Ann. Anim. Sci., Vol. 19, No. 3 (2019) 779–791 DOI: 10.2478/apas-2019-0023

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SUPPLEMENTAL EFFECTS OF EUCALYPTUS (EUCALYPTUS CAMALDULENSIS) LEAVES ON GROWTH PERFORMANCE, CARCASS CHARACTERISTICS, BLOOD **BIOCHEMISTRY AND IMMUNE RESPONSE OF GROWING RABBITS**

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Abstract

This research aimed to study the effect of dietary eucalyptus (Eucalyptus camaldulensis) leaves powder (EL) on growth performance, blood parameters, immune response and caecal microbiota in 84 growing Jabali and V-Line rabbits raised under high environmental temperature. The experiment started at 10 weeks of age and lasted 6 weeks. Rabbits were randomly distributed into three dietary eucalyptus treatments; control (no EL) and two groups supplemented with 0.1% and 0.2% EL powder. Rabbits were weighed at the beginning and at the end of the experiment. Feed intake, weight gain and feed conversion ratio were determined. Cell-mediated immune response was evaluated. At the end of the experiment, 54 rabbits were slaughtered (nine rabbits/sub-group). The results showed that the high level of EL (0.2%) had a negative effect on growth performance, dressing %, mid part % and significantly increased cell-mediated response. Rabbits fed the high level of EL showed shortening in caecum length. Significant linear reduction of total bacterial count and E. coli was observed in both groups given either 0.1% or 0.2% EL compared to the control group. Similar trend was found in the percentage of Salmonella sp. detection in both breeds. Jabali rabbits were significantly heavier than V-line rabbits and had better FCR and carcass traits except for fore part %. Additionally, they showed lower total microbial count. The current study indicated that EL could be utilized as an effective feed additive to improve cellular immunity and to reduce caecal bacterial counts in rabbits raised under high ambient temperature.

Key words: eucalyptus, caecal microbiota, antioxidant, immunity, rabbits

Nowadays, supplementation of natural feed additives and medicinal plants (phytogenics) to rabbit and poultry diets is a growing trend to improve both productive performance and health condition. Dietary phytogenics gained much interest in rabbit production as alternative to antibiotics to stimulate the growth by increasing the efficiency of feed utilization, enhance the immunity and the whole health status (Mancini et al., 2018; Parisi et al., 2018; Mancini et al., 2019). Moreover, dietary supplementation with natural antioxidants can enhance the oxidative stability of rabbit meat and consequently improve its shelf-life (Dalle Zotte, 2002).

Eucalyptus (Eucalyptus camaldulensis, EL) is a tall evergreen tree and now extensively cultivated in many regions worldwide. It contains several vital compounds including p-cymene, 1,8-cineole, β-phellandrene, spathulenol, cryptone aldehydes, cuminal, uncommon and phellandral, α -phellandrene, β -phellandrene leading to multi-functional characteristics such as antimicrobial, anti-inflammatory and antioxidative properties (Barra et al., 2010). In humans, EL is used to reduce nasal congestion in common cold during cold winter months (Sadlon and Lamson, 2010). Many investigators reported that feeding poultry a diet supplemented with eucalyptus improves productive traits, antioxidant status and immune response in laying hens and broilers (Abd El-Motaal et al., 2008; Farhadi et al., 2017). Supplementing diet with 0.1% EL can increase live body weight gain and growth rate in broiler chicks (Osman et al., 2007). Furthermore, polyphenols in eucalyptus leaves have shown various biological activities including antioxidant activity, antitumor activity and antibacterial activity (Salari et al., 2006; Bokaeian et al., 2010; Chen et al., 2017). Ahmed et al. (2005) reported that dietary EL had no significant effect on live body weight and daily weight gain in growing rabbits. Although many aspects of bioactive properties of eucalyptus have been explored in humans, data available of its effect on rabbit performance are very scarce. Our hypothesis focuses on the beneficial effect of the dietary EL in rabbit production, which has never been studied in rabbit under high ambient temperature. Therefore, the potential effects of dietary eucalyptus leaves inclusion on growth performance, blood parameters, immune response and caecal microbiota have been evaluated in growing rabbits raised under high environmental temperature.

Material and methods

This experiment was carried out at the experimental rabbit farm, College of Agriculture and Veterinary Medicine, Qassim University during summer season of 2017. The experiment started at 10 weeks of age and lasted 6 weeks. The averages of high and low ambient temperatures during the experimental period were $39^{\circ}C \pm 0.6$ and $24^{\circ}C \pm 0.8$, respectively (mean \pm SE). The animal care, handling and sampling procedures were approved by the Committee of Ethics and Animal Care of the Agricultural Research Center, Qassim University, Saudi Arabia.

Husbandry, diets and experimental design

The rabbits were kept individually under similar housing and management conditions inside a semi-closed rabbitry in wire mesh cages (50 cm \times 40 cm \times 40 cm) equipped with feeding hopper and drinking nipples. A factorial design of 2 \times 3 was followed in the present experiment. A total of 84 rabbits aged 10 weeks from two breeds (42 each) – Saudi local breed (Jabali, J) and imported Spanish breed (V-Line, V) – were randomly distributed into three dietary eucalyptus (*Eucalyptus camaldulensis*) treatments (14 rabbits /subgroup). The dietary treatments were control (no eucalyptus), while the other two groups were supplemented with 1 g and 2 g eucalyptus leaves powder/kg feed. Eucalyptus leaves were obtained from trees cultivated in the experimental research farm, Qassim University. The leaves were washed, airdried and ground into fine particles (powder). A commercial basal diet for growing rabbits containing 18.5% crude protein, 8.0% crude fibre, 3.0% crude fat and 9.4 MJ metabolizable energy/kg was used. The eucalyptus powder was mixed carefully with the ingredients of the basal diet before pelleting. Feed and water were available *ad libitum* to the animals.

Growth performance

All rabbits were weighed at the beginning and at the end of the experiment (16 weeks of age). Feed intake was individually recorded throughout the experimental period. Feed conversion ratio (FCR) was calculated as feed consumed divided by weight gain during the experimental period.

Assay for cell-mediated immunity

The *in vivo* response induced by injecting a mitogen was evaluated by injecting PHA-P into the left ear. At 14 weeks of age, 48 rabbits were randomly assigned (8 animals/subgroup) for cell-mediated immune response. Each animal was injected intradermally with 100 μ g PHA-P (Sigma Chemical Co., St Louis, MO, USA) in 0.1 ml of sterile saline. Upon injection, the site of needle was marked with permanent black ink to facilitate further measuring. The resultant swelling response in the ear was measured with a constant tension dial micrometer (Ames, Waltham, Massachusetts, USA) before injection and at 24, 48 and 72 h after PHA-P injection. The ear swelling was expressed as the difference between the thickness of the ear before and after injection.

Slaughter and carcass characteristics

At the end of the experiment, 54 animals were weighed, fasted for 12 h with free access to clean drinking water and slaughtered (n=9 animals for each subgroup). Upon bleeding, the rabbits were dissected according to Fathi et al. (2017). After skinning, the carcass was opened down and all organs and offal were removed. Hot carcass, skin, head, liver, heart, kidney and spleen were excised and weighed. The carcass was divided into three cuts; fore part, mid part and hind part. All data were expressed as a percentage of live body weight.

Blood collection, haematology and biochemical assay

Two blood samples were collected during slaughter from each rabbit into heparinized tubes for determination of haematological parameters and biochemical analysis. The haematological parameters were assessed by using Automatic Fully Digital Haematology Analyzer, BC-3000 Plus (Mindary, Bio-Medical Electronics Co., Ltd). These parameters were total count of red blood cell (RBC), haemoglobin (HGB), haematocrit (HCT) and platelets (PLT). The other blood samples were centrifuged (1500 × g for 12 minutes at 4°C) and the harvested plasma was stored at -20° C for further analysis. Total protein, albumin, cholesterol and triglycerides were determined in the plasma using commercial kits (Biomerieux, France). Serum triiodothyronine (T₃) and thyroxine (T₄) concentrations were measured using commercial ELISA kits (BioCheck[®], USA).

Determination of total antioxidant capacity

The same collected blood samples were used to determine the total antioxidant capacity (mmol/L) using commercial kit (Biodiagnostic[©] for diagnostic and research reagents, Dokki, Giza. Egypt, www.bio-diagnostic.com). This method exploits the ability of antioxidants to reduce hydrogen peroxide (H₂O₂). Determination was performed by the reaction of antioxidants in the sample with a defined amount of exogenously provided H₂O₂. The antioxidants in the sample eliminate a certain amount of the provided H₂O₂. The residual H₂O₂ is quantified colourimetrically by an enzymatic reaction which involves the conversion of 3,5,dichloro-2-hydroxy benzensulphonate to a coloured product.

Caecal microbiota

Caecum with its content from 9 rabbits per subgroup was separated and taken out. Caecum weight and length were measured. The caecal microbiota was characterized by enumerating the E. coli and Salmonella sp. populations. A segment of a 10-cm from each caecal sample was collected. Ten grams of caecal content per sample of different treatments were aseptically taken and homogenized in 90 ml of sterile diluent (0.1% peptone water) using a stomacher (Seward, Model 400, England) for 30 sec. Serial dilutions were prepared in peptone water (ISO 6887-1, 2003). Total aerobic bacterial count and E. coli were determined according to the methods of ISO 4833 (2003) and ISO 4832 (2006), respectively. The method used for Salmonella sp. isolation from different caecum content samples was carried out according to the method of ISO 6579 (2006). A sample of 25 g was pre-enriched in 225 ml of peptone water and incubated at 37°C for 16 to 24 h. For selective enrichment, 1 ml of peptone broth was transferred to 9 ml each of tetrathionate broth and incubated at 42°C for 24 h. From each selective enrichment broth a 5-mm loop full was streaked on selective plates of bismuth sulfite agar and incubated at 37°C for 24 h. Colonies on each flat medium were counted using a colony counter. Results were transformed as colony forming units (CFU) at log₁₀ per gram for the total aerobic bacteria and E. coli. Salmonella sp. was expressed as a percentage of detection.

Statistical analysis

Data were subjected to a two-way ANOVA using JMP Ver. 11 (SAS Institute, 2013) with breed and dietary eucalyptus supplementation level as fixed effects and by fitting into the following mathematical model:

$$Y_{ijk} = \mu + L_i + B_j + (LB)_{ij} + e_{ijk}$$

where:

 Y_{iik} = the observation taken on the k^{th} individual,

 $\mu = \text{overall mean},$

 L_i = the fixed effect of the j^{ih} eucalyptus supplementation level,

 B_i = the fixed effect of the *i*th breed,

 $(\dot{L}B)_{ii}$ = interaction between breed and eucalyptus supplementation level,

 e_{ijk} = random error assumed to be independent normally distributed with mean = 0 and variance = σ^2 .

All results are presented as means and the variability in data was expressed as pooled SEM. The significance of difference among the groups was assessed using Tukey's test. Statistical significance was considered when P<0.05.

Results

Growth performance

Table 1 shows the traits related to growth performance in two rabbit breeds fed a diet supplemented with EL. No significant difference was found among dietary treatments in final weight and weight gain. Rabbits fed a diet supplemented with 0.1% EL consumed significantly (P<0.01) more feed compared with the remaining groups. On the other hand, the group that received 0.2% eucalyptus had significantly lower feed intake compared with control. No significant difference was detected due to EL supplementation on FCR. A negative effect on growth performance resulting from adding high level of EL (0.2%) has been observed.

With regard to breed effect, growth performance of Jabali rabbits was much better than V-line rabbits. A significant improvement in body weight and weight gain (P=0.02 and 0.05, respectively) was observed in Jabali breed as compared to V-line at the end of the experiment. They also significantly consumed less feed and had a better FCR. However, significant interaction (P<0.001) was found for feed intake. Concerning the mortality rate during the experimental period, no significant difference either among dietary groups or between breeds was detected (data not shown).

Cell-mediated immunity

The results regarding the cellular immune response are presented in Table 2. Supplementation of EL at the level of 0.2% significantly (P<0.001) increased cell-

mediated response at all tested times post-injection compared to the control rabbits. Although 0.1% EL inclusion caused no significant increase compared to the control group after 24 h of PHA-P injection, it is worthy to note that the improvement of immune response had a curve linear with EL levels. Regarding breed effect, the results revealed that there was no significant difference between Jabali and V-line breeds. Interaction between EL level and breed did not exhibit a significant difference at all studied times.

Trait	Eucalyptus level (L)			Bree	SEM	P-value			
ITali	0%	0.1%			В	L*B			
Initial weight ¹ (g)	1 399	1 449	1 370	1 342	1 431	44.4	NS	NS	NS
Final weight ² (g)	1 967	2 068	1 952	1 838 b	2 059 a	41.3	NS	*	NS
Gain (g)	593	641	582	496 b	639 a	26.4	NS	*	NS
Feed intake (g)	2 337 B	2 540 A	2 228 C	2 468 A	2 324 B	44.9	**	***	***
FCR	4.59	4.45	4.54	4.97a	4.16 b	0.25	NS	*	NS

Table 1. Effect of eucalyptus level and breed on body weight, gain, feed intake and FCR in rabbits

a, b-values in rows with different letters differ significantly (P<0.05).

A, B, C – values in rows with different letters differ significantly (P<0.01).

 1 Body weight at 10 weeks of age, 2 body weight at 16 weeks of age, FCR – feed conversion ratio, SEM – standard error of the mean.

Carcass characteristics

In the present study, the dressing % in rabbits fed 0.1 EL was significantly (P<0.02) improved compared to the other two groups (Table 3). A negative effect was observed in the group fed a diet containing 0.2% EL for dressing % and mid part %. Rabbits of local breed (Jabali) had significant (P<0.01) improvement in all carcass characteristics, except for fore part %, compared to imported rabbits (V-line). Significant interaction was observed between EL level and breed for carcass weight, dressing % and fore part %. In terms of offal and internal organs, there were no significant differences due to either eucalyptus supplementation or breed (data not shown).

Table 2. Effect of eucaryptus level and breed on cen-mediated minimumty in rabbits										
Trait	Eucalyptus level (L)			Bree	d (B)	SEM	P-value			
11ait	0%	0.1%	0.2%	V-line	Jabali	SEIVI	L	В	L*B	
Swelling difference (mm)										
after 24 h	0.48 B	0.50 B	0.68 A	0.56	0.55	0.027	***	NS	NS	
after 48 h	0.27 B	0.31 B	0.50 A	0.38	0.35	0.023	***	NS	NS	
after 72 h	0.12 B	0.19 B	0.31 A	0.23	0.19	0.019	***	NS	NS	

Table 2. Effect of eucalyptus level and breed on cell-mediated immunity in rabbits

A, B - values in rows with different letters differ significantly (P<0.01).

SEM - standard error of the mean.

Trait	Eucalyptus level (L)			Bree	ed (B)	SEM	P-value				
	0%	0.1%	0.2%	V-line	Jabali	SEM	L	В	L*B		
Carcass weight (g)	1 043.7	1 123.7	1 028.7	909.7 B	1 141.6 A	32.05	NS	***	*		
Dressing (%)	51.9 b	53.2 a	51.0 b	48.60 B	53.69 A	0.58	*	***	***		
Fore part (%)	15.2	15.3	15.0	14.8	15.4	0.22	NS	NS	*		
Mid part (%)	15.1 ab	15.8 a	14.1 b	13.6 B	15.7 A	0.34	*	***	NS		
Hind part (%)	21.5	22.1	21.8	20.2 B	22.7 A	0.28	NS	***	NS		

Table 3. Effect of eucalyptus level and breed on carcass traits in rabbits

a, b - values in rows with different letters differ significantly (P<0.05).

A, B - values in rows with different letters differ significantly (P<0.01).

SEM - standard error of the mean.

Haematological parameters, blood biochemistry and antioxidant capacity

As shown in Table 4, no significant differences in blood haematology were observed due to EL inclusion. However, insignificant increase was found for HGB, RBC (P=0.07 and 0.10, respectively) in rabbits that received a diet supplemented with 0.1% EL.

Table 4. Effect of eucalyptus level and breed on blood haematology in rabbits

Tusit	Eucalyptus level (L)			Bree	d (B)	SEM	P-value			
Trait	0%	0.1%	0.2%	V-line	Jabali	SEM	L B 16 NS * 19 NS NS	L*B		
HGB (gm/dL)	12.0	13.0	12.3	11.5 b	12.8 a	0.26	NS	*	NS	
RBC (10 ⁶ /µL)	5.3	5.6	5.2	5.2	5.4	0.09	NS	NS	NS	
HCT (%)	34.4	35.7	34.2	33.6	35.2	0.68	NS	NS	NS	
PLT (10 ⁶ /mL)	257.9	205.1	263.5	355.2 A	198.7 B	27.26	NS	**	NS	

a, b – values in rows with different letters differ significantly (P<0.05).

A, B - values in rows with different letters differ significantly (P<0.01).

HGB - haemoglobin, RBC - red blood cells, HCT - haematocrit, PLT - platelets.

SEM - standard error of the mean.

Table 5. Effect of eucalyptus level and breed on	blood biochemistry and total antioxidant capacity in
r	bhite

Tabbits											
Trait	Eucalyptus level (L)			Bree	SEM	P-value					
	0%	0.1%	0.2%	V-line	Jabali	SEIVI	L	B NS NS *** NS	L*B		
Total protein (g/dL)	5.9 B	6.2 AB	6.6 A	6.3	6.2	0.11	**	NS	NS		
Albumin (g/dL)	3.9	4.0	4.2	3.9	4.1	0.06	NS	NS	NS		
Globulin (g/dL)	1.9 b	2.2 a	2.4 a	2.3	2.1	0.09	*	NS	NS		
Cholesterol (mg/dL)	111.6	110.9	106.7	137.4 A	94.7 B	6.79	NS	***	NS		
Triglycerides (mg/dL)	154.6	139.9	158.8	144.2	155.2	6.8	NS	NS	NS		
T ₃ (ng/mL)	1.18	1.07	1.36	1.12	1.23	0.08	NS	NS	NS		
$T_4 (\mu g/mL)$	6.35	6.84	5.59	5.78	6.52	0.24	NS	NS	NS		
Total antioxidant capacity	0.98 b	1.09 b	1.83 a	0.95 B	1.65 A	0.14	*	**	NS		

a, b – values in rows with different letters differ significantly (P<0.05).

A, B – values in rows with different letters differ significantly (P<0.01).

SEM - standard error of the mean.

A highly significant (P<0.02) difference between breeds was found for HGB and PLT. The highest value of HGB was recorded in Jabali breed (12.8 mg/dL), while the highest PLT value was recorded in V-line (355.2×10^6 /ml).

The results of blood biochemistry parameters and total antioxidant capacity as affected by EL supplementation and breed are presented in Table 5. Total protein content significantly (P<0.01) increased in rabbits fed a 0.02% EL (6.6 g/dL) compared to the others fed a 0.1% EL (6.2 g/dL) or the control group (5.9 g/dL). Albumin content was not affected by EL supplementation. Regarding globulin content, a significant (P=0.04) increase was found in both groups given EL compared to the control one. However, no significant effect due to EL supplementation on the cholesterol, triglycerides and thyroid hormones was detected. Both rabbit breeds performed similarly for all blood biochemical parameters except for cholesterol content, where V-line had a significantly (P<0.002) higher concentration compared to Jabali breed. Also, a significant (P<0.01) improvement in total antioxidant capacity of Jabali breed was noticed. As shown in Table 5, the activity of total antioxidant capacity in serum of rabbits fed a diet supplemented with 0.2% eucalyptus was significantly (P<0.05) elevated compared with the other groups. However, rabbits receiving lower level of EL did not differ from the control group.

Caecal microbiota

Caecal morphology and microbial profile as affected by EL inclusion and rabbit breed are shown in Table 6. Feeding rabbits on a diet containing 0.2% EL significantly (P<0.01) shortened the length of caecum (49 cm) compared to the unsupplemented group. It is worthy to note that EL supplementation was associated with a reduction of caecal microbiota. Significant (P<0.001) linear reduction in total bacterial count and *E. coli* was observed in both groups given either 0.1% or 0.2% EL compared to the control group. Similar trend was found for the percentage of *Salmonella sp.* detection in both breeds. Due to breed effect, it could be observed that the Jabali rabbits had significantly (P<0.001) shorter caecum and lighter weight % than those of V-line rabbits. Insignificant decrease in total microbial count was noticed in Jabali breed compared to V-line breed.

Trait	Euca	lyptus level	(L)	Bree	SEM	P-value					
11ait	0%	0.1%	0.2%	V-line	Jabali	SEIVI	L	В	L*B		
Caecum											
weight (g)	122.0	118.6	107.9	124.0	111.6	3.39	NS	NS	NS		
length (cm)	53.49 A	51.59 AB	49.01 B	53.84 A	49.97 B	0.64	**	***	NS		
weight (%)	6.2	5.7	5.6	7.0 A	5.3 B	0.22	NS	***	***		
Bacterial count (log10cfu/g)											
total	8.9 A	6.6 B	4.9 C	7.1	6.5	0.43	***	NS	NS		
E. coli	4.9 A	3.6 B	2.5 C	3.6	3.8	0.25	***	NS	NS		
Salmonella sp. ¹	100	80	60	80	80	_	-	_	-		

Table 6. Effect of eucalyptus supplementation and breed on caecal morphology and microbiota in rabbits

A, B, C - values in rows with different letters differ significantly (P<0.01).

SEM - standard error of the mean.

¹Detection percentage.

Discussion

Growth performance

Problems resulting from using antibiotics in animal feed have been frequently reported worldwide. Natural feed additives are greatly encouraged to replace antibiotics for improving growth performance and reducing residues in rabbit meat. However, to our knowledge there is no available literature concerning the effect of EL supplementation on carcass characteristics in rabbits raised under high ambient temperature. Most of the available reports on EL utilization as feed additives have been found in poultry. Dietary inclusion of EL in feeding rabbits did not significantly improve growth performance. These results are in accordance with those found by Ahmed et al. (2005) who reported that dietary EL had no significant effect on live body weight and daily weight gain in growing rabbits. Contrary to our results, Hassan et al. (2011) found a positive effect of EL dietary supplementation on growth performance in Japanese quail. The reason for this discrepancy may be due to the presence of higher levels of tannins in our EL sample. Our findings agree with the results of Farhadi et al. (2017) who stated that using EL powder in broiler at higher level had a deleterious effect on growth performance and deceased feed intake. This may be due to higher concentration of tannins. El-Adawy et al. (2008) reported that the tannins concentration was high and averaged 102.3 g/kg DM in EL. Tannins are present in eucalyptus and usually considered as antinutrients because they interact with dietary proteins and minerals and make them unavailable (Nyman and Björck, 1989; Moore et al., 2005). Moreover, tannins are known to interfere with enzyme activities (Mansoori et al., 2007). Similarly, Abd El-Motaal et al. (2008) stated that the body weight was not affected by EL supplementation in laying hens. Inclusion of EL at low level (0.1) significantly increased feed consumption, while the higher level significantly decreased feed consumption compared with the control group. This may be due to the higher eucalyptus oil administered in this treatment. However, further studies are still needed regarding its oil content and associated effects on feed consumption and feed conversion ratio. On the basis of breed effect, poor growth performance associated with imported breed (V-line) may be attributed to the negative effect of high ambient temperatures during the experimental period. The results of the present study agree with those of Fathi et al. (2017). Similarly, Iraqi et al. (2008) reported that the Egyptian Gabali breed exceeded the V-line breed in its weights and gain. On the other hand, under Saudi environmental conditions, Al-Dobaib (2010) stated that there was no significant difference between body weight at 10 weeks of age of three Saudi lines of rabbits compared with Spanish V-line. No significant interaction was observed between EL level and breed for growth performance.

Cell-mediated immunity

To our knowledge, there are no reports regarding the effect of feeding EL on cellular immunity of rabbits. Our findings revealed that the EL supplementation at level of 0.2% significantly (P<0.0001) improved cellular immunity. These improvements might be associated with tannin content in EL which is involved in enhancing the immune responsiveness via its immunomodulatory effect in growing rabbits (Parisi et al., 2018). Moreover, it is well known that EL has effective anti-inflammatory properties due to its contents of several biological compounds (Barra et al., 2010). In this context, Serafino et al. (2008) proved that Eucalyptus oil extract is able to implement the innate cell-mediated immune response, provide scientific support for an additional use of this plant extract, besides those concerning its antiseptic and anti-inflammatory properties. However, positive effect of eucalyptus on primary antibody response against SRBC in the broilers has been reported (Farhadi et al., 2017). Similar results were reported concerning the enhancement of immune profile in laying hens (Abd El-Motaal et al., 2008) and Japanese quail (Hassan et al., 2011). Laying hens fed a diet supplemented with 0.2% or 0.3% eucalyptus leaves were significantly hyper responder to PHA-P compared to hens given a control diet (Abd El-Motaal et al., 2008). Collectively, by taking into consideration our results in antioxidative properties, it seems more likely that dietary EL may enhance the immune responsiveness and the protection against diseases in growing rabbits under heat stress conditions.

Carcass characteristics

A significant increase in dressing of carcass and mid part occurred in rabbits that received a diet containing 0.1% EL. This trend was not was observed in the rabbits fed a diet containing 0.2% EL compared with the control rabbits. This may be due to increasing tannins amount in 0.2% EL level. On the other hand, Ahmed et al. (2005) reported that dietary EL had no significant effect on carcass traits including heart, liver and kidney percent in growing rabbits. These results are in agreement with those found by Mancini et al. (2019) when quebracho and chestnut were added to growing rabbit's diet. Based on the results of breeds, the superiority of carcass traits in Jabali breed is confirmed by the findings of Fathi et al. (2017), who found a significant improvement in carcass characteristics including dressing %, mid part % and hind part % in Jabali rabbits compared to V-line counterparts.

Haematological parameters and blood biochemistry constituents

No significant differences among dietary treated groups were found in blood haematology parameters. While in quails, Bello (2015) reported the supplementing diet with dried eucalyptus leaves resulted in a significant increase in the various haematological measurements (RBC, HGB and HTC). He attributed this improvement to the fact that eucalyptus contains and serves as a good source of iron, beta-carotene and vitamin C. In agreement with the current results of blood haematology, Fathi et al. (2017) reported that the local breed (Jabali) had higher HGB, RBC and HCT figures than those of the imported breed (V-line). This means that the local breed is more adapted to the hot environmental conditions prevailing in Saudi Arabia. Contrary to the current results, Fathi et al. (2017) reported that the Jabali rabbits had a superior number of platelets when compared with V-line rabbits. This inconsistency may be due to using different stock and varied health condition.

The dietary level of 0.2% EL significantly increased the concentration of blood total protein, and globulin compared to the control group. Ahmed et al. (2005) reported that dietary EL had no significant effect on total protein and significant in-

crease in serum albumin concentration in growing rabbits. In agreement with our findings, Abd El-Motaal et al. (2008) did not detect a significant effect due to EL supplementation on blood cholesterol content of laying hens. Inclusion of EL did not affect blood cholesterol and triglycerides concentrations. Due to breed effect, a significant reduction in blood cholesterol and significant increase in total antioxidant capacity were detected in Jabali breed compared to V-line which may have resulted from poor acclimatization of the imported breed. This fact is confirmed by the findings of Fathi et al. (2017).

Total antioxidant capacity

One of the significant results in the present study is the improvement of the antioxidative properties illustrated by total antioxidant capacity due to dietary supplementation of 0.2% EL (Table 5). The antioxidant properties of EL are related to its phytochemical active ingredients including polyphenols, 1,8-cineole, and tannins which play a vital role in scavenging of the free radicals and inhibiting lipid peroxidation (Luís et al., 2016). Similarly, Chen et al. (2017) indicated that dietary EL polyphenols supplementation increased the serum antioxidant status of laying hens. Also, Liu et al. (2011) observed that superoxide dismutase activity was significantly increased and malondialdehyde was significantly decreased in rabbits fed a diet containing different concentrations of tannins. Recently, there is a great interest in plant polyphenol antioxidants because of their protective effects against different diseases, including cardiovascular, inflammatory and cancer. Therefore, it is of interest to note that the dietary supplementation of 0.2% EL enhanced the antioxidative status, which probably resulted in improving the immunity of growing rabbits under high environmental temperature in the current study.

Caecal microbiota

Interestingly, the results of the present study revealed that dietary EL supplementation reduced the total bacterial count and E. coli (P<0.05, Table 6). In agreement with ceacal morphology and microbial profile of our results, Medic et al. (1992) found some antibacterial activities of eucalyptus species against E. coli. Similarly, Trivedi and Hotchandani (2004) revealed that oil of eucalyptus has antibacterial activity against Gram-positive as well as Gram-negative bacteria resistant to commonly used antimicrobial agents. Additionally, essential oil derived from eucalyptus showed a protective action in broilers against multiple respiratory pathogens mainly Mycoplasma gallisepticum and H9N2 influenza virus infections (Barbour et al., 2006 and 2011). Moreover, Mancini et al. (2019) proved tannins played a role as a protective factor of the intestinal mucosa and as a control of peristaltic activity in presence of digestive disorders. The mechanism of EL action is another approach that may be promising for further research. It is of interest to know how the EL shortens caecum length and reduces pathogenic bacteria. Therefore, it might be illustrated that dietary EL may affect the pathogenic bacteria in intestine during heat stress periods. Taken together, the previous studies obviously indicated that dietary EL supplementation plays a vital role in improving the immune response. Getting such benefits in growing rabbits is a necessity under high ambient temperature.

It could be concluded that supplementation of rabbit diets with either 0.1% or 0.2% EL greatly enhanced cellular immunity and total antioxidant capacity as well as lowered pathogenic bacteria in caecum without penalizing growth performance. Additionally, Jabali breed exhibited a higher production performance than that of the imported breed (V-line).

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Received: 11 XI 2018 Accepted: 8 III 2019