt_1

Where;

Wt_1 = Weight of petri dish
Wt_2 = Albumin / yolk weight
Wt = Weight of both petri dish + internal egg component (albumin / yolk)

The yolk diameter and albumin height of the egg were measured with electronic caliper. The shell weight with membrane was obtained by carefully replacing the opened part in the shell and weighing on the electronic scale. The thickness (mm) of the shell with intact membranes was measured at three different points and the average of the broad, sharp and middle part of the egg was obtained by using micrometer screw guage.

Table 1. Composition of the experimental diet (%) (Layers mash)

Ingredients (kg)	AMBD	FF+Toxin Binder	ACDTB	ACDWTB
Aflasafe Maize	50.220	-	-	-
Farm Maize	-	50.220	-	-
Contaminated Maize	-	-	-	-
Soyabean Meal	23.100	23.100	23.100	23.100
Wheat offal	13.460	13.460	13.460	13.460
Salt	0.350	0.350	0.350	0.350
Bone Meal	3.010	3.010	3.010	3.010
Oyster Shell	9.040	9.040	9.040	9.040
Lysine	0.200	0.200	0.200	0.200
Methionine	0.200	0.200	0.200	0.200
Vitamin C	0.006	0.006	0.006	0.006
Layer Premix	0.300	0.300	0.300	0.300
Toxin Binder	-	0.100	0.100	-
Oxytetracycline	0.100	0.100	0.100	0.100
Total (kg)	100.00	100.00	100.00	100.0
Calculated nutrients				
Crude Protein (%)	17.30	17.30	17.30	17.30
Crude Fibre (%)	4.56	4.56	4.56	4.56
Metabolizable Energy(Kal/kg)	2598.25	2598.25	2598.25	2598.25
Phoshorus	0.68	0.68	0.68	0.68
Lysine	1.05	1.05	1.05	1.05
Methionine	0.46	0.46	0.46	0.46
Calcium	4.34	4.34	4.34	4.34
Analysed nutrients				
Met energy(kcal/kg)	2874.38	2926.22	3048.16	3052.60
Crude protein	20.50	18.78	18.22	18.01
Ash	18.50	16.50	13.50	13.00
Crude fibre	7.05	7.16	7.21	7.25
Dry matter	90.49	90.09	90.40	90.29

Vitamin A - 10,000 IU, Vitamin D_3-2,000 IU, vitamin E-100 IU, Vitamin K-20 mg, thiamine B_1- 15 mg; Riboflavin B_2-40 mg,, Vitamin B_6 (pyridoxine) - 15 mg, Niacin – 150 mg; Vitamin B_{12}-0.015mg, Pantothenic acid-50 mg, Folic acid-5 mg, Biotin - 0.2 mg. Choline chloride-12mg; , Antioxidants- 1.25 g; Mn,-0.8 g; Zinc,-0.5 g; Iron - 0.2 g; copper-0.05g; iodine- 0.12 g; Selenium – 2 mg; cobalt – 2 mg. AMBD = Aflasafe maize-based diet, FF+Toxin binder = Farm feed + Toxin binder, ACDTB = Aflatoxin-contaminated diet with toxin binder and ACDWTB = Aflatoxin-contaminated diet without toxin binder

2.3 Statistical Analysis

All data obtained were subjected to descriptive statistics and one-way analysis of variance (ANOVA) in a completely randomized design using statistical analysis software (SAS, 2008). Means were separated using Duncan multiple range test [12].

3. RESULTS AND DISCUSSION

3.1 Effect of the Experimental Diets on Growth Performance of Layers

The results of the growth performance of laying hens fed experimental diets are shown in Table 2. Among the performance parameters assessed, only those of the initial body weight and mortality were not significantly different (P=0.05) across the treatment groups. The final live weight, daily feed intake, feed conversion ratio and daily weight gain were significantly influenced by the dietary treatment. It was observed that the final live weights of layers fed AMBD (T1) and FF+Toxin binder (T2) were significantly higher than those of the layers fed ACDTB (T3) and ACDWTB (T4). The highest value (1728.57±7.42 g) was recorded in layers fed on AMBD, while the least value was obtained in layers fed ACDTB (1545.71±6.30 g). The mean values obtained for daily feed intake was also significantly (P=0.05) higher for AMBD and FF+Toxin binder-fed hens compared to that of ACDTB and ACDWTB layers. Although, the

highest value (129.06± 8.75 g) was obtained in layers fed FF+Toxin binder diet, while the least was recorded in hens fed ACDTB diet (113.54±8.26 g). The feed conversion ratio of laying chickens fed Aflasafe maize-based diet (1.03±0.33) was significantly (P<0.05) lower while the value obtained for birds fed aflatoxin-contaminated diet without toxin binder (1.92±0.45) was significantly (P=0.05) higher compared to the control value. The weight gain of layers fed Aflasafe maize-based diet was observed to be significantly (P<0.05) higher compared to the birds fed the control diet (FF+Toxin binder), ACDTB and ACDWTB respectively.

Birds given AMBD had the highest weight gain among all the treatments. This may be due to the absence of *A. flavus* and *A. parasiticus* in the maize grain used to compound the diet which enhanced normal digestion, protein synthesis, proper nutrient absorption and utilization by the layers. A decrease in feed intake, growth rate, poor feed utilization, reduced egg production and increased liver fat levels have been reported as the main manifestations of chronic aflatoxicosis in layers [13,14]. The mean daily feed intake of layers fed the AMBD was observed to compare favorably with that of the farm feed (FF) and both were significantly higher than the feed intake of layers fed ACDTB and ACDTWB respectively. This could be as a result of the toxin effect ingested by the group of layers. This result is in agreement with the findings of Denli [14] who observed reduced feed intake in layers fed aflatoxin contaminated diets containing 1 mg/kg aflatoxin. The use of Aflasafe maize-based diet significantly offset this effect reaching values not significantly different from the control diet (FF+toxin binder). The presence of high concentration of aflatoxin in the aflatoxin-contaminated diet might have resulted into low feed quality and anorexia, which perhaps also resulted into the recorded lower feed intake as toxin level increased within the animal body. According to Pandey and Chauhan [15] who reported that the feeding of aflatoxin B1 at various dose to layers from 1st week to 40 weeks of age affected the feed consumption, as result of the reduction in the palatability of the feed, also caused depressed weight gain, reduction in egg production and egg weight at 3.91 mg/kg level and caused 11-47% dose-dependent mortality. The FCR value of layers fed AMBD showed a significant reduction compared to that of FF+Toxin binder. A lower FCR is an indication of high quality feed and it enables farmer to maximize profit. A lowered FCR value signifies a higher level of protein in the diet. This means it takes less feed to produce 1 kg of meat. Birds fed ACDTWB had the highest FCR value. This showed that aflatoxin contamination negatively influences the integrity of intestinal mucosa which perhaps affected selective reabsorption of nutrients in the birds. As stated by Aravind et al. [16], aflatoxin has been known to influence the metabolism of poultry, reducing the activity of enzyme that digest starch, protein, lipids, and also decreases blood protein. Contrary to these findings, Siloto et al. [17] did not observe any significant changes in the FCR of birds in his study. Birds fed ACDTB had the lowest weight gain. This might be because of the adverse effect of aflatoxins on the overall metabolism, digestion and protein synthesis, which causes DNA binding and inhibition of nucleic acid synthesis. It also shows that the binder added to this diet was not able to effectively bind the aflatoxin. This result is in agreement with the observation of Choi et al. [18] who observed decreased growth rate in birds fed 1 ppm aflatoxin-contaminated diet. Hamilton et al. [19] and Pearson et al. [20] reported significant decline in body weights of laying birds when fed 2.5 ppm aflatoxin. The % mortality of birds was not significantly influenced by the dietary treatment. This is in agreement with Rosmaninho et al. [13] and Aravind et al. [16] who noticed significant decrease in the body weight of birds fed naturally-contaminated diet containing aflatoxin. Although the % mortality of the birds was not significantly influenced by the experimental dietary treatments, the weekly mortality record which was significantly highest at the 10^{th} and 13^{th} week in birds fed ACDTB and ACDWTB could be due to dietary aflatoxin. This could have resulted in immunosuppression linked with increased mortality [21].

3.2 Effect of Experimental Diets on Egg Production Performance

The result of the egg production performance of laying hens fed experimental diets (Table 4) showed that the average egg weight, feed efficiency and feed intake were significantly affected (P=0.05) by the dietary treatment. The average egg number recorded was not significantly influenced. It was observed that the average egg weight of birds fed the ACDWTB (53.25 g) was significantly (P=0.05) lower than the egg weight of birds fed the FF+toxin binder (56.34 g). The average egg weight values of birds fed the AMBD and that of the ACDTB (56.34 g and 57.03 g) were not significantly

(P<0.05) altered compared to the control (FF+toxin binder) (56.36 g). The feed efficiency ratio (FER) of layers in terms of egg weight per feed intake was observed to be significantly influenced across the treatments. FER of layers fed AMBD (31.76±1.69 g) and those fed the control (FF+toxin binder) (31.59) were not significantly different (P=0.05). However, the feed efficiency ratio obtained in birds fed the ACDTB (25.34) and ACDWTB (22.6) were observed to be significantly depressed compared to the control. The average egg number of layers recorded per week was observed to range from 3.42 to 5.05 with the highest number obtained in birds fed the FF+toxin binder feed and the lowest value observed in birds fed the ACDWTB.

The egg production parameters evaluated were significantly influenced by the dietary treatment except that of the Average egg number. Addition of toxin binder to the farm feed incurred extra cost of feed formulation and the overall cost of production is also increased. In Aflasafe maize-based diet, the exclusion capability of the atoxigenic strain of *A. flavus* and *A. parasiticus* on the toxigenic strain enhanced a toxin free maize grains which do not need additional cost to procure toxin binder. Also the addition of Aflasafe to the field can still be effective for several years. Therefore, the use of Aflasafe maize-based diet reduces cost of production and help in maximizing the profit of farmers. The feed efficiency ratio in terms of egg production was significantly influenced by the dietary treatment. The FER value of birds in AMBD was the highest. This might be because of use of aflatoxin-free diet, which enhanced the bird's efficiency in converting the feed mass into desired output (i.e eggs). The FER value of birds in ACDWTB was the lowest. This might be as a result of aflatoxin influence of the poor feed quality and aflatoxin effect on the gastro intestinal tract (GIT) of the layers which adversely affected the required nutrient absorption which could have been channeled towards egg production and the subsequent influence on the reproductive system of the group of birds fed the diet which resulted into poor performance of the birds. The average egg number was not significantly influenced by the treatment. The hen day productions of the commercial layers fed dietary treatments were influenced in all the treatments. It could be observed that birds fed AMBD and FF+toxin binder had better hen-day production than birds on ACDTB and ACDWTB respectively. This might be probably because of the quality of diets with Aflasafe and that of farm feed without contamination. This result agrees with Iqbal et al. [22] and Verma et al. [23] who reported that feeding varying levels of aflatoxin to white leghorn layers resulted in a significant reduction in hen day egg production.

3.3 Effect of Aflatoxin on Egg Characteristics of Laying Chickens

The results on egg characteristics of layers fed the dietary treatments are shown in Table 3. Among the egg indices assessed, average albumen length and average shell thickness were not significantly influenced by the dietary treatments. The average egg length, average egg width, average yolk length, average yolk depth, average yolk width and average albumen width were significantly (P=0.05) affected by the dietary treatment. It was observed that the average of birds fed AMBD, FF+toxin binder and ACDTB were significantly (P=0.05) higher than that of birds fed ACDWTB. This same trend was recorded in the mean values of egg length, egg width and yolk length among the treatments. The average yolk depth (1.45 cm) of birds fed the AMBD was observed to be significantly (P<0.05) higher than that of birds fed FF+toxin binder (1.32 cm). Also, the average yolk depth of layers fed the ACDTB and ACDWTB depth (1.40 cm and 1.32 cm) were not significantly different from the layers fed the control diet (1.36 cm). The average yolk width observed for the layers fed AMBD, ACDTB and ACDWTB were not significantly altered compared to the control value. This same trend was observed in the average albumen width of the experimental birds. The average shell thicknesses recorded were not significantly influenced across the treatments.

The average egg weight of layers fed the Aflasafe maize-based diet and that of ACDTB were not significantly influenced as compared to the control [FF + toxin binder]. However, the average egg weight value of birds in (ACDTB) was significantly lower compared to the FF+toxin binder. The average egg weight of birds fed the Aflasafe maize-based diet compared favorably with that of the farm feed with toxin binder, depressed egg weight of layers was observed in ACDWTB which might be due to influence of the toxin levels in the feed which affected the feed efficiency and reproductive performance of the birds in accordance with Del Bianchi et al. [24]

Table 2. Growth performance of laying hens fed experimental diets

Parameters	AMBD	FF+Toxin binder	ACDTB	ACDWTB	SEM
Initial body wt (g/bird)	1650.00	1620.00	1590.00	1630.00	20.00
Final Live weight (g/bird)	1728.57 [a]	1680.00[a]	1545.71[b]	1552.86[b]	35.13
Feed intake (g/bird/day)	124.66[a]	129.06[a]	113.54[b]	115.14[b]	13.76
FCR	1.03[c]	1.84[b]	1.89[b]	1.92[a]	2.01
Feed Efficiency ratio	97.44[a]	54.91[b]	53.46[b]	49.61[b]	26.65
Weight gain (g/bird)	120.00[a]	70.00[b]	60.00[b]	60.00[b]	0.03
Mortality (%)	7.43	4.00	4.00	5.14	0.34

a-b-c :means within each row with different superscripts are significantly different (P<0.05).. FCR = Feed conversion ratio

Table 3. Egg production performance of laying hens fed the experimental diets

Parameters	Treatments				
	AMBD	FF+Toxin binder	ACDTB	ACDWTB	SEM
Average Egg Weight (g)	56.34[a]	56.35[a]	57.03[a]	53.28[b]	0.73
Feed intake (g/bird/day)	124.66	129.06	113.54	115.54	13.76
Feed efficiency (FE)	31.76[a]	31.59[a]	25.34[b]	22.63[c]	0.81
Average Egg Number/bird/Wk)	4.92	5.05	3.53	3.42	0.008

=Aflasafe maize-based diet, FF+Toxin binder = Farm feed + toxin binder, FE = Egg weight / Feed intake

Table 4. Egg characteristics of layers fed the dietary treatment

Parameters	AMBD	FF+Toxin binder	ACDTB	ACDWTB	SEM
Average Egg Weight (g)	56.34[a]	56.36 [a]	57.03 [a]	53.25 [b]	0.73
Average Egg Length (cm)	5.54 [a]	5.55 [a]	5.64 [a]	5.28 [b]	0.08
Average Egg Width (cm)	4.28 [a]	4.29 [a]	4.30 [a]	4.06 [b]	0.05
Average Yolk Length (cm)	4.18 [a]	4.20 [a]	4.23 [a]	4.00 [b]	0.06
Average Yolk Depth (cm)	1.45 [a]	1.36 [a]	1.40 [ab]	1.32 [c]	0.02
Average Yolk Width (cm)	16.42 [a]	16.11 [ab]	16.51 [a]	15.42 [b]	0.24
Average Albumen Length (cm)	9.33	9.61	9.65	9.28	0.15
Average Albumen Width (cm)	17.92 [a]	17.40 [ab]	18.13 [a]	17.02 [a]	0.28
Average Shell thickness (cm)	0.4	0.4	0.36	0.35	0.004

abc: means along the same row with different superscripts are significantly (p<0.05) different

This is also in agreement with the findings of Rizzi et al. [25] Zaghini et al. [26], Pandey and Chauhan [27] but disagrees with the result of Oliviera et al. [28] who observed no significant (P=0.05) difference in the egg weight of layers fed dietary treatments of 25, 50 and 100 ppb aflatoxin. Therefore, the use of Aflasafe maize-based diet may favour production and help in maximizing the profit of farmers. The depressed egg weight of layers in treatment 4 (ACDWTB) might be due to influence of aflatoxin levels in the feed and which affected the feed efficiency and reproductive performance of the birds. The result obtained in this study agreed with Rizzi et al. [25] who observed decreased egg weight in birds fed high aflatoxin level. However, this result disagrees with the findings of Oliviera et al. [29] who observed that egg weight of birds fed 25-100ppb were not significantly affected. The same trend was noticed in the values of average egg length, average egg width, average yolk length, and width and average albumen width. The average yolk length of layers in treatments 1 (AMBD) and 3 (ACDTB) were not significantly different compared to the control. However, the average yolk length of birds in treatment 4 was significantly lower compared to the control. Similar trend was observed in the average yolk width of the layers. According to Huff et al. [30] and Vieira [31], the aflatoxin contaminated diet caused reduction in yolk size and this was due to interference of aflatoxin with the liver metabolism of protein and lipids which are the main egg components. Fat deposition occurs in the liver

impairing lipid mobilization to the ovarian follicles and consequently resulting in eggs with small yolk size. Average yolk depth of birds in treatment 1 (AMBD) was significantly higher compared to the control (FF+toxin binder), the average yolk depth of birds in treatment 3 (ACDTB) was numerically higher, while the value in treatment 4 (ACDWTB) was numerically lower compared to those fed (FF+toxin binder). Washburn et al. [32] observed reduced yolk weight produced by layers fed diets containing 5mg/kg of Aflatoxin B_1. Average yolk depth of layers observed (AMBD) which was significantly higher compared to the control might be because the diet did not contain aflatoxin contamination which enhanced maximum transport of lipid to ovarian follicles from the liver. The internal quality of eggs is an important raw material in egg producing industry because of high demand for liquid egg, frozen egg, egg powder and yolk oil. Average albumen width values of birds in ACDTB are higher, while that of ACDWTB are lower compared to the AMBD. This might be as a result of aflatoxin level inclusion in the diets of birds in ACDWTB, while the binder effect in ACDTB may have been depleted or the adverse effect of aflatoxin on the liver which enhanced an increased average albumen weight. The average shell thickness of layers in AMBD was not significantly different from that of treatment 2 (FF+toxin binder), however, the values of layers in ACDTB and ACDWTB were significantly lower compared to these fed FF+toxin binder. This agree with the observation of Zaghini et al. [26] who reported reduced egg shell weight associated with feeding of 2.5 mg/kg of aflatoxin. However, the result of this work disagrees with the observation of Siloto et al. [17] and Galkate and Rokde [33], who observed higher egg shell thickness of 0.5 – 2.0 mg /kg of layers fed aflatoxin contaminated diet. It has been documented that feeding of aflatoxin causes poor calcium and phosphorus absorption and interference with Vitamin D3 metabolism which result in poor shell quality at higher levels of aflatoxins [34-36].

3.4 Effect of Experimental Diet on HDP of Laying Chickens

The Hen Day Production (HDP) of layers which received the AMBD and FF+ Toxin binder were recorded to be significantly higher than those layers given ACDTB and ACDWTB.

The hen day productions of the commercial layers fed dietary treatments were influenced in all the treatments. It could be observed that birds fed AMBD (T1) and FF+toxin binder (T2) had better hen-day production than birds on ACDTB (T3) and ACDWTB (T4) respectively. This might be probably because of the quality of diets with Aflasafe and that of farm feed without

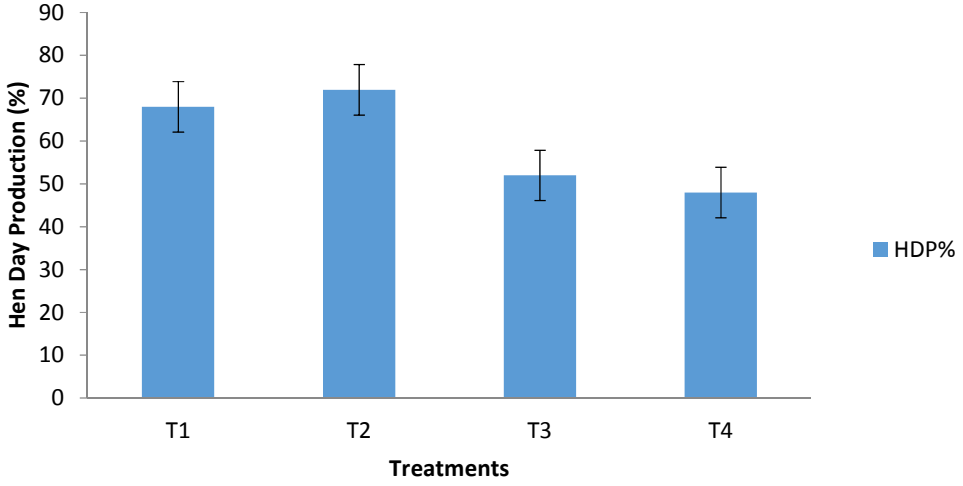


Fig. 1. Hen day production of laying hens fed experimental diets

contamination. This result agree with Iqbal et al. [22] and Verma et al. [23] who reported that feeding varying levels of AF to white leghorn laying chickens resulted in a significant reduction in hen day egg production.

4. CONCLUSION

Adverse effects of aflatoxin in layers can be prevented by the use of feed ingredients that contains no aflatoxin contamination through the mutual exclusion mechanism of *Aspergillus species* in the soil. Although aflatoxin binder is effective, but its cost implication could increase the overall production cost, especially on a large scale livestock farming. The use of biological means of mitigating aflatoxin (aflasafe-maize grain) in poultry diet is therefore recommended.

CONSENT

It is not applicable.

ETHICAL APPROVAL

As per international standard or university standard written ethical permission has been collected and preserved by the author(s).

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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