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Testing the Efficiency of Probiotic in Reducing the Growth of *Aspergillus flavus* in Corn Stored and its Effect on Performance of Broiler.

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ABSTRACT

This study has been done for testing the effectiveness and efficiency of probiotic in its ability to inhibit *Aspergillus flavus*, which secretes the aflatoxin B1 and compare it with materials that were approved to be effective in the inhibition of the growth of fungi and break down and reduction of mycotoxin. These are the filax substance and ozone gas. A laboratory experiment has been conducted to determine the isolates out of the fungus producing the aflatoxin B1. The isolate producing aflatoxin B1 has grown on the yellow corn seed. The probiotic was added along with the filax and they have been exposed to ozone gas and then they were stored from 30 days. A bio test was conducted to know the effects of biological toxin on the broiler meat. This study was conducted on 800 broiler chicks (Ross 308), of one day age, that have been weighed on 8 treatment (4 replicate per treatment and 25 chicks per replicate). The first treatment was (T1) which is the control diet that contains yellow corn not contaminated with aflatoxin B1, the second treatment is (T2) which is the diet that contains yellow corn contaminated with aflatoxin, while the third treatment (T3) it is used the yellow corn which is contaminated with toxin treated with ozone gas. The fourth treatment (T4) it is used the yellow corn which is not contaminated with toxin treated with ozone gas. In the fifth treatment (T5) a toxin contaminated yellow corn was used which is treated with the Iraqi probiotic. The sixth treatment (T6) the yellow corn not contaminated with toxin and treated with Iraqi probiotic was used. The seventh treatment (T7) the yellow corn was contaminated with the aflatoxin and treated with filax and the eighth treatment (T8) the yellow corn not contaminated with toxin was used but it was treated with filax. The results of the study indicated that the addition of ozone gas, the filax and the probiotic to the yellow corn contaminated with toxin led to a significant reduction in toxin amount in comparison with control treatment, which were contaminated with toxin without adding any other substances to them. A significant higher preponderance ($P<0.05$) and a clear improvement in productive performance of the birds that were fed with diets containing yellow corn and treated with the probiotic. Such improvement was clear also in the treatment to which the ozone gas and the filax were added. The study showed a significant deterioration ($p<0.05$) in productive characteristics of the birds in which yellow corn, contaminated with aflatoxin B1 was used from this study we conclude there is possibility to add the Iraqi probiotic to the diets of broilers meat that contain aflatoxin B1 to reduce the contamination of diets with toxin, in addition to the efficiency and obvious effectiveness of the filax and ozone gas in eliminating and destroying the toxin.

Keywords: Aflatoxin B1, Iraq Probiotic, Filax, Ozone Gas, Productive Performance, Broiler.

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INTRODUCTION

Most of the studies in the field of poultry breeding to the use of modern scientific methods in poultry feed manufacturing and began to concern the components of the bush, and they are free of contaminants and in particular fungus and outputs metabolic secondary of mycotoxins Mycotoxins, which is one of the main factors for the decline in production (Devegowda et al., 2005). As the poultry diets agricultural nstitute 50-60% of poultry diets (Shareef, 2010). As maize infected with a wide range of fungi producing toxins in the field before harvest ancircles it is suitable for the growth of fungi, especially maize, which cod during storage and which pose a threat to the health of humans and animals as a result of eating contaminated products to these toxins, there are many fungi that produce mycotoxins, but the most prevalent in the silo and used sex as food for poultry is Aspergillus flavas as found Hussain (2000), Mals (2004) that sex is more races A.flavas presence on the yellow corn used for feed grains as materials in Iraq and that the toxins that have been discovered from the production of this fungus aflatoxin, a poison which is the most dangerous toxins fungal on human and animal health even at the low level of pollution as well as developed by the mushroom product has on the growth and proliferation in over Thermal widely as found Hiti, (1977) that 76% of the isolates fungus A.flavas accompanying holds maize stored was producing aflatoxin B1, and was 54% of the corn stored samples contaminated with aflatoxin B1. And found Shareef (2007) that all poultry diets examined 185 samples were contaminated with poisons aflatoxin higher than the allowable level of 20 ppb. And aflatoxin contamination diets influence the performance of productive poultry to note Tiwari et al (1989) reduced weight gain and reduced immune rate of chicks fed diets contaminated with aflatoxin, compared with the control treatment. The modern strategies seek to reduce or eliminate the negative excitement of aflatoxin from poultry diets using the methods and methods of chemical, physical and herbal biogenic (Diaz et al, 2002, Munafi, 2011). So study aimed to conduct a laboratory experiment, and Khoznah and field to test the efficiency of probiotic Iraqi in reducing the growth of fungus A. flavas Alvarez poison aflatoxin B1 in maize, stored grain and compared with materials proved its efficiency and effectiveness in the inhibition of fungi, breaking down or reduction of mycotoxins a substance Alfablex and ozone gas and over the effect on the productive performance of broiler chickens.

MATERIALS AND METHODS

The Experiment Lab

Detection of aflatoxin b1 using thin layer chromatography (TLC) and quantification of AFBI using high performance liquid chromatography (HPLC).Isolate obtained from the laboratory of mycotoxin / college of agriculture , university of Baghdad , after multiplication at rice media and extraction of aflatoxin B1 by following reference method (AOAC , 2005). Toxins was qualitative detected by using TLC technology and matching deportation coefficient RF and notice the color of brilliance and its intensity and compared it with the standard of AFBI (sobolev and dorner ,2002 ; cocker et al ,1984). While quantity of AFBI evaluate in HPLC by using separation-column 18 reverse phase (4.5 mmid)ODS and the mobile phase consisting of 180 ml Aceton Enzal and 820 ml of distilled water and 10 ml acetic acid snow mixed in mixer before use and disposed of gas couss bubbles in mixture , use solvent injection by blending 100 ml of acetone enzal and 900 ml of distilled water and 10 ml acetic acids snow.In order to test efficiency of the filax and Iraqi probiotic in reducing *Aspergillus flavus* , three concentration of filax 0.1 ,0.2 and 0.3 ml were added. It is a commercial liquid produced by dutch selko company. It contains anumber of organic acid. These concentrations , mentioned above ,were added to 100 ml of PDA which is sterilized and cooled by use of food poisoning (dixit et al ,1916). The results led to the calculation of the average of diagonal perpendicular from each colony , after filling the comparison colony per class. The inhibition percentage was calculated by use of the following equation :

$$\text{The inhibition percentage} = \frac{C-T}{C} * 100$$

The average of control colony = c

The average of treatment colony = T

In the same way , the concentrations 0.03 , 0.03 , 0.1 , 0.2 and 0.5 have been done with a probiotic to each 100 ml PDA , 15-20 ml was added to a petri dish and the dishes that the isolate was grown on them and who are toxin productive were exposed to 3 and 4 kg/minute of ozone by 10,20 and 30 mintes for each concentrate with the average of three duplicate for each time and left treated without treatment for comparison. Each dish was wrapped with a 0.5 cm of diameter disk close to the edges fungi A.flavas colony, 6

days of age. Each treatment was repeated three times , then the dishes were incubated at 25 ± 2 C° for seven days. The isolate that produces the aflatoxin B1 was grown on the seeds of yellow corn in tightly closed glass bowls by 500 gram per glass bowl with an average of 3 replicates for each treatment, 2 gram of rice powder , which the toxin productive isolate is feeding on , were added , leaving it with the seeds of yellow corn for 5 days with daily mixing with 5 ml of water. The humidity of yellow corn was measured by the electronic hygrometer. The humidity rate was 19.5 %.the filax was sprayed with 0.2 % concentrate (mix with distilled water) and with 0.2 probiotic . the seed were exposed to ozone gas with 2 kg /minute concentrate for ten minute. The treatment were as follows: (T1) yellow corn+sterilized distilled water : control , (T2) toxins contaminated-only yellow corn : control , (T3) yellow corn contaminated with toxin and treated with filax , (T4) yellow corn not contaminated with toxin and treated with filax ,(T5) yellow corn contaminated with toxin and treated with Iraqi probiotic,(T6) yellow corn not contaminated with toxin and treated with Iraqi probiotic,(T7) yellow corn contaminated with toxin and treated with ozone gas , (T8) yellow corn not contaminated with toxin and treated with ozone gas. The treatments were mixed after 30 days by the electric blender and the opertions of excretion and purification were made on them , then the last extract was taken from purification and injected with 5 microliter in HPLC. The toxin concentration was calculated with the following equation according to (scudamer and patel ,2000).

$$\frac{\text{standaed toxin concentration}}{\text{sample}} = \frac{\text{standard toxin curve space}}{\text{sample curve space}}$$

The rate of reduction was calculated with the use of the following equation:

$$\text{Rate of reduction} = \frac{\text{concentration of control treatment}-\text{sample concentration}}{\text{concentration of control treatment}} * 100$$

The Experiment Field

In this experiment 800 chicks (ross308) were used of one day old with 46 gram the weight of each. The chicks were bred in a closed hall consisted on 32 box. The chicks were spread randomly to the box by 25 birds per box. The birds were fed with three diets according to the table 1. The treatments of the experiment were as follows : The first treatment was (T1) which is the control diet that contains yellow corn not contaminated with aflatoxin B1 , the second treatment is (T2) which is the diet that contains yellow corn contaminated with aflatoxin, while the third treatment (T3) it is used the yellow corn which is contaminated with toxin treated with ozone gas. The fourth treatment (T4) it is used the yellow corn which is not contaminated with toxin treated with ozone gas. In the fifth treatment (T5) a toxin contaminated yellow corn was used which is treated with the Iraqi probiotic. The sixth treatment (T6) the yellow corn not contaminated with toxin and treated with Iraqi probiotic was used. the seventh treatment (T7) the yellow corn was contaminated with the aflatoxin and treated with filax and the eighth treatment (T8) the yellow corn not contaminated with toxin was used but it was treated with filax was used. The productive performance were measured. They included average of body weight , weight gain , feed intake and feed conversion ratio during the period of the experiment (1-3) (4-6) weeks of age and the whole period of (1-6) weeks , as well as dressing percentage and relative weight of the carcass.

Statistical analysis

Experiment data were analysed with the used of complete random design CRD by use of statistical ready software (SAS,2007).intangible differences between the averages were compared using a polynomial Duncan test (Duncan , 1955).

Table 1. Percentage composition of the experimental diets

Ingredients %	Types of diets		
	Starter 1-10d.	Grower11-22d.	Finisher23-42d.
Yellow Corn	47.5	50.85	51.8
Wheat	10	10	10
Soybean Meal (48.5% CP) ¹	32	28	29.8

Meat Meal	2	5	5	2.5
Hydrogenated Vegetable Fat		3	4.15	4.9
Dicalcium Phosphate		0.7	0.5	0.25
NaCl		0.1	0.1	0.04
Limestone		1.2	1.14	0.45
Methionine		0.25	0.13	0.13
Lysine		0.25	0.13	0.13
Total		100	100	100
Calculated Values³				
M.E. Kcal/ Kg Diet		3033	3150	3190
Crude Protein %		22.75	21.11	20.9
Crude Fibre %				
Lysine, %				
Methionine Plus Cystine %				
Ca , %				
Available P, %				

¹ Soybean cake used an Argentine source of crude protein content by 48% and 2440 Kcal/ Kg M.E.

² Protein Meal User Product From Netherlands Origin (Brocon) Contain 40% Crude Protein 0.2107 Kcal / Kg Protein M.E., 0.5% Crude Fat 2.20% Crude Fiber 5%, Calcium 4.68% ,Phosphorus 3.85% Lysine 4.12%, Methionine 4.12% , Methionine Plus Cystine 0.42%, Tryptophan 0.38%, Threonine 1.70%. It Contains A Mixture Of Vitamins And Minerals Needed Believes Rare Birds Of These Elements.

³Based on National Research Council recommendations (1994).

RESULTS AND DISCUSSION

The Experiment Lab

Detection of aflatoxin b1 using thin layer chromatography (TLC) and quantification of AFBI using high performance liquid chromatography (HPLC).Detection results for AFBI by using (TLC) and standard for AFBI showed the isolate ability to produce AFBI. Quantification result by HPLC showed the fungus isolate was productive for AFB1 and concentration of lower and this consistent with previous studies showed that *A. flavas* is able to produce aflatoxin (AF) at higher, concentration AFB! : 186 – 740000 ppm (aziz et al ,2000 ; al-othman et al , 2014).

Results in table 2 indicate that there is a high efficiency reduction of the rarious concentrates of filax in reducing the toxin –productive treatment. The reduction rate reach to 87,100 and 100% of concentrates (0,0.2 and 0.3 %) respectively compared to the control treatment. The filax high efficiency in reducing *Aspergillus flavus* is because of what it contains of strong acids. Previous studies indicated that the brosil that contain 99% of propionic acid has a great effect in reducing the *Aspergillus flavus* growth reaching to 100 % by using concentrates of 11000-20000 ppm (Alhiti , 1977). Such results also agree with what al-nazal and et al (2004). The Al-falahi 2005 , Hussein 2008 and al-qassi 2014 found the effectiveness of a number of acids reduces many types of *Aspergillus flavus* that afflict seeds in the stores as well as in the feeds of poultry and animals. Saloumi (2007) mentioned the effectiveness of filax in reducing the growth of Fusarium while Hussein (2015) referred that there is no type of Fusarium in yellow corn when treated with filax solution while al-qaisi (2010) mentioned that in 0.1 % concentration , the percentage of reduction was 92 % against F.graminearum.

Table2.effect of different concentration of phylax in the isolation of aflatoxin in 7 days

Con. Of phyax	0.00	0.1	0.2	0.3
Inhibition %	0.00 c	87 b	100 a	100 a
Sg.	**			

Means in the same row with different superscripts were significantly different (P < 0.01).

Results of table 3 indicated the high effect of the Iraqi probiotic in reducing the *A.flavus* in low concentration in the implant dish. The concentrations 0.03 ,0.02,0.1,0.2 and 0.5 achieved reduction rate of

55,64,74,87 and 100 % respectively, compared to control treatment . the high efficiency collaborative Iraqi probiotic may return in reduction of treatment producing aflatoxin B1 to having a mix of bacteria such as lactobacillus , bifidobacterium , streptococcus and enterococcus, which compete with fungi on food and place in addition to its secretion of lactic acid , which has an effect in reducing the growth of fungi. Franco (2011) referred to the effectiveness the lactic acid bacteria that may reduce the growth of *F.graminearum* , in addition that some types of bacteria that comprise the vital mixture that produce fungal antibiotics that reduce the growth the fungal treatment such as bacterioines (osuliran and et al m 2010). Some bacterial types produce volatile fatty acids such as propionic and butyric , leading to the raise of PH to 6.6 and reducing the growth of the pathogenic fungi (Al-Khafaji , 2008).

Table 3. the effect of different concentration of the Iraqi probiotic in the isolation of aflatoxin production with 7 day.

Con.	0.00	0.02	0.03	0.1	0.2	0.5	Sg.
Colony diameter rate (cm)	9 a	4 b	3.5 b	2.1c	1.1cd	0 d	*
Inhibition %	0 e	55 d	65 cd	74bc	87ab	100a	**

Means in the same row with different superscripts were significantly different (P < 0.01) and (P<0.05).

The results of table 4 indicated that ozone gas by 4 g/m concentration with 10 minutes ahead of time compared to the concentration of 2 g/m and the control treatment , for the rate of reduction 100 and 96 % respectively . the high efficieny of ozone gas may return to the effect of gas on the cell tissue. Abdul wahab (2011) and others reached to the fact that exposing sample contaminated with certain types of fungi and aflatoxin to various doses of ozone gas in 20 ppm for 5 minutes , 40 ppm for 10 minutes and 50 ppm for 5 minutes were effective in reduction of numbers of fungi depending on the amount of dose and time of exposure.Oztekin and et al (2007) found through dipping the dried figs with ozone gas and other with an ozone sterilized water the number of contaminated tiny creatures reduced to 51.5 % when treated with ozone and to 8.77 when treated with the other method.

Tabel 4. The effect of different concentration of ozone gas in the isolation of aflatoxin B1.

Treatment	Con.	Inhibition percentage (min.)			Sg.
		10 min	20 min	30 min	
Ozone	2 g/min	94 a	85 a	71 b	**
	4 g /min	100 a	83 b	80 b	
Control	2 g/min	1.01 b	1.2 ab	2.3 a	*
	4 g/min	0 c	1.3 b	3.1 a	

Means in the same row with different superscripts were significantly different (P < 0.01) and (P<0.05).

Results of table 5 showed that adding the ozone with 4 g/m concentration for 10 minute and filax with 0.2concentration and the probiotic 2% to yellow corn contaminated with toxin has led to a significant reduction in the toxin amount relative to the control treatment. The percentage of reduction was 89.52 , 89.52 and 81.20 respectively. A previous study showed the efficiency of ozone in damaging aflatoxin B1 in yellow corn and dried fruits (al-haddad , 2013: tuma ,2015).such studies agree with what 21 reached that ozone can damage B1,B2,G1 and G2 aflatoxin with 50 micro g/ml concentration and that B1 and G2 toxin are more sensitive to ozone gas. They notice the damage of the double bond between the two carbon atoms 8 and 9 , which will lead to forming a more settled component called aflatoxinozonid. With the continuation of interaction, the other double bone will be damaged in the last component and the peripheral furan cycle will be opened leading to the formation of the AFB1- dialdehyde, which is less poisonous and mutated.

Table 5. testing the efficiency of the ozone and probiotic of Iraq and phylax in the detoxifictontion of aflatoxin in stored maize.

Reduction ratio %	Con. AFB1 ($\mu\text{g/kg}$)	Type of treatment
0.00 c	1.05 a	Contaminated control
0.00 c	0.16 b	Control with out contaminated
89.52 ab	0.11 b	Corn + aflatoxin + filax
100 a	0.00 b	Corn + filax

81.20 b	0.02 b	Corn + aflatoxin + Iraq probiotic
99.29 a	0.06 b	Corn + Iraq probiotic
89.52 ab	0.11 b	Corn + aflatoxin + ozone gas
81.25 b	0.03 b	Corn + ozone gas
*	**	sg

Means in the same row with different superscripts were significantly different ($P < 0.01$) and ($P < 0.05$).

The Experiment Field

The table 6 refers to a significant distinction ($P < 0.05$) in the rate of the treated bird bodyweight T3 in which the yellow corn contaminated with aflatoxin B1 and treatment with ozone was used at the age of 3 weeks , compared with the treatment bird T8 and found a significant difference with the remaining treatment, while at the age of 6 week , a significant distinction was noticed along with a significant increase in the average of bodyweight of the two treatment T3and T5 ,which were 2553.75 and 2516.05 grams the average of body weight respectively when compared with the treatment T8 , T4 and T2 but they are not different significantly of T7 , T6 and T1. They recorded a body weight average of 2450.09 , 2424.60 and 2452.81 grams respectively. Through the table there were no significant differences between the treatment T1,T4,T6,T7 and T8 at the end of experiment.

Table 6. Effect of the addition of ozone gas , filax and Iraq probiotic to a diet contaminated with aflatoxin B1 in body weight for the period (3 and 6) weeks.

treatment	Age weeks	
	3	6
T1	698.000^{AB} ± 11.00	2450.00 AB ± 31.10
T2	688.50^{AB} ± 3.50	2287.50C ± 66.50
T3	707.50^{AB} ± 0.50	2516.50 A ± 52.95
T4	698.50^{AB} ± 6.50	2373.60 B ± 33.90
T5	688.50^{AB} ± 3.50	2553.00 A ± 33.20
T6	703.50^{AB} ± 7.00	2360.80 B ± 29.50
T7	681.00^{AB} ± 1.00	2347.10 BC ± 38.40
T8	699.50^B ± 7.50	2424.60 AB ± 41.70
Sg.	*	*

Means in the same row with different superscripts were significantly different ($P < 0.05$).

As for the weight increase of birds , the table 7 indicates that there are no significant differences between the treatment during the first stage of the study (1-3) weeks ,while during the second stage (4-6) week all noticed there are significant differences in the weight increase average between the treatment for they significantly succeeded ($P < 0.05$) both the treatment T3 and T5 in comparison with the treatment T2,T4,T6 and T8 but they are not different significantly from the treatment T1 and T7 . the success in the average of weight increase of the bird treatment T5 compared other experiments treatments (T1,T2,T4,T6,T7 and T8) clear during the complete breeding period (1-6) weeks. They are not different significantly from the treatment T3 which in turn showed a significant distinction ($P < 0.05$) in the average of the weight increase compared to the treatment T2 which is not different significantly from the treatment T4, T6 and T8 .

The table 8 indicated that there are significant differences ($p < 0.05$) in the average of feed consumption for the experiment treatment during the first period (1-3) weeks , the feed consumption was increased significantlu ($P < 0.05$) for the treatment T2,T3 and T6 compared to treatment T4,T5 and T7and they are not different significantly from the treatment T8. No significant differences appeared between the treatment in amount of consumed feed for (4-6) weeks and no significant differences appeared during the whole period of the experiment (1-6) weeks. As for the feed conversion ratio , the table 9 showed that T4 recorded the best feed conversion ratio for the period 91-3) weeks with little significant difference ($P < 0.05$) with the treatment T2 and the are not different significantly from the remaining treatment. No significant differences appeared in the feed conversion ratio between the treatment for the period (4-6) weeks .

Table 7. effect of the addition of ozone gas , filax and Iraq probiotic to a diet contaminated with aflatoxin B1 in the increase in weight for the period (1-3),(4-6)and(1- 6) weeks.

treatment	Age weeks		
	1-3	4-6	1-6
T1	649.90 ±9.70	1761.00 AB ±53.00	2410.90 B ±62.70
T2	649.00 ±1.80	1625.50 C±110.50	2247.50 C±112.30
T3	655.05 ±1.95	1821.00 A±48.00	2476.05 AB±46.05
T4	664.40 ±10.90	1669.00 C±23.00	2333.60BC ±33.90
T5	655.25 ±1.55	1858.50 A±39.50	2513.75 A±37.95
T6	669.60 ±14.20	1715.00B±42.00	2384.60 BC±56.20
T7	638.80 ±5.40	1774.00 AB±35.00	2412.80 B ±40.40
T8	661.10 ±8.60	1646.00 C±158.00	2307.10 BC±149.40
Sg.	NS	*	*

Means in the same row with different superscripts were significantly different (P<0.05).

Table 8. effect of the addition of ozone gas , filax and Iraq probiotic to a diet contaminated with aflatoxin B1 in food consumption for the period (1-3),(4-6)and(1- 6) weeks.

treatment	Age weeks		
	1-3	4-6	1-6
T1	1095.75 ^{BC} ±1.10	3194.73±25.08	4290.48±23.98
T2	1149.05 ^A ±26.48	3146.79 ±61.24	4295.84 ±34.76
T3	1149.91 ^A ±15.80	3216.52±53.66	4366.43 ±37.86
T4	1075.54 ^C ±23.21	3253.21 ±17.67	4328.75±5.53
T5	1090.09 ^C ±4.37	3241.79 ±16.96	4331.88±12.58
T6	1169.20 ^A ±4.91	3177.84 ±13.19	4347.04 ±8.28
T7	1122.77 ^B ±16.33	3218.12±44.54	4340.89±28.20
T8	1130.68 ^{AB} ±9.78	3162.39 ±12.42	4293.07 ±22.21
Sg.	*	NS	NS

Means in the same row with different superscripts were significantly different (P<0.05).

Treatment T2 showed a significant increase (P<0.05) compared to the remaining treatment of the experiment. The treatment T5 showed a significant improvement (P<0.05) when compared with treatment T4,T6,T7 and T8 and there is a significant difference with the control treatment T1 and T3. The feed conversion ratio for T5 in which the yellow corn contaminated with toxin and treatment of probiotic 1/7 was used. The

table 10 shows a significant increase ($P<0.05$) in dressing percentage of treatment birds T3 compared to the treatment T1,T2,T4,T6 and T8 but they are not significantly different from T5 and T7 this significant increase appeared in the rate of chest piece to the same treatment T3 compared to all treatment used in experiment. They didn't appear any significant differences among all treatment in the percentage of the thigh piece and the drummer stick. Neither are they significant differences in the percentage of the eaten internal bowels for all treatment (11) but a clear calculated increase appeared in the weight of the liver in the treatment T2 compared to the rest of the treatment.

Table 9. effect of the addition of ozone gas , filax and Iraq probiotic to a diet contaminated with aflatoxin B1 in food conversion coefficient for the period (1-3),(4-6)and(1- 6) weeks.

treatment	Age weeks		
	1-3	4-6	1-6
T1	1.63 AB ± 0.25	1.85 ± 0.07	1.75 AC ± 0.05
T2	1.72 A ± 0.05	2.06 ± 0.07	1.89 A ± 0.06
T3	1.66 AB ± 0.02	1.85 ± 0.10	1.75 BC ± 0.04
T4	1.58 B ± 0.04	2.01 ± 0.03	1.79 B ± 0.005
T5	1.61 AB ± 0.02	1.83 ± 0.05	1.72 C ± 0.01
T6	1.68 AB ± 0.02	1.89 ± 0.01	1.79 B ± 0.02
T7	1.68AB ± 0.02	1.95 ± 0.04	1.81 B ± 0.03
T8	1.65 AB ± 0.02	1.99 ± 0.21	1.82B ± 0.10
Sg.	*	NS	*

Means in the same row with different superscripts were significantly different ($P<0.05$).

to a diet contaminated with aflatoxin B1 in food conversion coefficient for the period (1-3),(4-6)and(1- 6) weeks.

Through the results of the experiment it is noticed that a significant deterioration in the productive characteristics of the birds in the treatment from the rest of the treatment which filax, the Iraq probiotic and the ozone were added to them with significant differences reflected in the productive performance. And the treatment contaminated with toxin and treated with filax , Iraqi probiotic and ozone recorded clear significant differences from the control treatment. This is because of the high ability of filax to reduce the aflatoxin due to the fact that its efficiency in damaging the toxin is attributed to having protective acids which lead to opening of lactone cycle responsible for the toxin nature of component (Hameed ,2000). The (Hiti,1977) indicated that the propionic acid has a high ability to smashing the fungus toxins and that the citric and lactic acids are able to damage the fungus toxin (reiss , 1976). Hussein (2008) reached to reduction of aflatoxin contamination 20 , 30 and 60 ppm to 10.98 , 19.20 and 33.20 in million respectively when treated with filax. Salomi(2007), Baldawi (2007), al-qaisi(2010) and Hussein (2008) reached that filax has the ability to damage various fungus toxin. As for the Iraqi probiotic , it contains a group of bacteria that secrete lactic acid that damages the fungus toxin, such a group was used in removing toxins working as binding factors to the fungus toxin. The toxin are physically connected to the surface of the bacteria. These results agreed with (El-nazani et al ,2000). Moreover ,the cellular wall sugars of the yeast such as glucoxidase are considered and absorbing factor of the fungus toxin converting them into another non- poisonous. This research was largely used by moschini (2008) and et al and smith and et al (2008) .it is manufactured by Altech company with the trade name ® mycosorb.the study agrees with what Nof (2012) and other mention that the early exposure to microbes by broiler chicks meat of bacteria lactobacilli has an effective role in reducing the effect of aflatoxin B1 in the relative weight of liver , heart and the glandular stomach. Moreover ,the yeast wall connects the fungus toxin and this is confirmed by murthy and devegoda (2004) who studied the effect of adding 250 and 500 ppm of aflatoxin to diet of broiler at age of 5 weeks supported by the product glucomannan existing in the yeast wall. They both noticed that this product

connects 75 % of aflatoxin during 30 minutes leaning in the increase of this rate after 90 minutes to be 90% in addition to the ability of number of isolated lactobacilli bacteria strains of the brewed probiotic it connect aflatoxin molecules with bigger rate of Pediococcus and Leuconsitus . the results of Iraqi probiotic proved what the Roashan (2006) found who confirmed there is asignificant improvement in the average of body weight of broiler at 28 days of age with a rate 31.1 , 30,20.3 and 25.3 when adding 5 grams of tazri mixture of biomin and the bio former respectively to each kilogram of feed contaminated with aflatoxin compared to the treatment of toxin alone as a result of the action of probiotic against this toxin treatment of ozone have high efficiency in reducing toxin and the productive features improved because ozone works to damage toxin through conducting chemical changes in the various components. It interacts with aflatoxin through attacking the dual aliphatic bond between the carbon atoms 9and 8 leading to form unsettled intermediate components with uni-ozone called aflatoxinmonozonides and through rearrangement of atoms such components changes into other derivatives call aflatoxinozonides and then the dual bonds were cut in such components to composed other components represented by aldehydes , ketones and organic acids (doctor et al,2004). These results agreed with Drudents and king (2002) who confirmed that treatment the yellow corn contaminated with aflatoxin and ozone gas by 10-12 % of the total weight of gas used led to reduction of toxin by 92%.

Liver is among the most important organs in the body that are exposed to fungi toxins , and then many vital activities will be affected depending on the amount of fungi toxins and their concentrations (aruna et al , 1998). Aflatoxin is the most dangerous kinds of aflatoxin and the most powerful carcinogenic chemical components. This component enters into the body ,some of which is absorbed and the rest gets out with the wastes. The absorbed part reaches to the liver through P450 enzyme exist in the cytochrome system and then it is changed into epoxide (AFBO) AFB1-8-9-Epoxi which is connected with the nitrogenous base in the nuclear acids DNA and RNA causing a change in the succession of nitrogenous bases . so the carcinogenic and mutated effect appears (Crespi et al , 1991 ; benned and klich ,2003). Wytt (1991) and others confirmed the occurrence of damages in liver in the baby chicken as a result of feeding them on contaminated diets with aflatoxin B1 most vulnerable organs because it is noticed cirrhosis with greasy clumps inflation of gall channels in both chicken and ducks.From this study, we conclude that adding ozone gas with 4 g/m concentration for 10 yellow corn contaminated with aflatoxin B1 has led to significant reduction in the amount of toxin and to the reduction of the negative effect of the toxin in productive performance of the broilers fed on diets containing yellow corn contaminated with B1 toxin and the high efficiency of Iraqi probiotic whether in damaging toxin or as a nutrition substance that increased the nutritional value of the diet.

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