

## Advanced immunological effect of some herbal plants in Broilers

Abdalla, O.A., El-Boshy, M.E\*, Omnia E.E, Rasha M.I and Verginia M Farag\*

*Dept. of clinical pathology, Fac of vet. Med. Suez Canal Univeristy.*

*\*Dept. of clinical pathology, Fac of vet. Med. Mansoura Univeristy.*

### Abstract

One Hundred chicks, one day old were divided into 10 groups. The first group was control negative and the second was control positive. The other eight groups treated either by *Thymus vulgaris* (1 or 2 ml/ 1 litter drinking water) or *Zingiber officinale* (5 or 10 ml/ 1 litter drinking water) from the first day old till the end of the experiment. Experimental infection was done orally to 4 groups only of the 8 groups at 10 days

Growth parameters as body weights and feed conversion rates were the best in groups treated only with *Thymus vulgaris* or *Zingiber officinale* and higher doses were better than lower doses. Also, infected treated groups with higher doses of either *Thymus vulgaris* or *Zingiber officinale* extract were better than infected treated groups with low doses of either *Thymus vulgaris* or *Zingiber officinale* extract or infected only with the *E.coli* O78 strain.

Blood leukogram pictures and advanced immunological (IL-1 beta, 6, 10, 12 and Tumor necrosis factor) profiles showed better results with higher doses of either *Thymus vulgaris* or *Zingiber officinale* extract in compare to lower doses of the same extract either infected with *E.coli* O78 or not infected.

The experimentally infected groups showed better results in compare to control positive.

### Introduction

Supplementing broiler diets with some feed additives may be considered as an alternative to improve growth and feed conversion efficiency attempts have been made to use medicinal plants not only serve as a medical purpose but also contain aromatic substances and essential oils which are widely used in feed industries

(Heitzman, 1986; Khachatourians, 1998, Wary and Davies, 2000),.

One possible mechanism of how medicinal plants work is by improving broilers growth and feed utilization through the improvement of nutrients digestibility. This is through facilitating absorption of calorogenic nutrients across the gut wall by increasing its absorption capacity (Nelson et al, 1963). An improvement in nutrients

digestibility of broiler diets using medicinal plants such as thymus vulgaris and zingiber officinale (Abaza 2001, Al-Harathi 2002 and El-Husseiny et al, 2002).

Enteritis caused by *Escherichia coli* (colibacilliosis) is an important disease in the poultry industry because of increasing mortality and decreasing performance. *E. coli* is a normal inhabitant of the intestinal tracts of animals and is harmless as long as it is kept in check by other intestinal bacteria but when an imbalance occurs in bacterial flora of the intestinal tract of chicken at any age, *E. coli* may grow and cause an outbreak of colibacilliosis (Barnes et al, 2003).

The present experiment aimed to study effect of oral supplementation of herbal plants (*Thymus vulgaris* and *Zingiber officinale*) on normal and experimentally infected (by *E.Coli*) broiler chickens through evaluation of growth indices, leukogram as well as immunological parameters (in serum through evaluation of serum TNF- $\alpha$ , IL-1B, IL-6, IL-10 and IL-12).

### Materials and Methods

This experiment was conducted on one hundred (100) normal healthy Cobb broiler chicks one day old which were divided into equal 10 groups (each 10 chicks). Chicks of 8 groups only from one day old treated with *Zingiber officinale* aqueous extract (5 and 10 ml/L drinking water) or *Thymus vulgaris*

aqueous extract (1 and 2 ml/lit. drinking water). The other 2 groups, one of them is control negative group 1 (orally inoculated with distilled water only) the second is control positive group 2 (orally infected with *E.coli* serogroup *O*<sub>78</sub> at 10 day old by a dose of 0.5 ml *E.coli* serogroup *O*<sub>78</sub> of broth containing  $3 \times 10^8$  CFU (colony forming units of *E. Coli*) / chick) (Johnson et al, 2001 & Khalid 1990). The serogroup obtained from the Serology Unit Bank of Animal Health Research Institute in Dokki Giza. Balanced food and water were available (NRC, 1994) and libitum. No antibacterial agents were given to all groups. Infected groups kept in a separate room from non infected and all conditions were the same. Chickens in all groups inspected daily.

Indices for evaluation the growth performance (Body weight, Body weight gain, Feed utilization) were determined at 4 and 6 weeks old age. Blood samples will be collected from each chicken (two samples) at the end of the 4<sup>th</sup> and 6<sup>th</sup> weeks old of age in the experiment. First was collected without anticoagulant in sterile, clean and dry screw capped centrifuge tubes for serum separation, stored in deep freezer at -20°C for later determination of immunological values. Second one with anticoagulant (EDTA) for total leuckocytic count.

A blood film was made for differential leuckocytic count as soon

as possible after the collection of the blood sample with the anticoagulant, by the manual method. Two blood films were made for each sample, stained by Giemsa stain (*Feldman et al, 2000*). The slides were examined under oil immersion magnification (*Coles, 1986*).

#### **Immunological studies:**

At the end of the experiment (6 weeks old) advanced immunological studies were done as (tumor necrosis factor  $\alpha$ , IL-1B, IL-6 IL-10 and IL-12). Serum tumor necrosis factor  $\alpha$  was determined using ready made chicken tumor necrosis factor- $\alpha$  (TNF-  $\alpha$ ) ELISA Kit provided by Invitrogen Company according to *Tian (2005)*. Serum interleukin 1 beta was determined using ready made chicken IL-1B ELISA Kit provided by Invitrogen Company. Serum interleukin 6 was determined using readymade chicken IL-6 ELISA Kit provided by Invitrogen Company according to *Pedersen (2007)*. Serum interleukin 10 was determined using readymade chicken IL-10 ELISA Kit provided by Invitrogen Company according to *Lobell (1998)*. Serum interleukin 12 was determined using readymade chicken IL-12 ELISA Kit provided by Invitrogen Company according to *Trinchieri (1998)*.

#### **Statistical analysis:**

The mean values and standard errors will be calculated for the obtained data and the level of

significances for all means were determined using general linear model (GLM) of multiple-variant analysis (full factorial design) using SPSS/pc<sup>+</sup> V17 statistical package for windows. Two groups were significantly different if P value was statically lower than 0.05.

#### **Results and Discussion**

Natural plants extracts act as immunostimulants, antimicrobial, growth promoters and also have a good flavor (*Mukesh et al, 2012*).

Natural feed additives of plant origin are believed to be safer, healthier and less regarded as chemical hazards than synthetic additives (*Ghazalah and Ali 2008*).

**As shown in tables (1 and 2):** adding herbal plants (*Thymus vulgaris* and *Zingiber Officinale*) have potentials as growth and health promoter in chicken without adverse effects. Current study revealed that treated groups with either *Thymus vulgaris* (1 and 2 ml/1 liter drinking water) and *Zingiber Officinale* (5 and 10 ml/1 liter drinking water) aqueous extracts by different doses either experimentally infected with *Escherichia Coli* (O78 strain) are better to improve feed conversion rate and weight gain. Obtained results are in agree with (*Cross et al, 2007*) who reported that dietary thyme had a different effect when used as an herb or oil on weight gain and body mass.

Moreover, obtained results are harmony also with (*Ghazalah and*

*Ali 2008*) who reported that these results may be due to herbs and herbal products are incorporated in poultry diets to replace synthetic products in order to stimulate or promote the effective use of feed nutrients which may subsequently result in more rapid body weight gain, higher production rates and improved feed efficiency. Moreover, active components of herbs may improve digestion functions in broilers.

One possible mechanism of how medicinal plants work is by improving broilers growth and feed utilization through the improvement of nutrients digestibility. This is through-facilitating absorption of calorigenic nutrients across the gut wall by increasing its absorption capacity (*Nelson et al, 1963*).

The growth promoting active feed supplements improve stability of feed and auspicious impact the digestive micropopulation mostly through inhibition of pathogenic microorganisms' growth. In consideration of promoted health status of intestinal tract, farm animals are less exposed to the toxins produced by different microorganisms (*Frankič et al, 2009*). Obtained results are in harmony with (*Basilico and Basilico, 1999; Hernandez et al, 2004*) who recorded that the phenolic compounds carvacrol and thymol present in *thyme* exhibit considerable antimicrobial activity. That effect is mainly due to the lipophilic character of the active

principles, which permeate the cell membranes and mitochondria of the microorganisms and inhibit, among others, the membrane bound electron flow and there with the energy metabolism. This leads to a collapse of the proton pump and draining of the ATP pool. High concentrations of essential oils also lead to lysis of the cell membranes and denaturation of cytoplasmic proteins (*Helander et al, 1998*). Nevertheless, there is only limited data on *in vivo* effects of thyme and thymol.

**As shown in table (3 and 4) and advanced immunity study table (5):** avian leukocytes serve as the first line of defense against invading microorganism. The present study concluded that total leukocytic count, heterophils and lymphocytes were higher in experimentally infected groups with *E.coli O78* and treated with either *Thymus vulgaris* (1 and 2 ml/1 liter drinking water) and *Zingiber Officinale* (5 and 10 ml/1 liter drinking water) aqueous extracts by different doses in compare to either control positive or negative also in compare to treated groups only .Moreover, groups that treated with either thymus vulgaris or *Zingiber officinale* showed increase in the total leukocytic count in compare to both control positive and negative.

(*Craig, 1999 and Ghazalah and Ali, 2008*) are agree with us as they confimed that active components of herbs have a great influence on the function and reactivity of the

immune system of the farm animals and may stimulate the immune function in broilers.

The present results agree with (*Selitrennikoff, 2001*) who ensures that plants do not have an immune system directly comparable to that of animals. Thus, plants have evolved host defense mechanisms either by pre-existing or induced physical barriers by numerous antimicrobial compounds such as antimicrobial peptides, proteins and small molecular weight organic substances.

(*Demir et al, 2008*) are agree with the present results as they fed 1 day old male broiler chicks (Ross) by a starter diet supplemented by *thyme* powder until 21 d of age and a finisher diet until 42 day influenced significantly humeral immunity.

The present serum immunological studies (interleukins and tumor necrosis factor) are in harmony with (*Maha, 2010*) that confirm that thyme oil orally administration to normal, *Pseudomonas aeruginosa* infected and non infected mice at a dose level of 720µg/10 mice affect the neutrophil phagocyte activity that had potentially increased as a result of thyme oil administration associated with elevated expression of immunoglobulin G. The thyme oil ameliorating changes may be mediated via its immunomodulatory effects on phagocyte activity and releasing of immunoglobulins by B cell, while disagree with (*Ocana and Regler, 2012*) who confirm that addition of 5.15 and 25 µg/ ml of

thyme extracts for 24-48 hours to human macrophages derived from THP-1 monocytes caused significant reduction in production and gene expression of the proinflammatory mediators TNF- $\alpha$ , IL-1B, IL-6 and highly increased these parameters on the anti-inflammatory IL10 mainly occur with higher doses. Also (*Fachini-Queiros et al, 2012 & Juhas et al, 2008*) who ensured that thyme oil at concentrations of 1, 3, 10, 30 and 90 µg/ ml significantly decrease TNF- $\alpha$  and IL-6 & Dietary supplement of 5000 ppm thyme essential oil caused a significant inhibition of total m RNA IL-1B and IL-6 expression in the mouse colon respectively.

**As shown in table 3 and 4:** heterophil to lymphocyte ratio were increased in treated and treated infected groups in compare to control also globulin serum levels. These results are agree with (*Navid and Mahmoud, 2011*) as they use *Thyme* powder (dried thyme was supplied from local market and after fine milling, mixing) that was supplemented to 1 day following until 42 days to broiler chicks at 0.75%, 1%, 1.5%, and 2% doses. Heterophil to lymphocyte ratio was insignificantly increased in groups supplemented with thyme powder by except for the 1% dose in compare to the control. Globulin serum level insignificantly increased in all treated groups in compare to the control.

**Recommendation:** From the obtained data it can be concluded that 2 ml *Thymus vulgaris* extract or 10 ml *Zingiber officinale* extract / 1

litter drinking water from one day old age of the chick the best to use in the field.

**Table (1)** Some growth performance profiles at 4 weeks old age (mean  $\pm$ S.E) in broiler chicks experimentally infected with *E. coli* O78 and treated with either *Thymus vulgaris* or *Zingiber officinale*.

Group	Parameter	Body weight (gram)	Weight gain (gram)	Feed intake (gram)	Feed conversion rate
Gr 1	(control negative)	885 $\pm$ 1.581 <sup>e</sup>	838.69 $\pm$ 7.607 <sup>e</sup>	1435 $\pm$ 1.581 <sup>e</sup>	1.71 $\pm$ 0.014 <sup>f</sup>
Gr 2	(control positive)	801 $\pm$ 1.581 <sup>j</sup>	754.67 $\pm$ 2.139 <sup>j</sup>	1375 $\pm$ 1.581 <sup>j</sup>	1.82 $\pm$ 0.003 <sup>b</sup>
Gr 3	Zingiber officinale 5 ml	941 $\pm$ 1.581 <sup>d</sup>	895.46 $\pm$ 1.598 <sup>d</sup>	1482 $\pm$ 1.581 <sup>d</sup>	1.66 $\pm$ 0.001 <sup>g</sup>
Gr 4	Zingiber officinale 5 ml+ infected	814 $\pm$ 1.581 <sup>i</sup>	767.7 $\pm$ 6.529 <sup>si</sup>	1382 $\pm$ 1.581 <sup>i</sup>	1.8 $\pm$ 0.016 <sup>b</sup>
Gr 5	Zingiber officinale 10 ml	968 $\pm$ 1.581 <sup>c</sup>	922.24 $\pm$ 1.387 <sup>c</sup>	1495 $\pm$ 1.581 <sup>c</sup>	1.62 $\pm$ 0.002 <sup>h</sup>
Gr 6	Zingiber officinale 10 ml+ infected	833 $\pm$ 1.581 <sup>h</sup>	788.17 $\pm$ 0.934 <sup>h</sup>	1399 $\pm$ 1.581 <sup>h</sup>	1.78 $\pm$ 0.002 <sup>e</sup>
Gr 7	Thyme vulgaris 1 ml	985 $\pm$ 1.581 <sup>hb</sup>	938.71 $\pm$ 0.852 <sup>b</sup>	1501 $\pm$ 1.581 <sup>b</sup>	1.6 $\pm$ 0.002 <sup>i</sup>
Gr 8	Thyme vulgaris 1 ml+ infected	855 $\pm$ 1.581 <sup>hg</sup>	810.1 $\pm$ 1.615 <sup>g</sup>	1412 $\pm$ 1.581 <sup>g</sup>	1.74 $\pm$ 0.002 <sup>d</sup>
Gr 9	Thyme vulgaris 2 ml	1019 $\pm$ 1.581 <sup>ha</sup>	971.7 $\pm$ 0.776 <sup>a</sup>	1511 $\pm$ 1.581 <sup>a</sup>	1.56 $\pm$ 0.002 <sup>j</sup>
Gr 10	Thyme vulgaris 2 ml+ infected	876 $\pm$ 1.581 <sup>hf</sup>	830.53 $\pm$ 1.421 <sup>f</sup>	1431 $\pm$ 1.581 <sup>f</sup>	1.72 $\pm$ 0.002 <sup>e</sup>

The same letter in the same row is insignificant at P< (00.05)

**Table (2)** Some growth performance profiles at 6 weeks old age (mean  $\pm$ S.E) in broiler chicks experimentally infected with *E. coli* O78 and treated with either *Thymus vulgaris* or *Zingiber officinale*.

Group	Parameter	Body weight (gram)	Weight gain (gram)	Feed intake (gram)	Feed conversion rate
Gr 1	(control negative)	1854.82 $\pm$ 7.016 <sup>e</sup>	969.82 $\pm$ 6.382 <sup>d</sup>	1735 $\pm$ 1.581 <sup>d</sup>	1.79 $\pm$ 0.011 <sup>e</sup>
Gr 2	(control positive)	1588.19 $\pm$ 7.837 <sup>j</sup>	787.19 $\pm$ 6.341 <sup>i</sup>	1598 $\pm$ 1.581 <sup>h</sup>	2.03 $\pm$ 0.016 <sup>a</sup>
Gr 3	Zingiber officinale 5 ml	1955.25 $\pm$ 1.600 <sup>d</sup>	1014.3 $\pm$ 0.055 <sup>e</sup>	1780 $\pm$ 1.581 <sup>c</sup>	1.76 $\pm$ 0.016 <sup>f</sup>
Gr 4	Zingiber officinale 5 ml+ infected	1622.08 $\pm$ 8.157 <sup>i</sup>	808.08 $\pm$ 7.908 <sup>b</sup>	1600 $\pm$ 1.581 <sup>b</sup>	1.98 $\pm$ 0.020 <sup>b</sup>
Gr 5	Zingiber officinale 10 ml	2008.46 $\pm$ 8.790 <sup>c</sup>	1040.46 $\pm$ 9.724 <sup>b</sup>	1800 $\pm$ 1.581 <sup>b</sup>	1.73 $\pm$ 0.016 <sup>g</sup>
Gr 6	Zingiber officinale 10 ml+ infected	1702.33 $\pm$ 1.225 <sup>h</sup>	869.33 $\pm$ 1.581 <sup>g</sup>	1630 $\pm$ 1.581 <sup>g</sup>	1.88 $\pm$ 0.002 <sup>e</sup>
Gr 7	Thyme vulgaris 1 ml	2071.96 $\pm$ 2.800 <sup>b</sup>	1086.96 $\pm$ 1.445 <sup>a</sup>	1850 $\pm$ 1.581 <sup>a</sup>	1.7 $\pm$ 0.003 <sup>h</sup>
Gr 8	Thyme vulgaris 1 ml+ infected	1781.03 $\pm$ 2.080 <sup>g</sup>	926.03 $\pm$ 1.442 <sup>f</sup>	1690 $\pm$ 1.581 <sup>f</sup>	1.83 $\pm$ 0.003 <sup>d</sup>
Gr 9	Thyme vulgaris 2 ml	2113.67 $\pm$ 20.069 <sup>a</sup>	1094.67 $\pm$ 19.741 <sup>a</sup>	1850 $\pm$ 1.581 <sup>a</sup>	1.69 $\pm$ 0.032 <sup>h</sup>
Gr 10	Thyme vulgaris 2 ml+ infected	1825.47 $\pm$ 2.730 <sup>f</sup>	949.47 $\pm$ 2.925 <sup>e</sup>	1710 $\pm$ 1.581 <sup>e</sup>	1.8 $\pm$ 0.006 <sup>e</sup>

The same letter in the same row is insignificant at P< (00.05)

**Table (3)** Blood leucogram at 4 weeks old age (mean ±S.E) in broiler chicks experimentally infected with with *E. coli* O78 and treated with either *Thymus vulgaris* or *Zingiber officinale*.

Group	Parameter	TLC ×10 <sup>3</sup> /µl	Heterophils ×10 <sup>3</sup> /µl	Eosinophils ×10 <sup>3</sup> /µl	Basophils ×10 <sup>3</sup> /µl	Lymphocytes ×10 <sup>3</sup> /µl	Monocytes ×10 <sup>3</sup> /µl	H/L Ratio
Gr 1	(control negative)	22.77±0.387 <sup>a</sup>	7.49±0.173 <sup>b</sup>	0.77±0.153 <sup>b</sup>	0.000±0.000 <sup>a</sup>	13.93±0.472 <sup>a</sup>	0.59±0.136 <sup>d</sup>	0.54±0.021 <sup>f</sup>
Gr 2	(control positive)	24.56±1.352 <sup>f</sup>	8.21±0.555 <sup>e</sup>	0.82±0.102 <sup>ab</sup>	0.002±0.003 <sup>ab</sup>	14.71±0.837 <sup>a</sup>	0.82±0.213 <sup>cd</sup>	0.56±0.035 <sup>f</sup>
Gr 3	Zingiber officinale 5 ml	24.84±1.25 <sup>ef</sup>	8.58±0.373 <sup>fc</sup>	0.84±0.137 <sup>ab</sup>	0.002±0.002 <sup>ab</sup>	14.43±0.866 <sup>a</sup>	0.99±0.112 <sup>c</sup>	0.6±0.029 <sup>ef</sup>
Gr 4	Zingiber officinale 5 ml+ infected	26.12±0.983 <sup>de</sup>	9.89±0.508 <sup>de</sup>	0.86±0.226 <sup>ab</sup>	0.006±0.003 <sup>de</sup>	13.92±0.489 <sup>a</sup>	1.45±0.199 <sup>b</sup>	0.71±0.033 <sup>cd</sup>
Gr 5	Zingiber officinale 10 ml	25.45±1.047 <sup>ef</sup>	9.19±0.387 <sup>f</sup>	0.84±0.24 <sup>ab</sup>	0.004±0.001 <sup>ef</sup>	14.34±0.816 <sup>a</sup>	1.08±0.276 <sup>c</sup>	0.64±0.031 <sup>c</sup>
Gr 6	Zingiber officinale 10 ml+ infected	28.13±0.798 <sup>c</sup>	11.3±0.52 <sup>c</sup>	0.92±0.219 <sup>ab</sup>	0.011±0.002 <sup>bc</sup>	14.24±0.943 <sup>a</sup>	1.66±0.25 <sup>b</sup>	0.8±0.077 <sup>b</sup>
Gr 7	Thyme vulgaris 1 ml	25.33±1.224 <sup>ef</sup>	9.24±0.212 <sup>cf</sup>	0.83±0.104 <sup>ab</sup>	0.006±0.003 <sup>def</sup>	14.15±1.055 <sup>a</sup>	1.1±0.121 <sup>c</sup>	0.66±0.036 <sup>de</sup>
Gr 8	Thyme vulgaris 1 ml+ infected	30.22±1.652 <sup>b</sup>	13.13±0.563 <sup>b</sup>	0.98±0.223 <sup>ab</sup>	0.013±0.004 <sup>ab</sup>	14.03±0.944 <sup>a</sup>	2.07±0.289 <sup>a</sup>	0.94±0.056 <sup>a</sup>
Gr 9	Thyme vulgaris 2 ml	27.20±1.477 <sup>cd</sup>	10.39±0.573 <sup>d</sup>	0.88±0.247 <sup>ab</sup>	0.008±0.002 <sup>cd</sup>	14.38±0.648 <sup>a</sup>	1.55±0.245 <sup>b</sup>	0.72±0.04 <sup>c</sup>
Gr 10	Thyme vulgaris 2 ml+ infected	31.98±1.366 <sup>a</sup>	13.96±0.923 <sup>a</sup>	1.04±0.276 <sup>a</sup>	0.016±0.003 <sup>a</sup>	14.72±0.494 <sup>a</sup>	2.24±0.319 <sup>a</sup>	0.95±0.077 <sup>a</sup>

The same letter in the same raw is insignificant at P< (00.05)

**Table (4)** Blood leucogram at 6 weeks old age (mean ±S.E) in broiler chicks experimentally infected with with *E. coli* O78 and treated with either *Thymus vulgaris* or *Zingiber officinale*.

Parameter Group	TLC ×10 <sup>3</sup> /µl	Heterophils ×10 <sup>3</sup> /µl	Eosinophils ×10 <sup>3</sup> /µl	Basophils ×10 <sup>3</sup> /µl	Lymphocytes ×10 <sup>3</sup> /µl	Monocytes ×10 <sup>3</sup> /µl	H/L Ratio
Gr 1 (control negative)	24.22±0.593 <sup>e</sup>	6.27±0.303 <sup>f</sup>	1.288±0.105 <sup>d</sup>	0.000±0.000 <sup>e</sup>	15.38±0.391 <sup>ab</sup>	1.28±0.152 <sup>b</sup>	0.408±0.020 <sup>g</sup>
Gr 2 (control positive)	24.51±1.456 <sup>e</sup>	6.4±0.660 <sup>f</sup>	1.295±0.173 <sup>d</sup>	0.002±0.002 <sup>fg</sup>	15.4±0.905 <sup>ab</sup>	1.42±0.213 <sup>b</sup>	0.415±0.028 <sup>g</sup>
Gr 3 Zingiber officinale 5 ml	24.72±1.007 <sup>e</sup>	6.46±0.414 <sup>f</sup>	1.312±0.147 <sup>d</sup>	0.003±0.002 <sup>efg</sup>	15.4±0.500 <sup>ab</sup>	1.54±0.228 <sup>ab</sup>	0.42±0.020 <sup>g</sup>
Gr 4 Zingiber officinale 5 ml+ infected	27.57±0.773 <sup>cd</sup>	8.78±0.395 <sup>d</sup>	1.45±0.241 <sup>bcd</sup>	0.007±0.003 <sup>cde</sup>	15.28±0.601 <sup>ab</sup>	2.06±0.260 <sup>cd</sup>	0.575±0.028 <sup>d</sup>
Gr 5 Zingiber officinale 10 ml	25.33±1.014 <sup>e</sup>	7.16±0.410 <sup>f</sup>	1.35±0.175 <sup>d</sup>	0.004±0.003 <sup>efg</sup>	15.11±0.658 <sup>b</sup>	1.72±0.085 <sup>fg</sup>	0.47±0.031 <sup>f</sup>
Gr 6 Zingiber officinale 10 ml+ infected	30.18±1.047 <sup>b</sup>	10.95±0.652 <sup>b</sup>	1.59±0.292 <sup>abc</sup>	0.010±0.006 <sup>abc</sup>	15.1±0.591 <sup>b</sup>	2.54±0.207 <sup>c</sup>	0.73±0.053 <sup>b</sup>
Gr 7 Thyme vulgaris 1 ml	26.87±1.208 <sup>d</sup>	8.14±0.399 <sup>d</sup>	1.42±0.122 <sup>cd</sup>	0.005±0.002 <sup>def</sup>	15.36±0.841 <sup>ab</sup>	1.94±0.336 <sup>ef</sup>	0.53±0.032 <sup>e</sup>
Gr 8 Thyme vulgaris 1 ml+ infected	32.11±0.85 <sup>a</sup>	11.89±0.616 <sup>a</sup>	1.67±0.111 <sup>ab</sup>	0.012±0.006 <sup>ab</sup>	15.65±0.285 <sup>ab</sup>	2.89±0.211 <sup>b</sup>	0.76±0.039 <sup>ab</sup>
Gr 9 Thyme vulgaris 2 ml	28.46±1.521 <sup>c</sup>	9.72±0.854 <sup>c</sup>	1.5±0.125 <sup>bcd</sup>	0.009±0.004 <sup>bcd</sup>	14.99±0.685 <sup>b</sup>	2.25±0.248 <sup>d</sup>	0.65±0.037 <sup>c</sup>
Gr 10 Thyme vulgaris 2 ml+ infected	33.42±1.557 <sup>a</sup>	12.44±0.479 <sup>a</sup>	1.73±0.22 <sup>a</sup>	0.014±0.004 <sup>a</sup>	16.01±1.023 <sup>a</sup>	3.23±0.234 <sup>a</sup>	0.78±0.033 <sup>a</sup>

The same letter in the same raw is insignificant at P< (00.05)

**Table (5)** serum immunological studies at 6 weeks old age (mean  $\pm$ S.E) in broiler chicks experimentally infected with with *E. coli* O78 and treated with either *Thymus vulgaris* or *Zingiber officinale*.

Group \ Parameter	Interleukin 1 pg/ml	Interleukin 6pg/ml	Interleukin 10pg/ml	Interleukin 12 pg/ml	Tumor necrosis factor pg/ml
Gr 1 (control negative)	21.5 $\pm$ 3.72 <sup>f</sup>	15.7 $\pm$ 3.51 <sup>e</sup>	21.43 $\pm$ 2.00 <sup>e</sup>	14.23 $\pm$ 2.63 <sup>c</sup>	15.63 $\pm$ 3.47 <sup>c</sup>
Gr 2 (control positive)	142.17 $\pm$ 21.17 <sup>b</sup>	162.9 $\pm$ 23.08 <sup>b</sup>	101.8 $\pm$ 11.58 <sup>c</sup>	145.17 $\pm$ 28.46 <sup>a</sup>	75.60 $\pm$ 7.63 <sup>b</sup>
Gr 3 Zingiber officinale 5 ml	21.7 $\pm$ 4.36 <sup>f</sup>	21.47 $\pm$ 4.58 <sup>e</sup>	16.3 $\pm$ 1.05 <sup>e</sup>	19.33 $\pm$ 1.03 <sup>c</sup>	12.33 $\pm$ 1.53 <sup>c</sup>
Gr 4 Zingiber officinale 5 ml+ infected	88.2 $\pm$ 6.09 <sup>c</sup>	95.13 $\pm$ 8.41 <sup>c</sup>	83.63 $\pm$ 6.05 <sup>d</sup>	84.5 $\pm$ 12.17 <sup>c</sup>	56.67 $\pm$ 5.77 <sup>c</sup>
Gr 5 Zingiber officinale 10 ml	16.53 $\pm$ 1.7 <sup>f</sup>	14.4 $\pm$ 0.91 <sup>e</sup>	15.00 $\pm$ 2.91 <sup>e</sup>	17.87 $\pm$ 1.26 <sup>c</sup>	13.97 $\pm$ 2.56 <sup>c</sup>
Gr 6 Zingiber officinale 10 ml+ infected	71.37 $\pm$ 3.06 <sup>cd</sup>	78.77 $\pm$ 8.3 <sup>cd</sup>	57.17 $\pm$ 4.47 <sup>e</sup>	82.6 $\pm$ 5.15 <sup>c</sup>	38.07 $\pm$ 4.01 <sup>d</sup>
Gr 7 Thyme vulgaris 1 ml	61.9 $\pm$ 12.42 <sup>ed</sup>	72.83 $\pm$ 4.76 <sup>cd</sup>	44.7 $\pm$ 4.87 <sup>f</sup>	58.57 $\pm$ 3.07 <sup>d</sup>	39.67 $\pm$ 7.13 <sup>d</sup>
Gr 8 Thyme vulgaris 1 ml+ infected	193.00 $\pm$ 16.86 <sup>a</sup>	194.33 $\pm$ 28.13 <sup>a</sup>	139.17 $\pm$ 14.07 <sup>b</sup>	152.2 $\pm$ 8.35 <sup>a</sup>	92.93 $\pm$ 3.57 <sup>a</sup>
Gr 9 Thyme vulgaris 2 ml	48.4 $\pm$ 10.57 <sup>e</sup>	61.03 $\pm$ 2.55 <sup>d</sup>	72.3 $\pm$ 4.12 <sup>d</sup>	46.5 $\pm$ 3.37 <sup>d</sup>	34.03 $\pm$ 3.57 <sup>d</sup>
Gr 10 Thyme vulgaris 2 ml+ infected	146.63 $\pm$ 7.42 <sup>b</sup>	140.43 $\pm$ 16.21 <sup>b</sup>	183.9 $\pm$ 6.12 <sup>a</sup>	113.93 $\pm$ 10.13 <sup>b</sup>	67.77 $\pm$ 3.89 <sup>b</sup>

The same letter in the same raw is insignificant at  $P < (00.05)$

## References

**Abaza, I.M.K (2001).** The use of some medicinal plants as feed additives in broiler diets. Ph. D. Thesis, Faculty of Agriculture, Alexandria University.

**Al-Harhi M. A. (2002):** Performance and carcass characteristics of broiler chicks as affected by different dietary types and levels of herbs and spices as non classical growth promoters. Egypt. Poult. Sci. J., 22: 325-343.

**Barnes, H. J., Vaillancourt J-P. and Gross. W. B. (2003):** Colibacillosis. In: Diseases of Poultry, 11th ed., Iowa State University Press, Ames, IA, USA.

**Basilico, M. Z., and Basilico J. C., (1999):** Inhibitory effects of some spice essential oils on *Aspergillus ochraceus* NRRL 3174 growth and

ochratoxin a production. Lett. Appl. Microbiol. 29; 238– 241.

**Coles E H (1986):** Veterinary clinical pathology. 4<sup>th</sup> Ed. W.B. Saunders company, U.S.A.

**Craig, W. J., (1999):** Health-promoting properties of common herbs. American Journal of Clinical Nutrition 70 (suppl): 491S-499S.

**Cross, D.E., McDevitt, R.M., Hillman, K. and Acamovic, T. (2007):** The effect of herbs and their associated essential oils on performance, dietary digestibility and gut microflora in chickens from 7 to 28 days of age. Br. Poult. Sci. 48, 496-506.

**Demir E., Kilinc K., Yildirim Y., Fatma D and Eseceli H. (2008):** Comparative effects of mint, sage, thyme and flavomycin in wheat-



based broiler diets. *Archiva Zootechnica* 11:3, 54-63.

**El-Husseiny O, Shalash S. M. and Azouz H. M. (2002):** Response of broiler performance to diets containing hot pepper and/ or fenugreek at different metabolizable energy levels. *Egypt. Poult. Sci. J.* 22: 387-406.

**Fachini-Queiroz FC, Raquel K, Maria D C, Joice M C, Renata G, Ciomar A and Roberto K (2012):** Effect of thymol and carvacrol, constituents of thymus vulgaris l. Essential oil on inflammatory response. Evidence-based complementary and alternative medicine; 1-10.

**Feldman B F, Zink J G and Jain N C (2000):** Schalm's veterinary hematology. 5<sup>th</sup> Ed. Lippincott William and Wilkins. Canada: 1145- 1146.

**Frankič, T., Voljč, M., Salobir, J., and Rezar, V. (2009):** Use of herbs and spices and their extracts in animal nutrition. *Acta agriculturae Slovenica*, 94/2, 95-102.

**Ghazalah A.A. and Ali A.M. (2008):** Rosemary leaves as a dietary supplement for growth in broiler chickens. *Int. Poult. Sci.* 7, 234-239.

**Heitzman R. J. (1986):** Residues in Animal products. PP. 157-175 in: *Recent advances in Animal Nutrition*, W. Haresign and D. J. A. Cole, eds. Butterworths, London, UK.

**Helander, I.M., Alakomi H.-L., Latva-Kala, K., Mattila-Sandholm T.P.I., Smid, E.J.,**

**Gorris L.G.M., and Wright A. (1998):** Characterization of the action of selected essential oil components on gram-negative bacteria. *J. Agric. Food Chem.* 46, 3590-3595.

**Hernandez F., Madrid J., Garcia V., Orengo J., and Megi'as M. D., (2004):** Influence of two plant extracts on broilers performance, digestibility, and digestive organ size. *Poult. Sci.* 83; 169-174.

**Johnson LC, Bilgili SF, Hoerr FJ, McMurtrey BL and Norton RA (2001):** The effects of early exposure of cellulitis associated *E. coli* in one day old broiler chickens. *Avian pathology* 3 (2), 175-178.

**Juhás, S., Cikos S., CzikkovS. á, VeselJ. Il'kov á, G.á , Hájek T., Domaracký K., Domarack M., Bujnáková D., Rehák P., and Koppel J. (2008):** Effects of borneol and thymoquinone on TNBS-induced colitis in mice. *Folia Biol.* 54(1): p. 1-7.

**Khachatourians G. G. (1998):** Agriculture use of antibiotics and the evolution and transfer of antibiotic-resistant bacteria. *Can. Med. Assoc. J.* 159:1129-1136.

**Khalid, AM (1990):** Studies on natural and experimental *Escherichia coli* infection in chickens. *J. Egypt. Vet. Ass.*, 50 (3): 379-389.

**Lobell A (1998):** Methodology for determination of IL-10. *J. Exp. Med.*, 187 (9): 1543- 1548.

**Maha G S (2010):** Immunological and DNA Fragmentation Studies On The Protective Effect Of Thyme

Against Navelbine Induced Oxidative Stress In Mice Nature and Science; 8(8).

**Mukesh K. B., Jitender K.J., Satyanarayana Y. and Reddy A. D. (2012):** Animal and plant originated immunostimulants used in aquaculture. Scholars Research Library. J. Nat. Pro. Plant Resour; 2 (3): 397- 400.

**National Research Council (NRC) (1994):** Nutrient requirements of poultry. 9<sup>th</sup> Ed., National Academy PRESS, Washington, DC., USA.

**Navid H. M. and Mahmoud P. M. (2011):** The effects of different levels of Thyme on performance, carcass traits, blood parameters of broilers. Annals of Biological Research, 2 (4):379-385.

**Nelson F. E., L. S. Jensen, and M. C. Ginnis J. (1963):** Studies on the stimulation of growth by dietary Amoxicillins 2- Effect of Amoxicillins on metabolizable energy of the diet. Poult. Sci. 42: 209-219.

**Ocana M and Regler G (2012):** Effects of thyme extract oils (from thymus vulgaris, thymus zygis and thymus hyemalis) on cytokine production and gene expression of oxLDL-Stimulated THP-1-Macrophages, Journal of obesity; 11: 1-11.

**Pedersen B K (2007):** Role of myokines in exercise and metabolism. J. Appl. Physiol. 103 (3): 1093-98.

**Selitrennikoff CL (2001):** Antifungal proteins. App. Environ. Microbial; 67: 2883-2894.

**Tian B (2005):** Identification of direct genomic targets downstream of the nuclear factor signaling. J. Biol. Chem.;280 (17): 17435- 7448.

**Trinchieri G (1998):** Methodology for determination of IL-12. Adv. Immunol.; 70: 83- 243.

**Wary C and R. H. Davies (2000):** Competitive exclusion-an alternative to antibiotics. Vet. J., 59:107-108.

## الملخص العربي

### دراسات مناعية متقدمة على تأثير بعض النباتات العشبية في بدارى التسمين

مستخلص الزعتر أو الزنجبيل له تأثير فعال على مظاهر النمو الطبيعي في فراخ التسمين و المناعة و يمكن أن يكون له تأثير على نمو الفراخ المصابة معمليا بالميكروب القولوني عترة 078 . في دراستنا استهدفنا دراسة تأثير كل من مستخلص الزعتر أو الزنجبيل كل من بدارى التسمين الصحيحة أو المعدية معمليا بالميكروب القولوني عترة 078 على مظاهر النمو و صورة الدم و أيضا المناعة المتقدمة.

اجرينا الدراسة الفعلية على ١٠٠ فرخ عمر اليوم الواحد وتم تقسيمهم إلى ١٠ مجموعة الأولى منها ضابطة سالبة و الثانية ضابطة موجب معدية بالعترة البكتيرية فقط. الثماني مجموعات الأخرى عولجت من عمر يوم أما بالزعتر (١ أو ٢ملى/لتر ماء شرب) أو الزنجبيل (٥ أو ١٠ ملى/لتر ماء شرب). تم عدوى أربعة مجموعات فقط من الثمانية بالعترة الميكروبية عند عمر ١٠ يوم. قمنا بعد ذلك بقياس مظاهر النمو و بعض التغيرات الدموية و المناعية عند عمر ٤ و ٦ أسابيع و كانت النتائج كالآتي:-

اظهرت الأوزان و معدلات التحول الغذائي تحسنا ملحوظا في المجموعات المعالجة فقط بكل من الزعتر أو الزنجبيل بالجرعات العالية و كانت الأفضل. كما اظهرت المجموعات المعدية و المعالجة بجرعات عالية من كلا المستخلصين نتائج أفضل من الجرعات الأقل و المعدية فقط. بالنسبة لعينات الدم و المصل عموما اظهرت الجرعات العالية من كلا المستخلصين تحسنا ملحوظا في صورة الدم و قياسات المناعة المتقدمة و المؤشرات البيوكيميائية مقارنة بالجرعات الأقل سواء كانت غير معدية أو معدية. أيضا المجموعات المعدية و المعالجة بأى من المستخلصين أظهرت تحسنا ملحوظا بالمقارنة بالمجموعة الضابطة المعدية فقط. قمنا أيضا بأخذ عينات من (الكبد و القلب و الكلى و الأمعاء و الطحال و البرسا و الرئة) للفحص الباثولوجى و التى دعمت بقوة جميع النتائج السابقة حيث اظهرت بعض التغيرات المرضية الظاهرة في المجموعات المعدية فقط و المعدية و المعالجة بالجرعات الصغيرة من كلا المستخلصين و تحسن في تلك المعدية و المعالجة بالجرعات العلية من كل من الزعتر أو الزنجبيلز أيضا المعالجة فقط بالجرعات العالية أفضل من المعالجة فقط بالجرعات الأقل.