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Influence of the integration among oxytetracycline, oregano essential oil or garlic powder on intestine microbial population and productive performance of Japanese quail

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Abstract

Four essential oils (oregano, thyme, peppermint and garlic) were in vitroevaluated for their antimicrobial activity against some pathogenic microorganismwith concentrations of 10, 20, 50, 75, and 150µg/ml. The laboratoryresults showed that oregano and garlic essential oils were found to be more effective against all tested pathogenic microorganism.Consequently, the oregano essential oil and garlic powderwere chosen as feed additives on diets of quail birds. A total number of 480 Japanese quail chicks, 10 day old, were randomly selected, weighed and divided into eight experimental groups each of 60 birds (3 replicates each of 20 birds). The first group was fed on basal grower diet (considered as control). The second group was fed on diet supplemented with 1.0g oxytetracycline /kg diet. Birds of the 3rd, 4th and 5thgroups were fed on diets supplemented with oregano essential oil at levels of 50, 100 and 150 mg/kg combined with 0.5g oxytetracycline /kg diet, respectively, while birds of the 6th, 7th and 8th groups were fed on diets supplemented with garlic powder at levels of 0.1, 0.2 and 0.3 g/kg combined with 0.5g/kg oxytetracycline, respectively. The obtained results showed thatchicks fed on the dietsupplemented with 150 mg/kg oregano combined with 0.5g/kgoxytetracycline significantly improved body weight, body weight gain, feed conversion, carcass weight and economic efficiency.Moreover, it had the lowest intestine pathogenic populationscontent andmortality percentage followed by those fed on the diet supplementedwith 0.2 g/kg garlic powder combined with 0.5g/kg oxytetracycline.

Keywords: oregano essential oil, garlic powder, productive performance, microbial population

Introduction

Over the last few years, the use of antibiotics as growth promoters in animal and poultry feed has been decreased in several countries and it has been banned in others such as the European countries, because of the increasing concern regarding unsusceptible antibiotic bacteria. This shift has stimulated the search for alternative supplements, since use of medicinal plants or their essential oils are among these targeted alternative growth promoters(Williams and Losa, 2002; Lee *et al.*, 2003 and Hernandez *et al*., 2004). The medicinal plant extracts are characterized by several effects including antimicrobial activity (Lambert *et al*., 2001; Giannenas *et al*., 2003; Burt, 2004; Giannenas *et al*., 2004; Lee *et al.*, 2004 and Fukayama *et al.*, 2005) antioxidant activity (Basmacioğlu, H. *et al*., 2004; Giannenas *et al*., 2004 andFlorou-Paneri *et al*., 2006) enhancement of the immune system (Dorhoi*et al*., 2006) and consequently improvement of poultry performance.

Oregano (*Origanumvulgare*) is a species of *Origanum* genus which belongs to family Lamiaceae. Several studies have concluded that using the oregano products (oil extract or its dried leaf) as feed additives has enhanced broiler chicken performance (Halle *et al.*, 2001; Modeva and Profirov, 2003; Giannenas *et al.*, 2003 and Lihua *et al*., 2007) and reduce bacterial inhabitants in gastrointestinal tract such as *Clostridium perfringens*and *E. coli* (Giannenas*et al*., 2004 and Fukayama *et al*., 2005). Essential oils have been long known to possess antimicrobial properties (Mitsch*et al*., 2004). Several *in vitro* studies have demonstrated that essential oils eliminate pathogenic bacteria, among them *E. coli*, *Salmonella spp.* (including *Salmonella enteritidis* and *Salmonella typhimurium*), *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Staphylococcus epidermis*, *Klebsiellapneumoniae*, *Shigellaspp.*,*Proteus vulgaris*, and *Bacillus cereus* (Hammer *et al*., 1999; Inouye *et al*., 2003; Azaz *et al.*, 2004; Bagamboula *et al*., 2004 and Penalver *et al*., 2005).

Garlic (*Allium sativum*) is one of the oldest cultivated plant (Moyers, 1996). Several studies have concluded that garlic as a medical plants showed several effects including antimicrobial activity (Iwalokun *et al.*, 2004; Corzo-Martínez *et al.*, 2007 and Onibi *et al.*, 2009); inhibition of platelet aggregation (Apitz-Castro *et al.*, 1983); reduction in arterial blood pressure (McMahon and Vargas, 1993); prevention of fat infiltration of liver (Kim *et al*., 1995) and stimulation of immune functions (Sumiyoshi, 1997).

Garlic has rich organic sulfur compounds and precursors (allicin, diallyl sulfide, and diallyltrisulfide), whereallicin is the potentially

active component of garlic. These compounds provide garlic its biological properties and have been identified as having the hypocholesterolemic effect in human and animal products (Aouadi *et al*., 2000 and Chowdhury *et al*., 2002). Allicin and its derivatives has been shown to be bacteriostatic active against both gram positive or gram negative microorganisms as well as fungi such as *Candida albicans* and viruses including influenza viruses (Chang and Cheong, 2008). Additionally, garlic and garlic extracts have been shown to have antioxidant activity in various meat types (Yin and Cheng, 2003; Sallam *et al*., 2004 and Tang and Cronin, 2007).

Supplementing broiler chicken diets with garlic can produce chicken meat with favorable lipid profiles and can enhance eating quality because sensory panels found that thigh meat from chickens fed a garlic-supplemented diet had better texture and flavor (Kim, *et al*., 2008).

The objectives of the current study were to evaluate the effect of feeding quail birds on diets supplemented with different levels of either oregano essential oil or garlicpowder combined with the protective dose of oxytetracycline level 0.5 g/kg on the growth performance, mortality rate, feed efficiency, carcass yield, economic efficiency and microbial count in the small intestine.

Materials and Methods

This study was carried out in 2013/2014 as collaborative scientific work between the microbiological laboratories of Botany Department and the Poultry research Farm of Animal Production Department, Faculty of Agriculture, Benha University.

Four essential oils were *in vitro* evaluated as anti-microbial substances against pathogenic microorganisms to select some of them as feed additives on diets of quail birds.

Collection of medicinal plant materials

Plant extracts; oregano oil (*Origanummajorana*), thyme oil (*Thymus vulgaris*), peppermint oil (*Menthapiperita* L.) and garlic oil (*Allium sativum*) were obtained from National Authority for Control and Pharmaceutical Research to estimate their microbial activities against some pathogenic bacteria and fungi under laboratory condition.

Pathogenicmicroorganisms and growth media

Microorganisms were obtained from the culture collections of the Department of Microbiology at National Research Center. Bacterial strains;*Salmonella enteritidis* NAH77, *Staphylococcus aureus* NAM68, *Pseudomonasaeruginosa* NAM09, *Listeria moncytogenus*NAM98, *Yersinia enterocolitica* NAM45 and yeast strain *Candida albicans* NAA06 were chosen based on their clinical and pharmacological importance. The bacterial and fungal strains were used for evaluating antimicrobial activity of plant extracts. The bacterial strains were grown on Mueller-Hinton agar (MHA) plates at 37°C whereas the yeast were grown on Sabouraud dextrose agar at 28°C. The stock cultures were maintained at 4°C.

Antimicrobial activity of tested essential oils

In vitro, antimicrobial activity was assayed for both abovementioned essential oils and standard antibiotic (Oxytetracycline) with five dilutions (10, 20, 50, 75, and 150µg/ml) against pathogenic microorganisms. Antimicrobialactivities were investigated by agar diffusion methodaccording to (Kalemba and Kunicka, 2003).

Design of feeding experiment

A total number of 480 Japanese quail chicks, 10 day old, were randomly selected, weighed and divided into eight groups (treatments) each of 60 birds (each group contained 3 replicates of 20 birds), as shown in the following Table.

Table 1. Experimental design

Treatments	Description
T1	Birds fed on basal grower diet (control).
T2	Birds fed on basal diet supplemented with 1.0 g/kg oxytetracycline (20%).
T3	Birds fed on basal diet supplemented with oregano essential oil with level of 50 mg/kg + 0.5g/kg oxytetracycline (20%).
T4	Birds fed on basal diet supplemented with oregano essential oil with level of 100 mg/kg oregano essential oil + 0.5g/kg oxytetracycline (20%).
T5	Birds fed on basal diet supplemented with oregano essential oil with level of 150 mg/kg oregano essential oil + 0.5g/kg oxytetracycline (20%).
T6	Birds fed on basal diet supplemented with garlic powder level of 0.1g/kg+ 0.5g/kg oxytetracycline (20%).
T7	Birds fed on basal diet supplemented with garlic powder level of 0.2 g/kg+ 0.5g/kg oxytetracycline (20%).
T8	Birds fed on basal diet supplemented with garlic powder level of 0.3 g/kg+ 0.5g/kg oxytetracycline (20%).

Bird's management and estimated parameters

All Birds of experimental groups were kept under similar standard hygienic and environmental conditions. Battery cages with electric heater were used for brooding chicks. Chicks were fed ad-libitum on commercial grower ration formulated according to the recommended requirements of NRC, (1994) as showed in Table 2.

Table 2. Composition and calculated analysis of the basal diet.

Ingredients	%
Yellow corn	56.00
Soyabean meal (44%)	27.90
Corn gluten (60%)	12.00
di-calcium phosphate	1.70
Calcium carbonate	1.35
Salt	0.25
Hy-mix broiler premix	0.30
DL.methionine	0.20
Lysine	0.30
Calculated analysis	
ME(kcal/kg)	2948.3
CP (%)	24.39
Calcium	0.97
Available phosphorus (%)	0.45

*Each 2.5 kg of vitamins and minerals mixture contains:12000.000 IU vitamin A acetate; 2000.000 IU vitamin D3; 10.000 mg vitamin E acetate; 2000 mg vitamin K3; 100 mg vitamin B1; 4000 mg vitamin B2; 1500 mg vitamin B6; 10 mg vitamin B12; 10.000 mg pantothenic acid; 20.000 mg Nicotinic acid; 1000 mg Folic acid; 50 mg Biotin; 500.000 mg chorine; 10.000 mg Copper; 1000 mg Iodine; 300.00 mg Iron; 55.000 mg Manganese; 55.000 mg Zinc, and 100 mg Selenium.

Live body weight and feed consumption were weekly recorded. Weight gain and feed conversion were calculated and expressed in gram per chicks at the whole period of the experiment (from 10 to 42 days of age) according to the formula suggested by Broody, (1949).

Five birds for each experimental group were randomly chosen at the 42^{th} day of age for the determination of carcass traits. Birds chosen were deprived from feed for 4 hours before slaughtering after that they were individually weighed alive to the nearest gram. Slaughtering was performed according to recommended Islamic tradition. Birds were individually reweighed after complete bleeding. Shank and head were separately weighed. The proportional weights of giblets (empty gizzard, liver and heart) and total edible parts to live weight were calculated.

Mortality rate was recorded during the whole time of the experiment then expressed as percentage of total number of dead birds to the initial number of birds at the beginning of the experiment. The economic efficiency (EE) was calculated according to the cost of one-kg feed (included the price of feed additives) and the prices of one kg body gain during the same time of experiment.

Under aseptic conditions, small intestine contents for individual birds of each group were picked up for the microbial counts. Five birds were randomly selected to enumerate of microbial populations in small intestine (duodenum, jejunum, and ileum) of quail by using various dehydrated media (Biolife Ltd, Italy). The media were prepared and sterilized according to the manufacturer's instructions before being poured into sterile Petri dishes. The used media included tryptic soy agar (for total microbial count), Sabouradu dextrose agar (for cultivation and isolation of yeasts and molds), Pseudomonas selective agar (selective medium for isolation of *P. aeruginosa*), giolitti-cantoni agar with potassium tellurite 1% (for enumeration of *Staphylococcus aureus*)Mac Conkey agar OMS W/O CV (selective and differential medium for detection of coliforms and enteric pathogens) and SS agar (for selective medium for isolation of *Salmonella*spp and *Shigella*spp).Enumeration of microbial populations by plate count methods was carried out according to American Public Health Association, 1993.

Data were statistically analyzed by using SAS procedure guide (SAS, 2004), analysis of variance and least square means as well as Duncan new multiple range test for all of experimental groups.

Results and Discussion

Effect of essential oils on pathogenic microorganisms under laboratory condition.

Antimicrobial potential of essential oils were assessed in terms of inhibition zone expressed as centimeter for microbial growth. The results of the antimicrobial activity are illustrated in Fig (1).

The antimicrobial activity of tested essential oils increased with the increasing of concentration (µg/ml). Results revealed that *Y. enterocolitica*, *P. aeruginosa*, *L. moncytogenes* and *C. albicans* were more susceptible for essential oils than *Staph. aureus* and *S. enteritidis*.

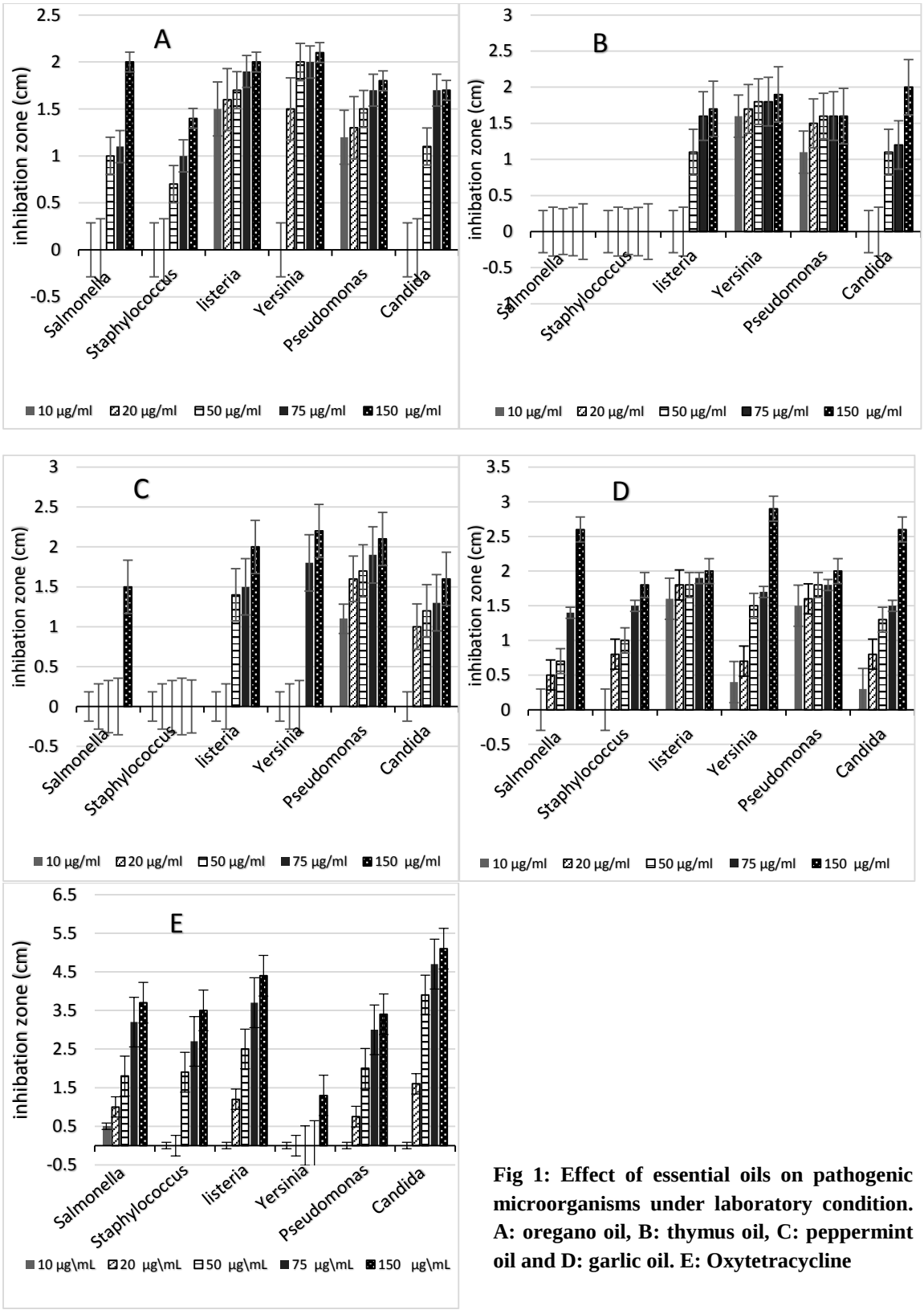

Fig 1: Effect of essential oils on pathogenic microorganisms under laboratory condition. A: oregano oil, B: thymus oil, C: peppermint oil and D: garlic oil. E: Oxytetracycline

Compared with thyme and peppermint, oregano and garlic essential oil were found to be more effective against all tested microorganisms. The minimum concentration of garlic essential oil which inhibits all tested microorganisms was 20 µg/ml.While the minimum concentrations oforegano essential oil and oxytetracycline were50 µg/ml and 150 µg/ml, respectively.

In vitro, several of studies have demonstrated that oregano essential oil eliminate pathogenic bacteria, among them *E. coli*, *Salmonella* spp, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Staphylococcus epidermis*, *Klebsiellapneumoniae*,

*Shigella*spp.,*Proteus vulgaris*, and *Bacillus cereus* (Hammer *et al*., 1999; Inouye *et al*., 2003; Azaz *et al.,* 2004; Bagamboula *et al*., 2004; Penalver *et al*., 2005; Kędzia and Hołderna-Kędzia, 2007)

Garlic has been shown several effects include antimicrobial activity (Iwalokun *et al*., 2004; Corzo-Martínez *et al.,* 2007; Onibi *et al*., 2009) where it contains compounds that inhibit *B. subtilis, Serratiamarcescens, Mycobacterium*spp, *B. cereus, Clostridium botulinum* type A, *E. coli, Lactobacillus plantarum, Leuconostocmesenteroides, Salmonella*spp, *Shigella* sppand *S. aureus*as well as*Aspergillusflavus, Aspergillusparasiticus, Candida albicans, Cryptococcus*sp, *Penicillium*sp, *Rhodotorula* spp are also inhibited.

Body weight and body weight gain

Quail chicks fed on diets supplemented with oregano (150 mg/kg) combined with 0.5g/kgoxytetracycline (T5) had highly significant increase in average of live body weight and weight gain at the whole time of the experiment followed by chicks fed on diet supplementedwith garlic powder level of 0.2 g/kg combined with 0.5g/k oxytetracycline (T4) compared with anther applied treatments (Table, 3). The improving in body weight and body weight gain of quail chicks due to addition of oregano essential oil to feed diet could be attributed to their ability to destroy pathogens in the digestive system and consequently increasing the production of digestive enzymes which improve utilization of digestive products (Hernández *etal*., 2004). Obtained results of this study are agreed with those reported by Alcicek*et al*., 2003; Ertas*et al*., 2005; Jamrozet *al*. 2005; Bozkurt*et al*., 2009 and Abdel-warth*et al*., 2012who found animprovement in body weight and body weight gain of broilers fed on diets supplemented with medicinal plant extracts. Concerning the addition effect ofgarlic powder levels in this study it was relatively similar to those reported by Raeesi*et al.* (2010) who found that broiler chicks fed on diets supplemented with 1 and 3% garlic powder have significant increasein body weight gain.

Feed consumption and conversion

Data presented in Table (4) revealed that no significant effect was found in feed consumption of quail birds due to applied treatments at all times of estimation, however, the total feed consumption at the whole period of experiment was significantly increased in treated chicks compared with control. This result is agreed with those obtained by Çabuk *et al.,*(2014)who indicated that herbs, plant extracts did not affect feed intake in broilerswhereas feed conversion ratiowas significantly improved by essential oil mixture and antibioticssupplementation. While highly significant variation was found in feed conversion due to applied treatments. Chicks fed on diets supplemented with oregano at level of 150 mg/kg combined with 0.5g/kg oxytetracycline (T5) showed the highest significant improvement in total feed conversion at the whole time of the experiment (2.01) followed by those fed on diet supplemented by level of 0.5g/kg oxytetracycline only (T2). These results are in agreement with those obtained by (Ertas *et al*., 2005 and Abdel-warth *et al*., 2012) who reported that addition of essential oils extracted from oregano, clove and anise improved feed conversion ratio compared to the control in broiler. Alp *et al*(2012)stated that dietary oregano essential oil and

Table 3. Least squares means and standard error (LSM ± S.E) for body weight (g) of birds of different experimental groups as affected by applied treatments.

Treatments	Body weight (g) at age of						Total body gain (g) at 10-42days
	10days	17days	21days	28days	35days	42days	
T1	58.51±0.81	83.60^{d}±4.77	109.60^{e}±7.45	148.28^{e}±7.58	186.10^{c}±8.29	219.34^{e}±7.29	160.82^{e}±7.17
T2	59.75±0.81	116.73^{c}±4.77	149.07^{cd}±7.45	184.08^{cd}±7.58	226.20^{bc}±8.29	256.99^{bc}±7.29	197.24^{bc}±7.17
T3	58.70±0.81	93.89^{d}±4.77	126.44^{de}±7.45	160.81^{de}±7.58	198.06^{c}±8.29	233.44^{de}±7.29	174.74^{cde}±7.17
T4	59.17±0.81	123.53^{c}±4.77	156.18^{bc}±7.45	198.08^{bc}±7.58	222.97^{b}±8.29	247.88^{cd}±7.29	188.71^{cd}±7.17
T5	59.43±0.81	157.65^{a}±4.77	185.57^{a}±7.45	227.16^{a}±7.58	275.65^{a}±8.29	308.66^{a}±7.29	249.23^{a}±7.17
T6	58.75±0.81	98.48^{d}±4.77	129.23^{de}±7.45	162.10^{de}±7.58	196.32^{c}±8.29	230.65^{de}±7.29	171.89^{de}±7.17
T7	59.90±0.81	139.28^{b}±4.77	175.14^{ab}±7.45	215.4^{ab}±7.58	258.35^{a}±8.29	272.72^{b}±7.29	212.82^{b}±7.17
T8	59.64±0.81	88.80^{d}±4.77	116.87^{e}±7.45	157.10^{e}±7.58	195.47^{c}±8.29	233.22^{de}±7.29	173.58^{de}±7.17

Where; Means with the same letters in each column are not significantly differed.
For more details of T1-T8 see Table 1

Table 4. Feed consumption and feed conversion of birds for different experimental groups as affected by applied treatments

Treatments	Feed consumption(g/bird/day)						Feed conversion (g feed/ g body gain) at 10-42days
	10-17days	17-21days	21-28days	28-35days	35-42days	Total (10-42days)	
T1	6.15±0.63	8.24±0.44	12.55±0.44	16.25±0.44	20.62±0.63	64.83^{b}±2.42	2.83^{a}±0.13
T2	6.12±0.63	8.21±0.44	12.52±0.44	16.22±0.44	20.59±0.63	65.65^{ab}±2.42	2.33^{bc}±0.13
T3	6.40± 0.63	8.16±0.44	12.47±0.44	16.17±0.44	20.87±0.63	67.08^{ab}±2.42	2.68^{ab}±0.13
T4	7.30±0.63	9.06±0.44	13.37±0.44	17.07±0.44	21.77±0.63	72.58^{ab}±2.42	2.69^{ab}±0.13
T5	7.26±0.63	8.35±0.44	12.66±0.44	16.36±0.44	21.73±0.63	71.36^{ab}±2.42	2.01^{c}±0.13
T6	7.14±0.63	8.89±0.44	13.20±0.44	16.90±0.44	21.61±0.63	73.74^{a}±2.42	3.02^{a}±0.13
T7	6.32±0.63	8.41±0.44	12.72±0.44	16.42±0.44	20.79±0.63	71.64^{ab}±2.42	2.38^{bc}±0.13
T8	5.86±0.63	7.95±0.44	12.26±0.44	15.96±0.44	20.33±0.63	70.35^{ab}±2.42	2.84^{a}±0.13

Means with the same letters in each column are not significantly differed

anticoccidial drugs supplementation significantly improved the FCR ($P < 0.05$) for broiler chicken from 21 to 42 days and 1 to 42 days of age, respectively, compared with the negative control diet. However,Fadlalla*et al*., (2010) showed that birds fed 0.3% garlic basal diet had significantly ($p<0.05$) lower total feed intake compared to other dietary treatments.

Carcass traits

Data presented in Table (5) showed significant variations in both proportion and absolute weight of carcass, giblets and total edible weight. It was clearly observed that chicks fed on diets supplemented with levels of either essential oregano oil or garlic powder combined with oxytetracycline as well as those fed on diets supplemented with oxytetracycline only had improved carcass traits compared with control one. In addition, chicks fed on diet supplemented with oregano (150 mg/kg)combined with 0.5g/k oxytetracycline (T5)had the highest absolute and proportion weights of carcass, giblets and total edibleweight followed by those fed on diets supplemented withgarlic powder by level of 0.2 g/kg combined with antibiotic level (0.5g/k) (T7) as well as those fed on diet supplemented with oxytetracycline level only (1.0 g/kg) (T2) compared with control (T1) (Table, 5). These results are agree with those reported by Alçiçek*et al*. (2003) who showed that supplementing essential oils at a concentration of 48 or 72 mg of an essential oil combination/kg of feed significantly improved carcass yield in broiler chicken. However, it disagree with those reported by Alp *et al*. (2012) who found that dietary treatment with oregano oil or the coccidiostatdrug had no effect on pre-slaughter weight, carcass weight, or carcass yield of broiler chicken.

Table 5. Least square means and standard error (LSM ± S.E) for Averages of absolute and proportional weights of carcass, giblets and total edible parts as affected by applied treatments.

Treatments	Live body weight(g)	Carcass weight		Giblet weight		Total edible weight	
		g	%	g	%	g	%
T1	219.9^{e}±3.6	142.9e±3.8	65.0f±0.51	7.4de±0.20	3.4abc±0.09	150.3e±3.82	68.4f±0.49
T2	260.7c±3.6	185.1c±3.8	71.0c±0.51	9.5b±0.20	3.7a±0.09	194.6c±3.82	74.6c±0.49
T3	240.3d±3.6	163.2d±3.8	67.9de±0.51	7.9cd±0.20	3.3bc±0.09	171.2d±3.82	71.2e±0.49
T4	253.1c±3.6	175.8c±3.1	69.4d±0.51	8.4c±0.20	3.3bc±0.09	184.2c±3.82	72.8d±0.49
T5	308.2a±3.6	233.1a±3.8	75.6a±0.51	10.5a±0.20	3.4ab±0.09	243.6a±3.82	79.1a±0.49
T6	231.9d±3.6	155.1d±3.8	66.9e±0.51	7.2e±0.20	3.1c±0.09	162.3d±3.82	70.0e±0.49
T7	278.6b±3.6	203.2b±3.8	72.8b±0.51	9.2b±0.20	3.3bc±0.09	212.5b±3.82	76.2b±0.49
T8	237.6d±3.6	161.6d±3.8	68.0de±0.51	7.6de±0.20	3.2bc±0.09	169.2d±3.82	71.2e±0.49

Means with the same letters in each column are not significantly differed

Table (6): Least square means and standard error (LSM ± S.E) for Mortality and economic efficiency (%) as affected by applied treatments.

Treatments	Mortality (%)	Economic efficiency (%)
T1	8.33^{a}±1.18	68.72^{c} ±2.96
T2	0.00^{c}	82.53^{ab}±2.96
T3	3.33^{bc}±1.18	74.98^{bc}±2.96
T4	3.33^{bc}±1.18	75.09^{bc}±2.96
T5	0.00^{c}	90.74^{a} ±2.96
T6	3.33^{bc} ±1.18	70.92^{c} ±2.96
T7	0.00^{c}	83.15^{ab}±2.96
T8	5.00^{ab}±1.18	73.78^{bc}±2.96

Means with the same letters in each column are not significantly differed.

5-Mortality rate

Generally, data in Table (6) showed that chicks fed on diets supplemented with either oregano essential oil or garlic powder recorded the lower mortality rate compared to the control group. The decrease in mortality rate in the treated groups could be attributed to the antimicrobial activity of both the oregano essential oil and garlic powder with complimentary effect of oxytetracycline that caused a reduction in colonization of pathogens in the gastrointestinal tract. These results are partial agreement with those reported by Alp*et al*. (2012) who found that broiler chicken receiving no dietary additives (negative control) had the highest mortality rate (11.25%), and birds receiving the coccidiostat drug had the lowest (4.0%), whereas this rate was intermediate for birds receiving the oregano oil (8.25%)at 42 days.

Economic efficiency

Obtained results in Table (6) demonstrated thatchicks fed on diets supplemented with different levels of essential oregano oil and garlic powder integrated with antibiotic showed highly significant effect on the economic efficiency percentage compared with control. As the same trend, chicks fed on diet supplemented with oregano 150 mg/kg combined with 0.5g /kg oxytetracycline (T5) recorded the highest economic efficiency (90.74%) followed by those fed on diet supplemented with garlic powder level of 0.2 g/kg combined with 0.5g /kg oxytetracycline (T7) (83.15%). However, chicks of control group that fed on basal diet had the lowest value of economic efficiency (68.72%). This improvement could be due to improving in either the feed utilization or reducing the amount of feed required to produce body weight gain as well as the decreasing in mortality percentage.

Microbial population in small intestine content of quail fed on diets supplemented with the oregano essential oil and garlic powder

Results in Table (7) showed that the diets supplementation with garlic or oregano oil combined with 0.5 g/kgoxytetracyclinedecreased thetotal microbial and pathogens count in small intestine compared with supplementation of oxytetracycline only.

Increasing the concentration of garlic or oregano supplementation led to increase their effect. The supplementation of garlic was more effective on pathogenic population than oregano oil.

Obtained data showed that supplementing diet with oregano or garlic led to Yeasts & molds disappear from small intestine content compared with control. Regarding the effect of oregano and garlic levels on pathogens, it is clearly found that using oregano or garlic with level of 150 mg/kg and 0.3 g/kg,respectively decreased bacterial total count and enumeration of *P. aeruginosa*, *Staph. aureus* and coliform group. However,*Salmonella*spp and*Shigella*spp werenot detected.This result is consistent with those obtained under laboratory condition illustrated in Fig 1.

The antimicrobial activity of essential oil caused a reduction in pathogens colonization in the gastrointestinal tract. (Apajalahti*et al*., 2001and Apajalahti*et al*., 2004). Many essential oils stimulate the growth of beneficial microorganisms and limit the number of pathogenic bacteria in poultry (Wenk, 2000).

Table 7. Microbial counts (log cfu/g) insmall intestine content of Japanese quail at 42 days of age.

Treatments		Microbial populations in samples from the intestinal contents of broilers (log cfu/g)					
		Microbial total count	Yeasts & molds	*Pseudo. aeruginosa*	*Staph. aureus*	Coliform group	*Salmonella &Shigella*
T1	duodenum	9.0 ^{bc}	5.7 ^{b}	5.03 ^{bc}	5.8 ^{ab}	6.8 ^{b}	4.5 ^{ab}
	Jejunum	9.3 ^{b}	4.0 ^{c}	5.06^{b}	5.7 ^{ab}	7.0 ^{b}	4.6 ^{a}
	Ilium	9.9 ^{a}	5.9 ^{a}	5.64 ^{a}	5.9 ^{a}	7.4 ^{a}	4.6 ^{a}
T2	duodenum	9.01 ^{bcd}	ND ^{d}	4.85 ^{bcde}	5.4 ^{cd}	6.5 ^{cd}	4.2 ^{bcd}
	Jejunum	9.0 ^{cde}	ND ^{d}	4.91 ^{bcde}	5.5 ^{bcd}	6.7 ^{bc}	4.2 ^{bcd}
	Ilium	9.03 ^{cde}	5.0 ^{c}	4.97 ^{bcd}	5.6 ^{bc}	6.8 ^{b}	4.4 ^{abc}
T3	duodenum	8.7 ^{cdefg}	ND ^{d}	4.80 ^{bcdef}	5.3 ^{cde}	6.4 ^{cd}	4.1 ^{de}
	Jejunum	8.8 ^{cdef}	ND ^{d}	4.81 ^{bcdef}	5.3 ^{cde}	6.4 ^{cd}	4.1 ^{cde}
	Ilium	8.9 ^{cdef}	ND ^{d}	4.82 ^{bcdef}	5.3 ^{cde}	6.5 ^{cd}	4.2 ^{bcde}
T4	duodenum	8.6 ^{efghi}	ND ^{d}	4.66 ^{cdefg}	5.0 ^{efg}	6.0 ^{efg}	3.9 ^{efg}
	Jejunum	8.6 ^{fghij}	ND ^{d}	4.65 ^{defg}	4.8 ^{fgh}	5.9 ^{efgh}	3.7 ^{fgh}
	Ilium	8.4 ^{ghijk}	ND ^{d}	4.54 ^{efgh}	4.7 ^{gh}	5.8 ^{fgh}	3.6 ^{ghi}
T5	duodenum	8.11 ^{kl}	ND ^{d}	4.28 ^{hi}	4.5 ^{hi}	5.5 ^{i}	3.3 ^{i}
	Jejunum	8.0 ^{kl}	ND ^{d}	4.18 ^{i}	4.5 ^{hi}	2.3 ^{j}	ND ^{j}
	Ilium	8.0 ^{kl}	ND ^{d}	4.15 ^{i}	4.3 ^{ij}	2.25 ^{k}	ND ^{j}
T6	duodenum	8.63 ^{cdefg}	ND ^{d}	4.81 ^{bcdef}	5.1 ^{def}	6.3 ^{d}	4.0 ^{def}
	Jejunum	8.62 ^{defgh}	ND ^{d}	4.74 ^{bcdef}	5.1 ^{def}	6.2 ^{de}	4.0 ^{def}
	Ilium	8.59 ^{defghi}	ND ^{d}	4.69 ^{cdefg}	5.1 ^{def}	6.1 ^{def}	3.9 ^{def}
T7	duodenum	8.2 ^{jkl}	ND ^{d}	4.34 ^{ghi}	4.6 ^{hi}	5.6 ^{hi}	3.3 ^{i}
	Jejunum	8.3 ^{ijkl}	ND ^{d}	4.48 ^{fghi}	4.6 ^{hi}	5.7 ^{ghi}	3.5 ^{hi}
	Ilium	8.3 ^{hijk}	ND ^{d}	4.49 ^{fghi}	4.6 ^{hi}	5.8 ^{fgh}	3.5 ^{hi}
T8	duodenum	7.0 ^{n}	ND ^{d}	3.0 ^{j}	ND ^{k}	0.66 ^{m}	ND ^{j}
	Jejunum	7.5 ^{m}	ND ^{d}	ND ^{k}	4.0 ^{j}	1.94 ^{l}	ND ^{j}
	Ilium	7.9 ^{l}	ND ^{d}	ND ^{k}	4.0 ^{j}	2.43 ^{j}	ND ^{j}

Means with the same letters in each column are not significantly differed

Conclusion

From the economic point of view, it could be concluded that using the basal diet supplemented with150 mg/kg oregano combined with 0.5g/kg oxytetracycline (T5) or 0.2 g/kg garlic powder combined with 0.5g/kg oxytetracycline (T7)for feeding of quail birds seemed to be adequate to achieve the favorable results. Sothe diets supplementationwith medicinal plants or their essential oils combined with the half subtherapeutic dose of antibiotic was recommended to reduce unsusceptible antibiotic bacteria and promote the growth performanceof birds.

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References

Abdel-Wareth A.A., Kehraus S., Hippenstiel F., Südekum K., 2012. Effects of thyme and oregano on growth performance of broilers from 4 to 42 days of age and on microbial counts in crop, small intestine and caecum of 42-day-old broilers. Animal Feed Science and Technology 178: 198– 202.

Alçiçek A., Bozkurt M. and Çabuk M., 2003. The effect of essential oil combination derived from selected herbs growing wild in Turkey on broiler performance. S. Afr. J. Anim. Sci. 33: 89–94.

Alp M., Midilli M., Kocabağlı N., Yılmaz H., Turan N., Gargılı A. and Acar N., 2012. The effects of dietary oregano essential oil on live performance, carcass yield, serum immunoglobulin G level, and oocyst count in broilers. J. Appl. Poult. Res. 21:630–636.

American Public Health Association 1993. Standard Methods for the Examination of Dairy Products, 16th ed. APHA, Washington, DC.

Aouadi R., Aouidet A., Elkadhi A., Ben Rayana M.C., Jaafoura H., Tritar B.and Nagati K.,2000. Effect of garlic (Allium sativum) on lipid metabolism in male rats. Nutr. Res., 20: 273-280.

Apajalahti J. H., Kettunen A., Bedford M. R., and. Holben W. E. 2001. Percent G + C profiling accurately reveals diet-related differences in the gastrointestinal microbial community of broiler chickens. Appl. Environ. Microbiol. 67:5656–5667.

Apajalahti J., Kettunen A., and Graham H. 2004. Characteristics of the gastrointestinal microbial communities, with special reference to the chicken. World's Poult. Sci. J. 60:223–232.

Apitz-Castro R., Cabrera S., Cruz M.R., Ledezma E. and Jain M.K., 1983. Effects of garlic extract and of three pure components isolated from it on human platelet aggregation, arachidonate metabolism, release reaction and platelet ultrastructure. Thromb. Res. 32: 155–169.

Azaz A.D., Irtem H.A., Kurkcuoğlu M., and Baser K.H.C., 2004. Composition and the in vitro antimicrobial activities of the essential oils of some Thymus species. Z. FürNaturforschung C J. Biosci. 59: 75–80.

Bagamboula C., Uyttendaele M. and Debevere J., 2004. Inhibitory effect of thyme and basil essential oils, carvacrol, thymol, estragol, linalool and p-cymene towards Shigella sonnei and S-flexneri. FOOD Microbiol. 21: 33–42.

Basmacioğlu H., Tokuşoğlu Ö. and Ergül M., 2004. The effect of oregano and rosemary essential oils or α-tocopheryl acetate on performance and lipid oxidation of meat enriched with n-3 PUFAs in broilers. Afr J AnimSci 34: 197–210.

Brody S., 1945. Bioenergetics and growth, with special reference to the efficiency complex in domestic animals (Oxford, England: Reinhold).

Burt S., 2004. Essential oils: their antibacterial properties and potential applications in foods--a review. Int. J. Food Microbiol. 94: 223–253.

Çabuk M., Eratak S., Alçicek, A., and Bozkurt M. 2014. Effects of Herbal Essential Oil Mixture as a Dietary Supplement on Egg Production in Quail. Sci. World J. 2014, 1–4.

Chang K.J. and Cheong S.H., 2008. Volatile organosulfur and nutrient compounds from garlic by cultivating areas and processing methods. FASEB J. 22: 1108–2.

Chowdhury S. R., Chowdhury S. D. and Smith T. K., 2002. Effects of dietary garlic on cholesterol metabolism in laying hens. Poult. Sci. 81: 1856 –1862.

Corzo-Martínez M., Corzo N., and Villamiel M., 2007. Biological properties of onions and garlic. Trends Food Sci. Technol. 18: 609–625.

Dorhoi A., Dobrean V., Zăhan M. and Virag P., 2006. Modulatory effects of several herbal extracts on avian peripheral blood cell immune responses. Phytother. Res. 20: 352–358.

Ertas O. N., Guler T., Ciftci M., Dalkilic B., and Yılmaz O. 2005. The effect of a dietary supplement coriander seeds on the fatty acid composition of breast muscle in Japanese quail. Review Med. Vet. 156:514–518.

Fadlalla I.M.T., Mohammed B.H., Bakhiet A.O. 2010. Effect of feeding garlic on the performance and immunity of broilers. Asian J. Poult. Sci., 4: 182-189.

Florou-Paneri P., Giannenas I., Christaki E., Govaris A. and Botsoglou N., 2006. Performance of chickens and oxidative stability of the produced meat as affected by feed supplementation with oregano, vitamin C, vitamin E and their combinations. Arch. Geflugelkunde 70: 232–240.

Fukayama E.H., Bertechini A.G., Geraldo A., Kato R.K. and Murgas L.D.S., 2005. Oregano extract as an additive in the broiler diet. R Bras Zootec 34: 2316–2326.

Giannenas I., Florou-Paneri P., Papazahariadou M., Christaki E., Botsoglou N.A. and Spais A.B., 2003. Effect of dietary supplementation with oregano essential oil on performance of broilers after experimental infection with Eimeriatenella. Arch. Anim. Nutr. 57: 99–106.

Giannenas I.A., Florou-Paneri P., Papazahariadou M., Botsoglou N.A., Christaki E. and Spais A.B., 2004. Effect of diet supplementation with ground oregano on performance of broiler chickens challenged with Eimeriatenella. Arch. Geflugelkunde 68: 247–252.

Halle I., Schubert R., Flachowsky G., Jahreis G., Bitsch R. and *et al*, 2001. Effects of essential oils and herbal mixtures on the growth of broiler chicks. In Vitamine Und Zusatzstoffe in Der Ernährung von Mensch Und Tier. 8. Symposium, 26. Und 27. September, 2001, Jena/Thüringen, Germany. (Friedrich-Schiller-Universität), pp. 439–442.

Hammer K.A., Carson C.F. and Riley T.V., 1999. Antimicrobial activity of essential oils and other plant extracts. J. Appl. Microbiol. 86: 985–990.

Hernandez F., Madrid J., Garcia V.,Orengo J. and Megias M.D., 2004. Influence of two plant

extracts on broilers performance, digestibility, and digestive organ size. Poult. Sci. 83: 169–174.

Inouye S., Abe S., Yamaguchi H. and Asakura M., 2003. Comparative study of antimicrobial and cytotoxic effects of selected essential oils by gaseous and solution contacts. Int. J. Aromather. 13: 33–41.

Iwalokun B.A., Ogunledun A., Ogbolu D.O., Bamiro S.B. and Jimi-Omojola J., 2004. In vitro antimicrobial properties of aqueous garlic extract against multidrug-resistant bacteria and Candida species from Nigeria. J. Med. Food 7: 327–333.

Jamroz D., Wiliczkiewicz A., Wertelecki T., Orda J., and Skorupińska J. 2005. Use of active substances of plant origin in chicken diets based on maize and locally grown cereals. Br. Poult. Sci. 46:485–493.

Kalemba D. and Kunicka A., 2003. Antibacterial and Antifungal Properties of Essential Oils. Curr. Med. Chem. 10: 813–829.

Kędzia B., and Hołderna-Kędzia E. 2007. Studies on effect of volatile oils on pathogenic bacteria, yeast fungi and dermatophytes. Post. Fitoter. 2:71–77.

Kim S.G., Nam S.Y., Chung H.C., Hong S.Y. and Jung K.H., 1995. Enhanced Effectiveness of Dimethyl-4, 4 "-dimethoxy-5, 6, 5", 6 "-dimethylene dioxybiphenyl-2, 2"-dicarboxylate in Combination with Garlic Oil against Experimental Hepatic Injury in Rats and Mice. J. Pharm. Pharmacol. 47: 678–682.

Kim Y.J., Jin S.K. and Yang H.S., 2009. Effect of dietary garlic bulb and husk on the physicochemical properties of chicken meat. Poult. Sci. 88: 398–405.

Lambert R.J.W., Skandamis P.N., Coote P.J. and Nychas G.J., 2001. A study of the minimum inhibitory concentration and mode of action of oregano essential oil, thymol and carvacrol. J. Appl. Microbiol. 91: 453–462.

Lee K. W., Everts H., Kappert H.J., Frehner M., Losa R. and Beynen A.C., 2003. Effects of dietary essential oil components on growth performance, digestive enzymes and lipid metabolism in female broiler chickens. Br. Poult. Sci. 44: 450–457.

Lee K.W., Everts H. and Beynen A.C., 2004. Essential oils in broiler nutrition. Int. J. Poult. Sci., 3: 738–752.

Lihua C., Ying Y., Yifu L. and Lei C., 2007. Effects of oregano oil on growth performance and carcass quality of broilers. China Poult 29: 9–11.

McMahon F.G. and Vargas R., 1993. Can garlic lower blood pressure? A pilot study. Pharmacother. J. Hum. Pharmacol. Drug Ther. 13: 406–407.

Mitsch P., Zitterl-Eglseer K., Köhler B., Gabler C., Losa R. and Zimpernik I., 2004. The effect of two different blends of essential oil components on the proliferation of Clostridium perfringens in the intestines of broiler chickens. Poult. Sci. 83: 669–675.

Modeva T. and Profirov Y., 2003. Influence of the oregano etheric oil on the weight gain and some blood biochemical indices in chickens. ZhivotnovdniNauki Bulg.

Moyers S.B., 1996. Garlic in health, history, and world cuisine (Suncoast Press).

NRC., 1994. Nutrient Requirements of Poultry. 9th rev. ed. Natl. Acad. Press, Washington, DC.Whey permeate containing 80% lactose was obtained from Berner Foods Inc (Roscoe, IL).

Onibi G.E.,Adebisi O.E.,Fajemisin A.N.,Adetunji A.V. and *et al*, 2009. Response of broiler chickens in terms of performance and meat quality to garlic (Allium sativum) supplementation. Afr. J. Agric. Res. 4: 511–517.

Penalver P., Huerta B., Borge C., Astorga R., Romero R. and Perea A., 2005. Antimicrobial activity of five essential oils against origin strains of the Enterobacteriaceae family. Apmis 113: 1–6.

Raeesi M., Hoeini-Aliabad S.A., Roofchaee A., Zare Shahneh A., Pirali S. 2010. Effect of periodically use of garlic (allium sativum) powder on performance and carcass characteristics in broiler chickens. World Academy of Science, Engine. Techno. 68; 1213-1219.

Sallam K.I., Ishioroshi M. and Samejima K., 2004. Antioxidant and antimicrobial effects of garlic in chicken sausage. LWT-Food Sci. Technol. 37: 849–855.

SAS, 2004. SAS/STAT guide for personal computers (SAS Institute).

Sumiyoshi H., 1997. New pharmacological activities of garlic and its constituents. Nihon Yakurigaku Zasshi Folia Pharmacol. Jpn. 110: 93 – 97.

Tang X. and Cronin D.A., 2007. The effects of brined onion extracts on lipid oxidation and sensory quality in refrigerated cooked turkey breast rolls during storage. Food Chem. 100: 712–718.

Wenk C. 2000. Recent advances in animal feed additives such as metabolic modifiers, antimicrobial agents, probiotics, enzymes and highly available minerals. Rev. Asian-australas. J. Anim. Sci. 13:86–95.

Williams P. and Losa R., 2002. Blending essential oils for poultry. Feed Mix 10, 8–9.

Yin M. and Cheng W., 2003. Antioxidant and antimicrobial effects of four garlic-derived organosulfur compounds in ground beef. Meat Sci. 63: 23–28.

التأثير المتكامل بين الأوكسي تتراسيكلين والزيت العطري للبردقوش و مسحوق الثوم على المحتوي الميكروبي للأمعاء والأداء الإنتاجي في السمان الياباني

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تم اختبار أربعة من الزيوت العطرية (البردقوش والزعتر والنعناع والثوم) معمليا لتقييم نشاطها المضاد ضد بعض سلالات الكائنات الحية الدقيقة المرضية بتركيزات 10، 20، 50، 75، و150 ميكروجرام / مل. وقد أظهرت النتائج المعملية أن كلٍ من الزيت العطري للبردقوس والثوم كانت أكثرها فعالية ضد كل المسببات المرضية المختبرة. ونتيجة لذلك فقد تم اختيارزيت البردقوش العطري ومسحوق الثوم كإضافة غذائية في علائق السمان. وفي تجربة تطبيقية فقد تم اختيار عدد 480 من طيور السمان الياباني عمر 10 أيام عشوائيا وتم تقسيمها إلى ثمانية مجموعات تجريبية كل منها 60 طائر (3 مكررات كل مكررة 20 طائر). المجموعة الأولى غذيت على عليقة أساسية بدون أي إضافات (معاملة مقارنة). المجموعة الثانية غذيت على عليقة مضاف اليها 1.0 جم / كجم من المضاد الحيوي أوكسيتتراسيكلين. وغذيت طيور المجموعة الثالثة والرابعة والخامسة على علائق مضاف اليها الزيت العطري للبردقوش بتركيزات 50 و 100 و 150 ملجم / كجم بالإضافة الي 0.5جم / كجم من المضاد الحيوي أوكسيتتراسيكلين، على التوالي. في حين غذيت طيور المجموعة السادسة والسابعة والثامنة على علائق مضاف اليها مسحوق الثوم بتركيزات 0.1، 0.2 و 0.3 جم / كجم بالإضافةإلي 0.5جم/كجم من المضاد الحيوي أوكسيتتراسيكلين على التوالي. وأظهرت النتائج التي تم التوصل اليها أن تغذية السمان على عليقة مزودةبالزيت العطري للبردقوش بمعدل 150 ملجم / كجم أدي الي تحسن كبير في وزن الجسم، الزيادة المكتسبة في وزن الجسم، معامل التحويل الغذائي، وزن الذبيحة والكفاءة الاقتصادية.علاوة على ذلك فقد انخفض محتوي الأمعاء من المسببات المرضية وانخفضت نسبة النفوق، وقد تلي هذه المعاملة من حيث الاهمية المعاملة التي تغذى فيها السمان على علائق تحتوي على 0.2جم / كجم من مسحوق الثوم.