Original Article. January-December 2019; Vol. 9:1-7. Received: 01/03/2019 Accepted: 04/08/2019

http://dx.doi.org/10.21929/abavet2019.918

Effect of *Thymus daenensis* on immunity and performance in broiler

Efecto de *Thymus daenensis* sobre la inmunidad y rendimiento en pollo

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ABSTRACT

Thymus daenensis Celak is one of the species of *Thymus* that is endemic to Iran. *T. daenensis* is distributed in most parts of Iran, particularly over the Zagros and some parts of the Alborz mountain ranges. The leaves and flowering parts of Thyme plants were traditionally used for various medical purposes. For the study of thyme (*Thymus daenensis*) effect on performance and immunity in chickens, 180 one-day-old broiler chicks divided into 4 groups by 3 replicates, randomly. All chickens reared under the same condition and received 0, 0.25, 0.5 and 1 percent of thyme on diet from 5 to 42 days old, continuously. Control chickens feed basic diet did not receive any supplement on feed. Growth indices including body weight, weight gain, and feed conversion ratio (FCR) were determined and recorded at the end of each week. In 42 days old, chickens slaughtered and blood was sampled without anticoagulant for determine of antibody titer against Newcastle disease (ND) vaccine. The results showed that continuous consumption of 0.5 and 1 percent of thyme can increase weight gain, reduce feed consumption and FCR. In addition, the supplement of 0.5 and 1 percent of thyme can increase antibody titer in chickens but the differences were not significant between treatment and control groups. Therefore, it concluded that a supplement of 0.5 and 1 percent of thyme in chicken nutrition can improve the growth performance and no negative effect on the immunocompetence of broiler chickens.

Keywords: Chicken, productive parameters, medicinal plants, immunity.

RESUMEN

Thymus daenensis Celak es una de las especies de tomillo endémicas de Irán. *T. daenensis* se distribuye en la mayoría de las partes de Irán, particularmente en los Zagros y en algunas partes de las cadenas montañosas de Alborz. Las hojas y las partes florales de las plantas de tomillo se utilizan tradicionalmente con fines médicos. Se estudió el efecto del tomillo (*Thymus daenensis*) sobre el rendimiento e inmunidad en pollos, 180 aves de engorda de un día de edad se dividieron al azar en 4 grupos y 3 repeticiones. Todos los pollos se criaron en las mismas condiciones y recibieron 0, 0.25, 0.5 y 1 por ciento de tomillo en la dieta de 5 a 42 días.. El grupo control recibió la dieta básica sin suplemento. Los índices de crecimiento como

peso corporal, ganancia de peso e índice de conversión alimenticia (ICA) se registraron semanalmente. A los 42 días, los pollos se sacrificaron y se tomaron muestras de sangre sin anticoagulantes para determinar el título de anticuerpos contra la vacuna contra la enfermedad de Newcastle (ENC). Los resultados mostraron que el consumo de 0.5 y 1 por ciento de tomillo puede incrementar la ganancia de peso, reducir el consumo de alimento y ICA. Además, el suplemento a 0.5 y 1 por ciento de tomillo puede aumentar el título de anticuerpos en los pollos,.. Por lo tanto, se llegó a la conclusión que suplementar la dieta con 0.5 y 1 por ciento de tomillo se puede mejorar el crecimiento sin tener efecto negativo en la inmunocompetencia de los pollos.
Palabras clave: Pollos de engorda, parámetros productivos, plantas medicinales, inmunidad.

INTRODUCTION

Antibiotics in poultry diets can lead to antibiotic residual and resistance in poultry products such as meat and egg (Izadi *et al*., 2013). Using herbal plants with fewer side effects and no problems associated with residue in poultry product such as meat and egg can be a good option. Due to the increasing demand for the use of organic chickens, the use of herbal plants in poultry foods has become more popular worldwide as an alternative to antibiotics.

Thymus (commonly known as Thyme) is an herbal plant belonging to the *Lamiaceae* family, It is widely distributed throughout Europe, Asia and North Africa (Zarshenas and Krenn, 2015). *Thymus daenensis* Celak is one of the species of *Thymus* that is endemic to Iran. *T. daenensis* is distributed in most parts of Iran, particularly over the Zagros and some parts of the Alborz mountain ranges (Bahmani *et al*., 2014). Fresh Thyme has one of the highest antioxidant levels among herbs. It is accompanying with minerals and vitamins that are essential for health. Its leaves are one of the rich sources of potassium, iron, calcium, manganese, magnesium, and selenium (Bolukbasi and Erham, 2007). Thymol is the main phenolic component that is responsible for its antioxidative activity (Gholami-Ahangaran *et al*., 2015). Thyme contains many flavonoids, phenolic antioxidant components such as *zeaxanthin*, *pigenin, lutein, luteolin* and *thymonin* (Bahmani *et al*., 2014). Thymol has been used as an antimicrobial medicinal drug. Also, Thymol improves liver function that influences performance (Bolukbasi and Erham, 2007). The leaves and flowering parts of Thyme plants were traditionally used for various medical purposes; as an antispasmodic, antitussive, expectorant, carminative, anti-inflammatory or tonic agent. Thyme has antibacterial and antioxidant properties due to containing thymol and carvacrol (Zarshenas and Krenn, 2015).

The antioxidants may be defined as molecules that prevent cell damage against free radicals and are critical for maintaining optimum health. Living cells require adequate amounts of antioxidant in order to avoid the harmful effect of reactive oxygen species (ROS) and to prevent damage to the immune system (Kiokias *et al*., 2018). During the inflammatory processes, the activation of phagocytes and/or the action of bacterial products with specific receptors are capable of promoting the assembly of the

multicomponent flavoprotein NADPH oxidase, which catalyzes the production of high amounts of the superoxide anion radical. Under this circumstance, neutrophils and macrophages are recognized to produce superoxide free radicals and oxygen peroxide, which are necessary for defense against pathogens. In this condition, antioxidants are essential to regulate the reactions that release free radicals (Puertollano *et al*., 2011). Antioxidants play a role as cofactors at the level of cytokine regulation, antioxidant nutrients commonly included in the diets or herbs improve immune function and protect against infectious disease. As a result, dietary antioxidants have been related to modulating the host susceptibility or resistance to infectious pathogens (Ang *et al*., 2018).

There are some studies that showed consumption of thyme as powder, extract or essential oil in partial time of growing period can increase growth in chickens while some studies represented thyme has no effect in performance. The current study was conducted to evaluate the effect of continuous orally administrated thyme powder from the early growing period to slaughtering on performance and immunity responses of broiler against Newcastle disease (ND) vaccine.

MATERIALS AND METHODS

In this study, 180 Ross strain broiler chicks were randomly divided into four equal groups with three replicates in 12 separate pens. So per pen 15 broiler chicks were reared until 42 days of age. All chickens in different groups received feed and water freely (*Ad libitum*). Diets were prepared for all groups a based on corn-soy and were balanced according to the requirements of NRC (1994). All chickens received thyme powder from 5 days old until 42 days old, based on the following: the first group was the control group and not received powdered thyme (group 1). The second, third and fourth groups received powdered thyme to the 0.25 (group 2), 0.5 (group 3) and one percent (group 4) of the diet, respectively. All chickens vaccinated with Newcastle disease (ND) vaccine at 10, 19 and 28 days old. The weight gain, feed intake and feed conversion ratio at 42 days of age were calculated and compared.

At 42 days old, all chickens were slaughtered and blood was sampled without anticoagulant for determine of antibody titer against ND vaccine. The antibody titers were measured by conventional haemagglutination inhibition (HI) test.

All data were analyzed with SPSS software and statistic program of one way ANOVA. If there is a statistical difference between the mean of data in different groups, the difference was expressed by Tukey test. The significant difference level was considered less than 0.05.

RESULTS

Growth performance

At 42 days old, the highest weight gain observed in broilers that received 0.5 percent of thyme but had no significant difference with chickens that received one and 0.25 percent of thyme, while is significantly higher than the control group (P< 0.05). Comparison of the cumulative feed intake at the end of the sixth week of the growing period showed no significant difference between groups. The FCR in chickens fed 0.5 and 1.0 percent thyme was significantly lower than control chickens and chickens received 0.25 percent of thyme. There were no significant difference in FCR in chickens received 0.5 and 1.0 percent of thyme in the diet (Table 1).

Table 1. Growing performance in different studied groups

Treatments	1 (0 %)	2 (0.25%)	3 (0.5%)	4 (1.0%)
Cumulative WG (g) in sixth week	2124 ± 140[b]	2160 ± 179[ab]	2328 ± 182[a]	2276 ± 143[ab]
Cumulative FI (g) in sixth week	3842 ± 341	3867 ± 265	3698 ± 175	3675 ± 170
Cumulative FCR in sixth week	1.81 ± 0.09[a]	1.79 ± 0.21[a]	1.59 ± 0.16[b]	1.61 ± 0.14[b]

BW: Body Weight, FI: Feed Intake, FCR: Feed conversion rate.

*Data presented as mean ± standard deviation.

[a, b, c]: different words represents significant differences Tukey between groups in each row (P<0.05).

HI Titer

Although the HI titers in chickens fed 0.5 and 1.0 percent of thyme were higher than others, comparison of HI titers revealed that there were no significant differences between treatment groups (Table 2).

Table 2. HI titer against ND vaccine in 42 days old

Treatments	1 (0)	2 (0.25%)	3 (0.5%)	4 (1.0%)
HI titer	3.9 ± 1.4	3.8 ± 1.5	4.1 ± 1.0	4.5 ± 1.7

*Data presented as mean ± standard deviation.

DISCUSSION

One main concern of the consumers of poultry meat products is a great to use of chemicals in poultry foods as well as the antibiotic residual and transmission of genes related to antibiotic resistance (Izadi *et al*., 2013). So, using of natural compounds in poultry food is popular, recently. Medicinal plants can be a good alternative to antibiotics in poultry diets. Iran for particular climatic is one of the rich sources of herbal plants (Bahmani *et al*., 2014). Thyme in *Lamiaceae* family is used for the improvement of the nervous problems, treatment of depression and insomnia, and has antibacterial, anti-parasitic and anti-fungal effects. It is used as a carminative, antispasmodic, digestive aid,

anti-cough and cold treatment, traditionally (Akbarinia and Mirza, 2008). One of the main genera is *thyme daenensis* that grows in different regions of Iran, especially Zagross Mountain areas (Noori *et al*., 2011). The main compounds are thymol and carvacrol that they have an antimicrobial effect (Jang *et al*., 2017). According to available information, thyme antibacterial effect was evaluated in different infections *in vitro* and in clinical trials in different species of animals and sometimes humans (Bolukbasi and Erham, 2007). Some reports are available for the antimicrobial effect of thyme in poultry (Kivanc *et al*., 1996; Akhondzadeh *et al*., 2004). For example, Kivanc *et al.* (1996) showed that thyme can reduce oocysts of coccidia and clostridial populations in broiler chickens. In addition, Akhondzadeh *et al.* (2004) stated that different percentages of thyme essential oils can have a negative effect on the growth of *Salmonella typhimurium*.

In the recent study, three doses of 0.25, 0.5 and 1.0 percentage of *thyme daenensis* from 5 days old until 42 days old were used in poultry diets. Results showed that adding the thyme to the chicken nutrition has no effect on feed intake, while doses of 0.5 and 1.0 percent thyme significantly increase weight gain and feed conversion compared to the control chickens. Previously, it has been demonstrated that thymol, as the main compound extracted from thyme, as a hydrophobic component, involved in function of the cell wall in bacteria (Noori *et al*., 2011). Thymol can cause a change in membrane permeability to K + and H + cations. The increase in cell permeability leads to leakage of ions and eventually leads to bacterial death (Haselmeyer *et al*., 2015). The reduction in bacterial pathogens in intestines can cause improvement in gastrointestinal health and leads to increased growth performance. Furthermore, some reports suggest that the effective ingredients in herbal plants play a role in the performance and digestive enzymes secretion and affect on the function of the digestive tract (Rahimi *et al*., 2011). Probably improvement in the enzyme activity indirectly increased digestion, absorption and eventually weight gain and feed efficiency in the present study.

Improving immunity responses to vaccines in poultry production is very important to prevent common important diseases. A variety of different factors such as chicken immunity, chicken age, and diet, vaccination failure, the incidence of immune suppressive diseases can induce immunity response to the vaccine. Some studies demonstrated that some herbs such as coneflower (*Echinacea purpurea*) were most effective as immunostimulator in vaccination immune response because of the stimulation of the non-specific immune system. It is thought that immune enhancement of *Echinacea* is provided by certain polysaccharides, flavonoids, and isobutylamides (Rehman *et al*., 1999). It seems that thyme that is rich in active compounds such as flavonoids, act as an antioxidant, may enhance the immune function. The results of this study showed that the thyme has no effect on antibody response against ND vaccine that this result was be agreed with the finding of Rahimi *et al*. (2011) that studied aqueous extract of thyme (*thymus vulgaris*) on SRBC and ND vaccine responses in chickens. Therefore, the results

of this study revealed the addition of 0.5 and 1.0 percent of thyme in poultry nutrition can improve the growth performance and no negative effect on the immunocompetence of broiler chickens.

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