ISSN 1110-0419

Annals Of Agric. Sc., Moshtohor,
Vol. 44(1): 125-133, (2006).

PRODUCTIVE PERFORMANCE OF JAPANESE QUAIL FED ON DIETS MIXED WITH NATURAL ANTI-AFLATOXIN COMPOUNDS

BY

Hala M. Gamal El-Deen*; Azza A. Mustafa and Nahed S.Yousef*****

* Poultry Nutrition, Biological and Environmental Dept., Faculty of Home Economic,

** Mycology, Biological and Environmental Dept., Faculty of Home Economic,

*** Food science and Technology Dept., Faculty of Home Economic, Al-Azhar University, Tanta, Egypt.

ABSTRACT

Some natural and organic compounds were investigated as a possible aflatoxin B_1 decomposer in spoiled layer fodder. The most effective of these compounds were mixed with fresh layer fodder in order to evaluate their effect on the performance of Japanese quail. The natural compounds were tested as aflatoxin B1 decomposers at levels of 1, 2 or 3%. It was found that 3% tea extract, citric acid or dried garlic gave the highest effect on reducing aflatoxin B_1 which reached 207.0, 214.2 and 237.6 ppt respectively as compared with 413.75 ppt in the spoiled control untreated fodder. Therefore, tea extract, citric acid and dried garlic were individually mixed with fresh layer basal diet at the level of 3%. Egg quality, carcass values and blood biochemical analysis of Japanese quail showed similarity among the different experimental quails as compared with the control diet. Plasma lipid was however, decreased significantly on adding either dried garlic or citric acid. It might be concluded that 3% tea extract, dried garlic or citric acid are safe additives to the diet of Japanese quail. These additives not only had significantly reduced aflatoxin B_1 but also did not exert any harmful effect on the performance of the Japanese quail.

Key words: Aflatoxin B1, tea extract, garlic, citric acid, feed additives and quail performance

INTRODUCTION

Shortage in cereal production accompanied with increasingly continuous demands on livestock feeds is considered a major problem in Egypt. In addition the presence of moulds in feedstuff due to the bad handling and unfavorable storage conditions is another important problem (Mohamed, 2003).

Mycotoxins are toxic compounds produced by fungi, and aflatoxin B_1 is the most potent of the aflatoxins (Wood, 1989). When aflatoxin B_1 and B_2 contaminated food or feed is consumed, the toxins are metabolized to aflatoxins

M_1 and M_2 and excreted into the tissues, biological fluids, and milk of lactating animals, including breast milk resulting in a significant decrease in blood total protein and blood albumin (Pegram & Wyatt, 1986).

It has been reported that tea extract had strong inhibitory effect on moulds producing aflatoxins and also, had anti-toxigenic activity on the produced aflatoxins B_1, B_2, G_1 and G_2 (Hasan, 1999 and Yousef, 2002).

Organic acids such as acetic, propionic, sorbic and citric acid are commonly used as preservatives in food industry and were found to be effective in preventing fungal growth and subsequently the aflatoxin production (Viera, 2003).

It has been also mentioned that not only performances of laying hens were not affected by diet supplemented with up to 10 % sun dried garlic paste, but also, concentrations of serum and yolk cholesterol were reduced (Chowdhury *et al.*, 2002). Moreover, it has been found, in a recent study that 3% sterilized garlic oil had moderate antifungal effect on molds contaminated fodder (Yousef *et al.*, 2005).

Therefore, the present study was carried out in order to evaluate the effect of some natural and organic compounds on aflatoxin B_1 levels in the fodder. Also, the performance of Japanese quail fed on diets mixed with these natural materials was followed.

MATERIALS AND METHODS

Evaluating some natural compounds as aflatoxin decomposers and the productive performance of the Japanese quail layers fed on diets supplemented with these materials was carried out at Faculty of Home Economic, Al-Azhar University. Two experiments were performed, the first one aimed at evaluating the effect of some organic acids and natural compounds on reducing the concentration of aflatoxins on a spoiled diet. The second experiment was executed by using the results obtained in the first one, according to the low concentrations of aflatoxins, the compounds in choice being 3% dried garlic, tea extract (25%), and citric acid.

1- Evaluation of natural components as aflatoxin decomposers:

The formulated feed (Table 1) was stored and left till it was spoiled with moulds. The contaminated feed was then treated with 1, 2 or 3 % of acetic acid, lactic acid, citric acid, cinnamic acid, cinnamaldhyde, dried garlic, garlic oil and dried tea. Also, 25 g of garlic paste and dried tea were individually extracted in 100 ml boiling water, then 1, 2 or 3 % of these extracts were separately added to the contaminated fodder. The treated feeds were then stored at room temperature for 15 days and the aflatoxin B_1 concentration was determined according to Ridascrean^R Aflatoxin B_1 Enzyme immunoassay for quantitative analysis of aflatoxin B_1 (R-Biopharm AG Darmstadt, Germany).

2- Evaluation of natural compounds effect on performance of Japanese quail:

Seventy-two of five weeks old Japanese quail laying hens as a commercial strain obtained from Fac. of Agric., Cairo University were used in this experiment. Equal numbers of hens were randomly distributed into four groups, a control and three treatment groups. Each group contained 18 birds was divided into 6 replicates (3 hens each) and raised in wire cages for 8 weeks period in an open system house. The Japanese quails were given fresh formulated (NRC, 1994) diets (Table 1) and water, adlibitum, during the study. The control group was fed on the basal diet, while the experimental groups were fed on the same diet mixed with 3 % of dried garlic paste, tea extract or citric acid.

Table (1): Composition and calculated analysis of the basal diet.

Ingredients	%
Corn (yellow)	62.01
Soybean meal (44 %)	22.95
Corn gluten (60 %)	6.0
Dicalcium phosphate	1.5
Limestone	3.5
Salt	0.32
Oil (plant)	1.0
Premix*	0.4
DL-Methionine	0.1
Lysine	0.22
Total	100
Calculated analysis	
Crude protein	19.91
Crude fiber	2.28
Crude fat	3.76
Metabolizable energy	2966.69
Calcium	2.53
Available Phosphorus	0.398
L. lysine	1.09
Meth+ Cys.	0.489

* Premix provided/Kg of the final diet: Vit. A, 12500 IU; Vit.D3, 2500 IU, Vit.E (91% alpha-tocopherol- acetate) 20.9 mg; Vit. B1, 1.25 mg; B2, 5.5 mg; Vit. B6, 3.5 mg; Vit. B12, 12 mg, pantothenic acid, 9 mg; folic acid, 0.9 mg; biotin, 60 mg; Trace minerals, Fe, 35 mg; Cu, 7 mg; Zn, 50 mg; Mn, 80 mg; I, 350 mg; Se, 150 mg; Co, 400 mg.

Egg quality:

The daily egg production of each group was individually weighed and recorded, from the first laid egg up to the end of the experimental period. Records of egg production and egg weight together with feed consumption were used to calculate the values of feed conversion ratio. Feed intake was calculated as g feed /layer /day and g feed /layer /6 weeks. Eggs were randomly taken from the daily production to determine yolk, albumin and shell parameters.

Haugh units, as an indicator for albumin quality, were calculated on the basis of the individual egg weight and the albumin height using the formula reported by Neshein *et al.* (1979).

$$\text{Haugh unit} = 100 \log (H + 7.57 - 1.7\, W^{0.37})$$

Where:
H = Albumin height, mm
W = Egg weight, g
7.57, 1.7 and 0.37 are constants

Carcass characteristics:

At the end of the experiment, 6 quails of each treatment were randomly selected, weighed and slaughtered. After bleeding, scalding and feather picking by hand, liver, kidney, spleen and gizzard were eviscerated and weighed. Small intestine thickness (SIT) was determined and was used as indicator of the effect of the tested growth promoters on microorganisms populations in Alimintary canal (Stutz *et al.*, 1983).

Biochemical analysis:

At the end of the experiment, 6 birds of each treatment were randomly taken and slaughtered and blood samples were collected in a sterile heparinized centrifuge tubes. The samples were then centrifuged for 20 min. at 3000 rpm and plasma samples were then stored at –20°C until analyzed. Plasma total protein was determined according to Biuret method described by Henery (1964). Plasma creatinin was determined according to Bartels (1971) and albumin was estimated as mentioned by Doumas *et al.* (1972). Plasma globulins were obtained by subtracting albumin values from total protein values. Plasma total lipids were determined according to Knight *et al.* (1972) and total cholesterol according to Watson (1960). Glutamic pyruvic transaminase (GPT) and glutamic oxalo acetic transaminase (GOT) were determined as described by Reitman and Frankel (1957).

Statistical analyses

The data obtained were subjected to analysis of variance according to SPSS (1997). Significant differences among individual means were analyzed by Duncan's multiple range test (Dancan, 1955).

RESULTS AND DISCUSSION

Evaluation of natural compounds as aflatoxin decomposers

The influence of chemical and herbal compounds at levels of 1, 2 and 3% on aflatoxin B_1 formed in the experimental fodder is shown in Table 2. It can be seen that aflatoxin B_1 was significantly affected by the high levels added of the natural compounds. The lowest B_1 concentration was reached when 3% of either tea extract, citric acid or dried garlic paste was mixed with the experimental fodder. B_1 concentration reached 207.0, 214.2 and 237.6 representing a percent decrease, compared to control, of 49.97, 48.23 and 42.57 for 3% dried tea, citric acid and dried garlic, respectively. Other natural compounds exerted however, weaker effect on aflatoxin B1.

These results were in agreement with those of Reiss (1976) who mentioned that aflatoxin B_1 and G_1, which induced by *Aspergillus parasiticus* were suppressed in bread supplemented with citric acid and lactic acid .In addition, feeding toads on minced garlic or garlic oil resulted in marked reduction

in the incidence of tumors induced by aflatoxin B1 (El-Mofty *et al.*, 1994). Moreover, Yen and Chen (1995) also stated that water extract of tea showed strong antimutagenic action against aflatoxin B_1. Moreover, Hasan (1999) reported that tea extract at the level of 3% had antitoxigenic activity on aflatoxin production. Also, he found that tea extracts at 1 and 3% had reduced aflatoxin production in liquid broth and suggested that tannin and caffeine might be the effective agents.

Table (2): Effect of different levels of natural and organic compounds mixed with layers diets on aflatoxin B_1 (ppt*).

Organic or natural products	Supplemented level, %				Overall
	0	1	2	3	
Dried garlic	413.75	383.2	259.2	237.6	323.44
Garlic extract (25%)	413.75	295.2	261.0	257.4	306.38
Garlic oil	413.75	329.4	293.8	250.0	323.04
Dried tea	413.75	370.4	325.8	304.2	353.54
Tea extract (25%)	413.75	322.2	268.2	207.0	302.79
Acetic acid	413.75	412.2	336.6	306.0	367.14
Lactic acid	413.75	360.0	334.8	275.4	345.98
Citric acid	413.75	354.6	273.2	214.2	313.94
Cinnamic acid	413.75	349.2	327.6	313.2	350.94
Cinnamaldhyde	413.75	390.6	295.2	259.2	333.69
Overall	413.75[a]	356.7[b]	298.04[c]	262.44[c]	

* ppt = part per trillion

a, b and c means in the same row within the same item followed by different superscripts differ significantly at $P< 0.05$.

Evaluation of natural compounds effect on performance of Japanese quail

As a result of the previous data, 3% of either dried garlic, tea extract or citric acid were mixed with fresh layer diets in order to evaluate their effect on the performance of Japanese quail. Table 3 indicates that egg quality of Japanese quail responded positively to the experimental diet. Egg weight (g), feed intake (g/layer/day) were significantly higher than the control. Total egg number, egg mass (g/layrt/6 weeks) were, however insignificantly higher than the control. On the other hand, feed conversion (kg feed/kg eggs) was almost similar in the experimental diets and the control. Other parameters of egg quality were found to be similar as a result of the different experimental diets. The insignificant differences in Haugh units indicated that the laid eggs tend to have no watery albumin as a result of adding garlic, tea extract or citric acid.

The same observations were mentioned by Chowdhury *et al.* (2002) who mentioned that egg quality of different strains was not affected by feeding diet supplemented with up to 10% sun dried garlic past. Also, in an earlier studies Brown and Lee (1985) reported that citric acid had not affected the feed efficiency ratio of broiler chicks.

Table (3): Effect of 3% tea extract, dried garlic and citric acid mixed with layer diets on egg quality of Japanese quail.

Parameters	Control	Tea extract	Dried garlic	Citric acid
Egg number, layer /6 weeks	34.1	35.7	35.17	35.24
Egg weight, g	10.12^{a}	12.25^{b}	11.97^{b}	11.85^{b}
Egg mass, g/layer/6 weeks	345.09	437.3	420.98	417.59
Feed intake, g feed/layer/ day	38.05^{a}	50.1^{b}	45.96^{b}	43.49^{b}
Feed intake, g feed/layer/6 weeks	1712.25^{a}	2254.5^{b}	2068.2^{b}	1957.05^{b}
Feed conversion, Kg feed/Kg eggs	4.96	5.15	4.91	4.69
Egg weight, g	10.35^{a}	12.34^{b}	12.03^{b}	12.14^{b}
Shell, %	21.12^{a}	15.22^{b}	15.85^{b}	15.73^{b}
Albumin, %	45.64^{a}	50.309^{b}	50.896^{b}	49.186^{b}
Yolk, %	33.24	34.47	33.26	34.68
Shell weight, g	2.18	1.878	1.907	1.91
Albumin weight, g	4.72	6.208	6.12	5.97
Yolk weight, g	3.44	4.253	4.001	4.21
Yolk volume, cm^3	4.26	4.4	4.7	4.6
Albumin volume, cm^3	5.7	5.4	5.6	5.6
Albumin height, mm	6.84	6.86	7.22	6.86
Yolk height, mm	11.82	13.48	12.25	12.36
Yolk diameter, mm	4.36	4.35	4.45	4.41
Haugh unit	101.58	100.40	102.20	100.64

a, b, ... means in the same row within the same item followed by different superscripts differ significantly at $P< 0.05$.

Data presented in Table 4 shows carcass performance as affected by adding 3% tea extract, dried garlic or citric acid to the layer diet. It can be seen that carcass values were generally comparable in the different treatments as compared with the control diet. However, adding dried garlic resulted in a significant increase in heart percent and tea extract produced slight increase in intestine thickness. It has been mentioned in a similar studies that small intestine thickness calculated as small intestine weight (g) / small intestine length (cm) was not affected when broiler chicks were fed on a diet supplemented with organic acids. The thickness of intestinal wall is considered a good indicator for the number of microbial populations in intestinal lumen (El-Afifi, 1997).

Data illustrated in Table 5 indicates that adding 3% tea extract, dried garlic or citric acid to the layer diet had no harmful effect on blood parameters of Japanese quail. Moreover, a significant decrease in plasma lipid was noticed when adding either dried garlic or citric acid. Plasma lipids (mg/100 ml) reached 459.0 and 465.9 when dried garlic and citric acid were added to the diet, respectively as compared with 492.6 for the control treatment. Also, El-Kerdawy (1996) reported a similar reduction in blood total lipid in rabbits as a result of adding garlic or citric acid to the diet. In the same direction El-Afifi (1997) mentioned that adding garlic to the poultry diet did not cause any deleterious effect on plasma total protein, albumin or globulin.

Table (4): Effect of 3% tea extract, dried garlic and citric acid mixed with layer diets on carcass performance of Japanese quail.

Organs	Control	Tea extract	Dried garlic	Citric acid
Alive weight, g	316.50	333.8	318.80	302.0
Liver, %	3.00	3.70	3.27	3.28
Gizzard, %	2.02	2.14	2.00	2.30
Heart, %	0.71^{a}	0.70^{a}	0.96^{b}	0.73^{a}
Spleen, %	0.060	0.049	0.086	0.060
SIT*, g/cm	0.14	0.17	0.13	0.13

* SIT = small intestine thickness

a and b means in the same row within the same item followed by different superscripts differ significantly at $P< 0.05$.

Table (5): Effect of 3% tea extract, dried garlic and citric acid mixed with layer diets on blood parameters of Japanese quail.

Parameters	Control	Tea extract	Dried garlic	Citric acid
Protein, g/dl	3.60	3.38	3.60	3.64
Albumin, g/dl	2.09	2.03	1.88	2.14
Glubulin, g/dl	1.52	1.35	1.68	1.5
A/G ratio	1.38	1.50	1.12	1.43
Lipids, mg/100 ml	492.6^{a}	490.1^{a}	459.0^{b}	465.9^{b}
Cholesterol, mg/dl	135.25	133.95	132.75	132.78
GPT, U/l	5.38	6.50	7.00	6.65
GOT, U/l	110.8	109.5	108.0	109.2
Creatinin, mg/dl	1.63	2.10	2.03	2.08

A,b ... means in the same row within the same item followed by different superscripts differ significantly at $P< 0.05$.

Finally, it might be stated that adding 3% tea extract, dried garlic or citric acid not only had reduced aflatoxin B_1 during storage of the experimental diet but also had no negative effect on the performance of the Japanese quail. Therefore, feeding layer Japanese quail with fodder mixed with 3% of either tea extract, dried garlic or citric acid might be safe and recommended.

ACKNOWLEDGEMENT

The authors are grateful to Al-Azhar University for the partial financial support (from chapter 2, item 3) of this study.

REFERENCES

Bartels, H. (1971): Estimation of creatinine. Clin. Chem. Acta., 33: 81.

Brown, D.R. and Lee, S.L. (1985): Effect of citric and ascorbic acids on performance and intestinal pH of chicks. Poultry Sci., 64: 1399.

Chowdhury, S. R.; Chowdhury, S.D. and Smith, T.K. (2002): Effect of dietary garlic on cholesterol metabolism in laying hens. Poultry Sci., 81: 1856–1862.

Doumas, B.; Watson, W. and Biggs, H. (1972): Albumin standard and measurements of serum with bromocresol green. Clin. Chemical. Acta, 31: 87.

Duncan, D. (1955): Multiple range and multiple F test. Biometrics, 11: 1-42. Eddition, Assol. Offic. Anal. Chem., Arlington, AV. USA.

El-Afifi, Sh.F. (1997): Nutritional studies on onion and garlic supplemented to poultry feed. Ph. D thesis, Poult. Prod. Dept., Fac. Agric., Ain Shams Univ.

El-Kerdaway, D.M.A. (1996): Acidified feeds for growing rabbits. Egypt. J. of Rabbit Sc., 6 (2) 143-156.

El-Mofty, M.M.; Katr, S.A.; Essawy, A. and Gawad, H.S.A. (1994): Preventive action of garlic on aflatoxin B1 indiced carcinogenesis in the toads Bufo regularis. Nutrition and Cancer, 21 (1): 95-100.

Hasan, H. (1999): Role of caffeine and tannin in anti- toxigenic properties of coffee and tea. Cryptogamic Mycology, 20 (1): 17-21 .

Henery, R.J. (1964): A colorimetric method for the determination of the total protein. Clin. Chemical. Harper & Raw Publisher New York, pp. 181.

Knight, J.A.; Anderson, I.S. and Rawle, J.M. (1972): Chemical basis of the sulfa-phosphate vanillin reaction for estimating total serum lipid. J. Biol. Chem. 226: 497.

Mohamed, E.A.A. (2003): Studies on the performance of some local strains of chickens as affected with fungi toxins. Ph. D. Thesis. Fac. of Agric., Al-Azhar Univ.

Neshein, M.C.; Austic, R.E. and Cord, L.E. (1979): Poultry production, 12th Ed., Lea & Febiger, Philadelphia.

NRC National Research Counsel. (1994): Nutrient requirement of poultry 9th Ed. National Academy Press, Washington, DC., USA, Academy of Sci.

Pegram, R.A. and Wyatt, R.D. (1986): The relationship of certain blood parameters to aflatoxin resistance in Japanese quail. Poult. Sci. 65: 1652 – 1658.

Reiss, J. (1976): Prevention of the formation of mycotoxins in whole wheat bread by citric acid and lactic acid. Experimenta. 32: 168-169.

Reitman, S. and Frankel, S. (1957): A colorimetric method for the determination of serum glutamic oxaloacetate and glutamic pyruvic transaminase. Am. J. Clin. Path., 28: 56-58.

SPSS. (1997): SPSS User's Guide Statistics Version 8. Copyright SPSS Inc., USA. Washington, DC., USA.

Stutz, M.; Johnson. S.L. and Judith, F.R. (1983): Effect of diet, bacitracin and body weight restrictions on intestine of broiler chickens . Poult. Sci., 62: 1626–1632.

Viera, S.L. (2003): Nutritional implications of mould development in feed-stuffs and alternative to reduce the mycotoxin problem in poultry feeds. World's Poult. Sci. J., 59: 111.

Watson, D. (1960): A simple method for the determination of serum cholesterol. Clin. Chem. 5: 639.

Wood, G. E. (1989): Aflatoxin in domestic and imported foods and feeds. J. AOAC. 72: 543–548.

Yen, G. and Chen, H. (1995): Antioxidant activity of various tea extracts in relation to their antimutagenic. J. of Agric. and Food Chem. 43 (1): 27–32.

Yousef, Nahed S. (2002): Tea extracts as possible natural food preservative for organic food. Annals of Agric. Sc., Moshtohor, 40 (2): 939-949.

Yousef, Nahed S.; Gamal El-Deen, Hala M. and Abdel-Monsef, Rehan A. (2005): Antifungal effect of some essential oils against different fungi isolated from broiler feed mixtures. Egypt. J. Agric. Res., 83 (2): 1075-1085.

الأداء الانتاجى للسمان اليابانى المغذى على علف مخلوط بمواد طبيعية مضادة للسموم الفطرية (الافلاتوكسين)

هالة محمد جمال الدين*، وعزة أحمد مصطفى، ناهد سامى يوسف*****

* تغذية دواجن- قسم علوم بيئية وبيولوجية - كلية الاقتصاد المنزلى.

** ميكولوجى - قسم علوم بيئية وبيولوجية - كلية الاقتصاد المنزلى.

*** قسم علوم وتكنولوجيا الأغذية - كلية الاقتصاد المنزلى - جامعة الأزهر - طنطا - جمهورية مصر العربية.

تم دراسة تأثير إضافة بعض المواد الطبيعية والمركبات العضوية على علف ملوث بالفطريات واختبار هذه الاضافات كمواد مضادة للتوكسينات الفطرية. هذه المواد الطبيعية والعضوية تم خلطها بالعلف بتركيزات ١ و٢ و٣ %.

لوحظ من بين هذه المواد المضافة أن مستخلص الشاى وحامض الستريك والثوم الجاف بتركيز ٣ % أعطت أعلى تأثير على أفلاتوكسين ب١ ووصل تركيز الأفلاتوكسين ٢٠٧ و٢١٤,٢ و٢٣٧,٦ جزء فى التريليون على التوالى بالمقارنة بالعلف الغير مضاف له أى من هذه المواد حيث كان تركيز الافلاتوكسين به ٤١٣,٧٥ جزء فى التريليون .

وأجريت تجربة بهدف تقييم خواص السمان اليابانى المغذى على العلف المضاف له مستخلص الشاى وحامض الستريك والثوم الجاف بتركيز 3 % . ولوحظ أنه عند التغذية على العلف المخلوط بهذه المواد لم يؤثر بصفة عامة على صفات الجودة للسمان من حيث كمية البيض وصفات الذبيحة وأيضا التحليل الكيميائى للدم. وقد أظهرت التجارب نقصان تركيز لبيدات الدم نتيجة لإضافة الثوم الجاف أوحامض الستريك. ومن هذه النتائج يتضح أن إضافة ٣% مستخلص الشاى أوالثوم الجاف أوحامض الستريك تعتبر إضافة آمنة لعلف التسمين للسمان. وهذه الإضافات قللت من التوكسينات الفطرية بالعليقة بالاضافة أنها لم تحدث تأثير سلبى على السمان اليابانى.