Effect of addition of eucalyptus and rosemary leaves alone or in mixture on nutrient intake and some serum biochemical profile of local Awassi lambs

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Abstract

The present experiment aimed to evaluate the effects of inclusion of dried eucalyptus leaves (*Eucalyptus camaldulensis*) and dried rosemary leaves (*Rosmarinus officinali*s L.) alone or in mixture on nutrient intake, growth performance and some biochemical blood profile in local lambs. Four Awassi male lambs were randomly assigned according to a 4×4 Latin square design to four period and four treatments. The treatments were: Basal diet only without any additive and served as control (T1), 2g eucalyptus leaves. $head^{-1}.day^{-1}$(T2), 2g rosemary leaves. $head^{-1}.day^{-1}$ (T3), 2 g of mixture (1g eucalyptus leaves+1g rosemary leaves). $head^{-1}.day^{-1}$(T4). Animals were fed a concentrate at rate 3% of live body weight, while wheat straw was offered *ad libitum*. Results showed that the intakes of all nutrients were not significantly affected by addition of eucalyptus and rosemary leaves alone or their mixture. Statistical analysis showed that total weight gain, average daily gain and feed conversion ratio enhanced significantly ($P \leq 0.05$) in lambs fed T2 and T3 compared with those lambs fed T1 and T4. The concentrations of serum glucose, total protein, and triglycerides were not affected. While, serum urea and urea nitrogen concentrations were decreased ($P \leq 0.05$) in T2, T3 and compared with T1 and T4. Result also revealed that malonaldehyde levels decreased ($P \leq 0.05$) in T2, T3 and T4 compared with control (T1). It can be concluded from the results that addition either eucalyptus leaves or rosemary leaves as single additive exhibited better performance than mixture additive without adverse effect on lambs health.

Keywords: Awassi lambs, *Eucalyptus camaldulensis*, *Rosmarinus officinali*s, nutrient intake, growth performance, blood parameters.

Introduction

Recent years have seen increasing interest in the possibilities of phytogenic products as natural substitutes for antibiotics and growth promoters in ruminant nutrition, since they might improve health states and enhance animal performance [1,2,3,4]. Among aromatic and medicinal plants are eucalyptus (*Eucalyptus camaldulensis* Dhnn.) leaves and rosemary (*Rosmarinus officinalis* L.) leaves, the active ingredient of these plants is primarily alkaloids, flavonoids, phenolics, terpenes, and essential oils [4, 6]. Eucalyptus (*Eucalyptus camaldulensis*), is a perennial evergreen tree that falls under the genus eucalyptus within the family Myrtaceae [7]. The major components of essential oil eucalyptus leaves were 1,8-cineole (eucalyptol) , *p*-cymene, *γ*-terpinene, and *β*-pinene [6, 8], which showed

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antioxidant, antimicrobial activities [7, 9]. Rosemary (*Rosmarinus officinalis* L.) belongs to the Lamiaceae family and is a perennial aromatic herb that originates from the Mediterranean area [3]. The major components of rosemary essential oil are camphor, 1.8-cineole, α and β-pinene, caryophyllene, and limonene) [10], which showed antibacterial properties against both gram-positive and gram-negative bacteria [11]. Furthermore, the phenolic diterpenes found in rosemary leaves (carnosol, carnosic acid, rosmanol, methyl carnosate, and rosmarinic acid) have a strong antioxidant capacity that has been extensively studied [3,11]. Therefore, we hypothesized that a mixture by equal proportions will be more beneficial on growth performance and animal health, this study was undertaken to evaluate the efficiency of adding eucalyptus and rosemary leaves alone or in mixtures on nutrient intake, growth performance, and serum biochemical parameters of Awassi lambs.

Material and Methods

All animal care and experimental procedures of this study were approved by the Central Committee of Bioethics at the University of Kufa, Iraq. protocol No. 13186/2025). Eucalyptus (*Eucalyptus camaldulensis* Dehn.) leaves were picked from trees growing at University of Kufa, while rosemary (*Rosmarinus officinali*s L.) leaves were obtained from local market in Najaf.

Animals, Diet and Management:

This experiment was conducted at the privet field at Al-Mashkhab District, Al-Najaf Province, Iraq. Four Awassi male lambs with 5-6 months of age, an average weight of 34.77±1.23 kg which were obtained from the local market in Najaf and housed in individual pens (1.5×2m). Each pen was equipped with the roughage, concentrate diet and drinking water container. Concentrate diet were offered twice daily morning and evening at a rate of 3% of live body weight followed with wheat straw *ad libitum.* All lambs were vaccinated against internal and external parasites before start the experiment. Each period was lasted 21 days (14 days preliminary period and 7 days for recoded). The dietary treatments were follows: T1: Basal diet only without any additive; T2: 2g of eucalyptus leaves. head^{-1}. day^{-1}; T3:2g of rosemary leaves. head^{-1}. day^{-1}; and T4:2g of mixture (1g eucalyptus leaves+1 g rosemary leaves). head^{-1}. day^{-1}. the residual concentrate and wheat straw were daily collected and weighted to calculate nutrient intake for each lamb. Animals were weighed weekly during each period, the changes in body weight were recorded from the start and the end of each period (during 7 days).

Chemical analysis

The samples of concentrate diet, wheat straw, eucalyptus leaves and rosemary leaves were ground in laboratory grinder

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with 1mm sieve and analysis for dry matter (DM), Ash, Crude protein (CP), Ether extract (EE), Crude fiber (CF) According to AOAC methods [12]. The organic matter (OM)was calculated 100-ash. The Nitrogen free extract (NFE) was calculated using equation: NFE=OM-(CP+EE+CF). The metabolizable energy was calculated using equation: ME (MJ / kg DM) = 0.012×CP + 0.031×EE + 0.005 ×CF+ 0.014 ×NFE [13]. the chemical composition of concentrate diet, wheat straw, eucalyptus and rosemary leaves used in this study are presented in Table 1.

Table 1. Ingredients and Chemical composition of concentrate diet, wheat straw, Eucalyptus and Rosemary leaves (On DM% basis)

Ingredients	Concentrate diet	Wheat straw	Eucalyptus leaves	Rosemary leaves
Barley grain	45			
Wheat bran	35			
Yellow corn	10			
Soybean meal	8			
Limestone	0.5			
Salt	0.5			
Premix	1			
Chemical composition (%)				
Dry matter (DM)	89.51	92.14	92.47	94.60
Organic matter (OM)	93.87	92.41	94.11	94.79
Ash	6.13	7.59	5.89	5.21
Crude protein (CP)	15.39	2.13	8.60	6.46
Crude fiber (CF)	7.88	41.87	15.96	19.65
Ether extract (EE)	3.30	1.53	3.47	5.50
Nitrogen free extract (NFE)[1]	67.3	46.88	66.08	63.18
Metabolizable energy (MJ/Kg DM)[2]	12.75	9.39	12.16	12.31

[1]NFE= OM- (CP+CF+EE).; [2]ME (MJ / kg DM) = 0.012×CP + 0.031×EE + 0.005 ×CF+ 0.014 ×NFE [13].

Serum Biochemical Profile:

Blood samples 10 ml were collected at the end of each period from the jugular vein of each lamb at 3 h after morning feeding into serum separating tubes. After that the blood Serum were centrifuged at 3000 rpm for 20 min. for obtaining the serum. The collected serum was kept at −18 °C until analyses and transferred to the laboratory in the same day of collection. Serum was analyzed for total protein, urea, urea nitrogen, glucose and triglycerides concentrations and Malonaldehyde concentration (MDA). These biochemical parameters were analyzed by spectrophotometry using commercial kits.

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Statistical Analysis

Data were analyzed as a 4×4 Latin square design using General Linear Model (GLM) procedure of SAS [14]. The statistical model was: $Yijk = \mu + Ai + P_j + T_k + Eijk$, where: μ= is the overall mean; A_i= the effect of animal *i*; Pj= the effect of period j; Ti= the fixed effect of dietary treatments; E_{ijk}= the standard error effect. The significant differences among means were tested using Duncan multiple test range [15], the significant effects were declared at $P \leq 0.05$.

Results and Discussion

Nutrient intake

No significant effect was observed for nutrient intake with feeding eucalyptus, rosemary leaves alone or in combination (Table2). Similar results reported by many studies using eucalyptus leaves as feed additive in lactating buffalo [5, 16], in Ossimi lambs [17], in Swamp Buffalo [18]. Furthermore, [19] stated that addition 0.5 g eucalyptus essential oil.head^{-1}.day^{-1} in Du Han hybrid sheep did not alert the intake of dry matter, organic matter, crude protein, and fiber compared with those in control group. on the other hand, [20] found decrease in feed intake in cattle supplemented with 0,100, 200 g^{-1}.h^{-1}. d^{-1} of eucalyptus leaves. Moreover, no significant different on feed intake when supplementation of rosemary leaves in fattening sheep [21, 22, 23]. In addition, [1] reported increase DM intake (g.day^{-1}) in Karadi lambs fed on 7.5g/Kg of rosemary leaves but no significant differences when expressed intake as g.kg^{-1} metabolic body weight. Moreover, [24] reported no significant difference in DM, CP and ME intake when supplemented lambs diet with 5 and 7.5 g/kg of rosemary leaves. Numerous researches have suggested the influence of supplementary rosemary leaves or rosemary oil on feed intake and palatability; nevertheless, their findings were variable and inconsistent.[25] reported that that feed intake of Damascus goats where not affect when supplemented their diet with 1% rosemary leaves, while [26] reported increased DM, OM, CP and fiber in Dorper lambs fed rosemary leaves at 1%. The inconsistent effect of Rosemary essential oils on feed intake were also reported depending upon the type and doses as well as their properties may differently depending on the chemical composition of their active components [3].

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Table 2. Effect of addition of eucalyptus and rosemary leaves alone or in mixture on nutrient intake in Awassi lambs (Mean ± SE)

Intake ($g.day^{-1}$)	Treatments				P
	T1 Control ($0\ g.h^{-1}.d^{-1}$)	T2 Eucalyptus ($2\ g.h^{-1}.d^{-1}$)	T3 Rosemary ($2\ g.h^{-1}.d^{-1}$)	T4 Mixture ($2\ g.h^{-1}.d^{-1}$)	
TDMI	1273.84±86.99	1226.01±95.89	1255.05±127.25	1224.10±138.01	N.S.
TOMI	1249.17±86.38	1202.26±75.28	1230.90±125.74	1200.10±136.29	N.S.
TCPI	162.23±4.80	156.18±5.03	158.80±10.59	157.72±11.83	N.S.
TCFI	229.08±33.83	220.36±28.85	228.41±39.44	215.33±41.22	N.S.
TEEI	38.63±1.77	37.19±1.62	37.92±3.09	37.36±3.40	N.S.
TNFEI	840.29±7.60	808.81±4.29	826.36±7.26	826.39±8.31	N.S.
TMEI	16.17±0.94	15.57±0.84	15.91±1.47	15.58±1.61	N.S.

N.S.: Not significant; T1: Control without additives, T2: 2 g eucalyptus leaves.$head^{-1}.day^{-1}$, T3: 2 g rosemary leaves.$head^{-1}.day^{-1}$, T4: 2 g Mixture (1 g eucalyptus leaves + 1 g rosemary leaves). $head^{-1}.day^{-1}$ TDMI: Total dry matter intake, TOMI: Total organic matter intake, TCPI: Total crude protein intake, TCFI: Total crude fiber intake, TEEI: Total ether extract intake, TNFEI: Total nitrogen free extract .

Growth performance

Growth performance of lambs although it made at a shot-term feeding 7 days in the current study. [27] stated that growth performance of Dorper lambs when measured as a short-term feeding (7 days) or at long term feeding (90 days) are similar. Results in Table 3 revealed that total weight gain (TWG) and average daily gain (ADG) were increased ($P \leq 0.05$). The feed conversion ratio (FCR) was improved ($P \leq 0.05$) in lambs fed eucalyptus leaves and rosemary leaves alone compared with those fed mixture and control (Table 3). These results are in harmony with results reported by [17] when fed sheep low levels ($0.1g.kg^{-1}$ live body weight. day^{-1}) of eucalyptus leaves and [2] also reported increased total and daily body weight of calves fed eucalyptus leaves ($22\ g.\ h^{-1}.\ d^{-1}$) compared with those fed control diet and contributed that to increase the digestibility of nutrient or enhanced health of calves. Similarly, TWG, ADG and FCR enhanced when rosemary leaves added to the diet of Karadi sheep [1, 24]. In Barki lambs [23]. However, [21] did not found improvement in body weight gain and average daily gain in Saidi sheep fed 2.5% of Rosemary leaves. Furthermore, [26] reported that final body weight, total weight gain and average daily gain increased leaves compared with those lambs fed 1%Nigella sativa seed and combination. The variable effect on fattening performance in the different studies might be strong different in type, source and concentrations of essential oil and active components among experiment [4, 9].

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Table 3. Effect of addition of eucalyptus and rosemary leaves alone or in mixture on growth performance in Awassi lambs (Mean ± SE)

Growth Performance (7 days)	Treatments				P
	T1 Control (0 $g.h^{-1}.d^{-1}$)	T2 Eucalyptus (2 $g.h^{-1}.d^{-1}$)	T3 Rosemary (2 $g.h^{-1}.d^{-1}$)	T4 Mixture (2 $g.h^{-1}.d^{-1}$)	
IBW (kg)	34.94±3.78	34.45±2.73	34.61±2.18	35.08±1.86	N.S.
FBW (kg)	35.83±3.80	35.73±2.65	35.80±2.22	35.78±1.80	N.S.
TWG (kg)	$0.90±0.11^{bc}$	$1.28±0.17^{a}$	$1.19±0.09^{ab}$	$0.71±0.08^{c}$	*
ADG ($g.day^{-1}$)	$127.86±15.61^{bc}$	$183.21±24.97^{a}$	$169.29±12.92^{ab}$	$101.07±11.26^{c}$	*
FCR	$10.10±1.31^{ab}$	$6.76±0.83^{c}$	$7.36±0.83^{bc}$	$11.82±0.93^{a}$	*

Means within same raw with different letter are significantly different (P≤0.05), N.S.: Not significant; *: (P≤0.05);

T1: Control without additives, T2: 2 g eucalyptus leaves.$head^{-1}.day^{-1}$, T3: 2 g rosemary leaves.$head^{-1}.day^{-1}$, T4: 2 g Mixture (1 g eucalyptus leaves + 1 g rosemary leaves). $head^{-1}.day^{-1}$. IBW: initial body weight, FBW: Final body weight, TWG: total weight gain, ADG: Average daily gain, FCR: feed conversion ratio.

Serum Biochemical parameters:

The effect of addition of eucalyptus leaves and rosemary leaves alone or in mixture on serum biochemical parameters are presented in Table 4. Results revealed that the concentrations of serum glucose (SG), total protein (STP), triglyceride (STG) were not significantly different among treatments. These results are in agreement with results obtained [16] who found that addition eucalyptus leaves as did not significantly affect blood glucose concentration in dairy cows. [22] also reported that SG, STP and STG concentration were not affected by addition 0.5, 1% of rosemary leaves to Rahmani lambs ration. Similarly, [28] reported that SG, STP concentrations did not affect significantly when Ghezel sheep fed 100, 200 and 400 $mg.day^{-1}$ of rosemary essential oil. However, [25] and [29] reported increased SG concentrations in Damascus goat and Norduz lambs when fed rosemary leaves and rosemary essential oils. Furthermore,[1] reported that supplemented Karadi lambs with rosemary leaves significantly increased the concentrations of SG.

Results in Table 4 also showed that serum urea (SU) and urea nitrogen (SUN) were decrease (P≤0.05) in lambs fed eucalyptus leaves (T2) and rosemary leaves (T3) compared with those fed T1 (control) and T4 (mixture). These results are in harmony with results by [5] when fed eucalyptus leaves to Buffalo. [1, 21, 24] found a significant decrease in SUN and SU when fed sheep on rosemary leaves. However, SUN and SU where not affect by adding eucalyptole or rosemary leaves [15, 25]. According to [1] the highest response in live weight growth was associated with reduced serum urea concentration, which may imply improve efficiency utilization of the nutrients available in the diets. The current study results showed significant (P≤0.05) lower malonaldehyde (MDA) levels with eucalyptus, rosemary and

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mixture, with superiority decreased (P≤0.05) with rosemary leaves (Table 4), this may be due to strong antioxidant activity presence in rosemary leaves. these results are in harmony with previous results by [22] in Rahmani lambs fed rosemary and [30] when calves fed rosemary leaves. All serum biochemical paraments in the current study are within normal rage of healthy lambs.

Table 4. Effect of addition of eucalyptus and rosemary leaves alone or in mixture on blood biochemical parameters in Awassi lambs (Mean ± SE)

Serum Biochemical Parameter	Treatments				P
	T1 Control (0 $g.h^{-1}.d^{-1}$)	T2 Eucalyptus (2 $g.h^{-1}.d^{-1}$)	T3 Rosemary (2 $g.h^{-1}.d^{-1}$)	T4 Mixture (2 $g.h^{-1}.d^{-1}$)	
SG (mg. dL^{-1})	91.82±8.58	91.44±8.36	95.75±5.03	98.01±8.75	N.S.
SU (mg. dL^{-1})	48.63±5.46^{a}	39.75±5.27^{b}	40.45±4.90^{b}	40.64±7.19^{ab}	*
STP (g. dL^{-1})	5.29±0.45	4.95±0.30	4.93±0.48	4.80±0.37	N.S.
SUN (mg. dL^{-1})	22.96±0.55^{a}	18.55±0.46^{b}	18.88±0.22^{b}	19.97±1.36^{ab}	*
STG (mg. dL^{-1})	22.25±0.74	21.75±0.61	21.25±0.78	20.75±1.03	NS
MDA (nmol. L^{-1})	5.16±1.43^{a}	4.86±3.38^{b}	4.43±3.30^{c}	4.39±1.01^{bc}	*

Means within same raw with different letter are significantly different (P≤0.05), N.S.: Not significant; *: (P≤0.05);

T1: Control without additives, T2: 2 g eucalyptus leaves.$head^{-1}$.day^{-1}, T3: 2 g rosemary leaves.$head^{-1}$.day^{-1}, T4: 2 g Mixture (1 g eucalyptus leaves + 1 g rosemary leaves). head-1.day^{-1}. SG: Serum glucose; SU: Serum Urea; STP: Serum total protein; SUN: Serum urea nitrogen; STG: Serum triglyceride; MDA: Malondialdehyde

Conclusion

Based on this study, it could be concluded that addition dried eucalyptus, rosemary leaves powder could improve health status and enhanced growth performance of Awassi lambs. However, further research is still required to optimize dosage and understand long term effects.

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