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# Synergistic Influences of Essential Oils Blends and Their Active Substances on Blood Biochemical Constituents Under Climate Change of Growing Saidi Lambs.

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## Abstract

This study investigated the synergistic effect of two blends of essential oils from medicinal plants as a phytogenic additive on the active substances and on various blood serum parameters of Saidi lambs. The initial additive comprised a 1:1:1 Rosemary, Thyme, and Peppermint oil mixture (RTPOM). The second additive comprised a 1:1:1 Lemongrass, Marjoram, and Black Seed oil mixture (LMBOM). The two groups were orally given to Saidi lambs by syringe. The 1st group (5 lambs) was administered a control diet devoid of essential oil blends supply (concentrate feed mixture to roughage ratio of 70:30); the 2nd group (5 lambs) was administered a control diet supplemented with 0.03% RTPOM on a DM basis; and the 3rd group administered a control diet supplemented with 0.03% LMBOM on a DM basis. The experiment spanned 180 days. GC-MS analyzed all essential oils of medicinal plants for their bioactive substances. Results revealed that the principal bioactive substances for RTPOM were eucalyptol and thymol for Rosemary; thymol and p-Cymene for Thyme; and (-)-carvone and limonene for Peppermint. The principal bioactive substances for LMBOM were citral and 1,3,8-p-menthatriene for Lemongrass; terpinen-4-ol and γ-terpinene for Marjoram; and heptane, 2,2,4,6,6-pentamethyl-, thymoquinone, and benzene,1-methyl-3-(1-methylethyl)- for Black Seed. A substantial rise in serum total protein ($P=0.0069$) and globulin ($P=0.0453$) and a decrease in cholesterol ($P=0.0429$) were seen with the LMBOM and RTPOM rations. Furthermore, the incorporation of essential oil blends led to a significant reduction in rectal temperature ($P<0.0001$) in a hot and cool environment compared to the lambs in control.

**Keywords:** Essential oils; active substances; blood parameters; lambs and rectal temperature.

## Introduction

Essential oils are naturally occurring biological chemicals prevalent in various plant tissues. Their salient fragrance and bioactive properties possess diverse biological effects, which may influence the performance of ruminants.

Botanical essential oils can differ substantially in composition but generally consist of two chemical categories: terpenes (including their oxidized derivatives, known as terpenoids) and shikimates. The latter is less common and mainly exists as phenylpropanoids **[1]**. Notably, the antimicrobial characteristics of essential oils have been recognized for decades, and numerous scientific researches have been conducted **[2, 3, 4, and 5].** Additionally, the utilization of essential oils as feed additives may enhance gut microbiota health and bolster ruminant immune responses, leading to sustainable and healthier food for human consumption **[6]**.

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Several studies were done on the effects of herbal plants alone, such as Rosemary (*Rosmarinus officinalis*), Thyme (*Thymus vulgaris* L.), Peppermint (*Mentha piperita*), Lemongrass (*Cymbopogon citratus*), Marjoram (*Origanum majorana*), and Black seed (*Nigella sativa*) on serum blood biochemical constituents. They enhanced total protein, albumin, and globulin. In this context, these herbal plants lowered blood cholesterol levels and enhanced certain liver and kidney functions in ruminants **[7, 8, 9, 10, and 11]**.

Recent scientific interest has focused on the amalgamation of many natural additives, which demonstrated superior results compared to individual natural additives, likely due to the synergistic effects among the herbal components that improve the efficacy of their dietary incorporation **[12, 13, and 14]**. Furthermore, feeding a combination of essential oils increases antimicrobial activities, optimizes ruminal fermentation, and enhances animal performance compared to the individual additives **[15, 16, and 17]**.

Climate change may have potential consequences for animal productivity and growth, as well as the quantity and quality of available feed, water sources, and overall animal health **[18]**. Rearing small ruminants is a relevant activity in arid and semi-arid regions, as these animals are adapted to hot climates, possessing capabilities for survival, production, and reproduction **[19].** Evidence strongly supports the notion that heat stress significantly reduces sheep productivity **[20].** Physically, growing lambs are particularly susceptible to thermal stress **[21].** Reportedly, the utilization of feed additives, such as antioxidants, is the most often embraced approach to mitigate thermal stress economically **[22]**. Phytogenics, short for bioactive substances derived from herbal plants, have been employed in human and animal nutrition to augment performance and mitigate disorders in situations associated with thermal stress **[23]**.

Considering the possible great active substances significance of medicinal plants and, to enhance the efficacy of essential oils, furthermore, the adverse impacts of climate change and mitigating its severity, this study aimed to test the ability of two blends of essential oils as a phytogenic additive about the active substances and influences on some blood biochemical constituents of growing Saidi lambs in hot and cool conditions.

## MATERIALS AND METHODS

### Lambs, experimental procedures, and diets:

The experiment spanned 180 days and involved 15 male Saidi lambs aged 4-5 months of 18.47 ± 0.45 kg body weight. The animals were assigned to three experimental groups with a completely randomized approach. Before the trial's initiation, all animals underwent treatment for internal and external parasites and vaccination against prevalent infectious diseases. All groups received isonitrogenous and isoenergetic diets following **NRC [24]**, with daily rations formulated to fulfill maintenance and production requirements. The daily ration was divided into two equal portions, administered at 8:00 and 16:00, guaranteeing the animal's unrestricted access to water. Furthermore, minerals and vitamin blocks were accessible for unrestricted selection**.**

Throughout the experiment, the 1st group was the control (received 70% concentrate feed mixture (CFM) with 30% alfalfa hay without any oral administration); the 2$^{nd}$ (control plus a 1:1:1 Rosemary, Thyme, and Peppermint essential oil mixture); and the 3$^{rd}$ group (control plus a 1:1:1 Lemongrass, Marjoram, and Black seed essential oil mixture) were orally administered in a dose of

0.03% on a dry matter (DM) basis to lambs immediately prior to feeding throughout the entire 180 days experimental period. Feed items were evaluated following **A.O.A.C. [25]**. The chemical composition of the experimental feed is shown in Table 1.

**Table 1. Chemical composition of the diet offered to Lambs (% DM basis).**

| Composition | Dry matter % | Organic matter % | Crude protein % | Crude fiber % | Ether extract % | Nitrogen Free Extract % | Ash, % |
|---|---|---|---|---|---|---|---|
| **CFM*** | 93.76 | 83.89 | 15.06 | 16.26 | 3.06 | 49.51 | 16.11 |
| **Alfalfa hay** | 94.23 | 79.14 | 11.95 | 25.18 | 2.70 | 39.32 | 20.86 |
| **Basal diet** | 93.90 | 82.47 | 14.13 | 18.94 | 2.95 | 46.45 | 17.53 |

**CFM= Concentrate feed mixture; consists of per ton: 500 yellow corn, 200 wheat bran, 150 cotton seed meal, 125 soybean meal, 10 sodium chloride, 10 calcium carbonate, and 5 minerals plus vitamins additives.*

**Analysis of essential oils:**

Gas chromatography-mass spectrometry (GC-MS) instrument stands were used for essential oil analysis to identify the active substances of each essential oil individually. The analysis was conducted at the Chemistry Department, Faculty of Science, Aswan University, Egypt. The components were identified by comparing their mass spectra with those established by NIST21 and NIST107 **[26]** and comparing them with literature sources' data **[27]**. The co-chromatography process frequently exposes essential oils to genuine chemicals.

**Blood samples and biochemical analysis:**

Blood sampling was obtained monthly before the morning feeding from all lambs by simple vein puncture of the jugular vein (10 ml of blood per animal) into clean glass tubes. Upon collection, samples were promptly placed on ice and centrifuged at 2,500 rpm for 20 minutes at 4°C to isolate serum. Serum samples were frozen at -20ºC until assayed. The blood serum was analyzed for total protein, albumin, and globulin, cholesterol triglycerides, cholesterol, glucose, creatinine, uric acid, urea, AST and ALT. Blood analysis was conducted in the Animal Production Laboratory, Department of Animal and Poultry Production, Faculty of Agriculture and Natural Resources, Aswan University, Aswan, Egypt.

**Meteorological data and physiological parameters:**

Air temperature (AT) and relative humidity (RH%) inside the farming building were measured three times daily at 8.00, 12.00, and 15.00 hours using a digital thermometer. Temperature-humidity index (THI) values under Egyptian conditions were calculated using the following equation **[28]:**

$$THI= AT - [(0.31 - 0.31 \times RH/100) \times (AT - 14.4)]$$

The obtained values indicate the following: The values below 22.2 indicate no heat stress, those between 22.2 and 23.3 indicate moderate heat stress, and those between 23.3 and 25.6 indicate severe heat stress and those exceeding 25.6 indicate extreme heat stress. The rectal temperature was measured weekly during the experiment using a mercury thermometer.

**Analytical statistics**:

An analysis of variance (ANOVA) was employed to examine the collected data using the standard linear modeling approach **[29]**. Statistical significance was determined using **[30]** multiple comparison tests with a significance level of P<0.05.

## Results and discussion

### Active substances:

The results of the GC-MS chromatogram analysis of *R. officinalis, T. vulgaris* L., and *M. piperita* essential oils revealed 21, 10, and 7 active compounds, respectively (Table 2). Results indicated that the higher area percentage principal bioactive substances were Eucalyptol and thymol for *R. officinalis*; thymol and p-cymene for *T. vulgaris* L.; and (-)-carvone and Limonene for *M. piperita*. Several studies suggest that thymol and (-)-carvone possess diverse biological effects, including attenuation of oxidative stress, antimicrobial, anti-inflammatory, preservation of anti-cancer activity, a spasmolytic effect, and hepatocyte integrity **[31, 32, 33, 34, and 35]**.

**Table 2. Principal active substances of *Rosmarinus officinalis, Thymus vulgaris* L., and *Mentha piperita* evaluated essential oils using GC-MS analysis.**

| Compound Name | R. Time | % of total peak area | | |
|---|---|---|---|---|
| | | ***Rosmarinus officinalis*** | ***Thymus vulgaris* L.** | ***Mentha piperita*** |
| Eucalyptol | 4.958 | 35.74% | - | 7.06% |
| Thymol | 7.467 | 16.56% | 36.75% | - |
| (1S)-2,6,6-Trimethylbicyclo[3.1.1]hept-2-ene | 3.722 | 9.78% | - | - |
| (+)-2-Bornanone | 6.245 | 7.49% | - | - |
| Bicyclo[5.2.0]nonane, 2-methylene-4,8,8-trimethyl-4-vinyl- | 8.62 | 3.60% | - | - |
| endo-Borneol | 6.442 | 3.58% | - | - |
| o-Cymene | 4.858 | 3.37% | - | - |
| Camphene | 3.936 | 3.25% | - | - |
| D-Limonene | 4.914 | 1.96% | - | 11.23% |
| Cyclohexene, 1-methyl-4-(1-methylethylidene | 6.657 | 1.82% | - | - |
| Estragole | 6.715 | 1.75% | 4.89% | - |
| Terpinen-4-ol | 6.542 | 1.53% | - | - |
| γ-Terpinene | 5.263 | 1.53% | 3.19% | - |
| D-Carvone | 7.144 | 1.50% | - | - |
| Phenol, 2-methyl-5-(1-methylethyl)- | 7.558 | 1.49% | - | - |
| β-Ocimene | 5.683 | 1.22% | - | - |
| Heptane, 2,2,4,6,6-pentamethyl- | 4.422 | 0.98% | 7.10% | 5.45% |
| l-Menthone | 6.309 | 0.90% | - | - |
| 2,6-Dimethyldecane | 5.208 | 0.73% | - | - |
| β-Pinene | 4.296 | 0.64% | - | - |
| Naphthalene, 1,2,3,5,6,8a-hexahydro-4,7-dimethyl-1-(1-methylethyl)-, (1S-cis)- | 9.478 | 0.59% | - | - |
| p-Cymene | 4.857 | - | 24.27% | - |
| (+)-3-Carene | 5.263 | - | 10.36% | - |
| Phenol, 2-methyl-5-(1-methylethyl)- | 7.56 | - | 3.68% | - |
| Benzene, 1-ethyl-2,3-dimethyl- | 4.962 | - | 3.66% | - |
| 3-Carene | 5.685 | - | 3.36% | - |
| Decane, 3,6-dimethyl- | 5.208 | - | 2.75% | - |
| (-)-Carvone | 7.141 | - | - | 69.01% |
| α-Pinene | 3.728 | - | - | 2.91% |
| (-)-β-Bourbonene | 8.324 | - | - | 2.36% |
| Undecane, 5,7-dimethyl | 5.211 | - | - | 1.99% |

Furthermore, recent studies have highlighted that eucalyptol and p-cymene possess anti-inflammatory, anti-oxidative, anti-apoptotic, immune-regulatory, and nephroprotective properties **[36, 37, and 38]**. Notably, limonene was identified as a possible therapeutic agent for alleviating intestinal inflammation and maintaining gastrointestinal health **[39]**.

The results of the GC-MS chromatogram analysis of *Cymbopogon citratus, Origanum majorana*, and *Nigella sativa* essential oils revealed 14, 12, and 7 compounds, respectively (Table 3). Results indicated that the principal bioactive substances with a higher area percentage were citral and 1,3,8-p-menthatriene for *Cymbopogon citratus*; terpinen-4-ol and γ-terpinene for *Origanum majorana*; and heptane, 2,2,4,6,6-pentamethyl- and thymoquinone for *Nigella sativa*. Evidence suggests that citral, terpinene-4-ol, and thymoquinone improved efficient antimicrobial activity, anti-inflammatory effects, liver function, and antioxidant enzyme levels **[40, 41, 42, 43, and 44]**.

**Table 3. Principal active substances of *Cymbopogon citratus, Origanum majorana*, and *Nigella sativa* evaluated essential oils using GC-MS analysis.**

| Compound Name | R. Time | % of total peak area | | |
|---|---|---|---|---|
| | | ***Cymbopogon citratus*** | ***Origanum majorana*** | ***Nigella sativa*** |
| Eucalyptol | 4.958 | 2.86% | - | - |
| (1S)-2,6,6-Trimethylbicyclo[3.1.1]hept-2-ene | 3.722 | - | 4.73% | - |
| Bicyclo[5.2.0]nonane, 2-methylene-4,8,8-trimethyl-4-vinyl- | 8.62 | - | 7.24% | - |
| o-Cymene | 4.858 | - | 9.29% | - |
| Cyclohexene, 1-methyl-4-(1-methylethylidene | 6.657 | - | 2.88% | - |
| Terpinen-4-ol | 6.542 | - | 30.44% | - |
| γ-Terpinene | 5.263 | - | 12.14% | 7.32% |
| Heptane, 2,2,4,6,6-pentamethyl- | 4.422 | 9.18% | 3.46% | 27.06% |
| 2,6-Dimethyldecane | 5.208 | - | - | - |
| β-Pinene | 4.296 | 6.91% | - | - |
| (+)-3-Carene | 5.263 | - | 4.89% | - |
| α-Pinene | 3.728 | - | - | 13.17% |
| Citral | 7.318 | 24.10% | - | - |
| 1,3,8-p-Menthatriene | 7.078 | 22.43% | - | - |
| Nonadecane, 2-methyl | 11.373 | 5.89% | - | - |
| Tetratetracontane | 9.089 | 4.92% | - | - |
| 2-Isopropyl-5-methyl-1-heptanol | 7.369 | 4.12% | - | - |
| 2-Bromotetradecane | 9.494 | 3.63% | - | - |
| 7,9-Di-tert-butyl-1-oxaspiro(4,5)deca-6,9-diene-2,8-dione | 14.561 | 3.54% | - | - |
| Hexadecane | 10.057 | 3.33% | - | - |
| 2-Bromo dodecane | 11.935 | 3.22% | - | - |
| (E)-β-Famesene | 7.166 | 3.05% | - | - |
| Heptadecane | 11.197 | 2.84% | - | - |
| (+)-4-Carene | 5.715 | - | 8.32% | - |
| Benzene, 1-methyl-3-(1-methylethyl)- | 4.858 | - | 8.22% | 16.21% |
| 1,3-Cyclohexadiene, 1-methyl-4-(1-methylethyl)- | 4.766 | - | 5.96% | - |
| Caryophyllene | 8.62 | - | 2.45% | - |
| Thymoquinone | 7.182 | - | - | 16.38% |
| Undecane, 5,7-dimethyl- | 5.208 | - | - | 14.10% |
| Tetradecane, 2,6,10-trimethyl- | 7.372 | - | - | 5.77% |

**Blood biochemical constituents:**

The results of the blood serum parameters are displayed in Table 4. Data revealed that both RTPOM and LMBOM dietary supplementation had significant effects on immunity functions, such as total proteins (*P=0.0069*) and globulin (*P=0.0453*), in comparison to the control. At the same time, a significant decrease in serum cholesterol (*P=0.0429*) was seen with the LMBOM and RTPOM supplementation. Conversely, no significant differences were noticed (*P>0.05*) for serum albumin, triglycerides, and glucose. Furthermore, critical physiological functions, including liver function indicated by AST and ALT enzyme levels, as well as kidney function represented by urea, uric acid, and creatinine concentrations, were not adversely or significantly (*P>0.05*) influenced by the dietary supplementation of RTPOM or LMBOM in growing Saidi lambs. All results of blood serum components fell within the established normal levels **[45].**

**Table 4. Effect of administration of essential oil blends on serum biochemical parameters of growing Saidi lambs.**

| Item | Experimental rations | | | SEM | *p value* |
|---|---|---|---|---|---|
| | Control | RTPOM | LMBOM | | |
| Total Protein g/dl | 6.73[b] | 7.53[a] | 7.35[a] | 0.11 | 0.0069 |
| Albumin g/dl | 3.94 | 4.12 | 4.04 | 0.04 | 0.2481 |
| Globulin g/dl | 2.79[b] | 3.42[a] | 3.31[a] | 0.12 | 0.0453 |
| Urea mg/dl | 43.27 | 43.96 | 46.70 | 1.23 | 0.269 |
| Uric acid mg/dl | 24.66 | 27.59 | 21.46 | 2.15 | 0.5147 |
| Creatinine mg/dl | 1.72 | 1.67 | 1.67 | 0.07 | 0.9481 |
| AST U/l | 130.39 | 130.41 | 115.24 | 3.36 | 0.1037 |
| ALT U/l | 30.99 | 31.63 | 30.99 | 0.70 | 0.9147 |
| Cholesterol mg/dl | 88.00[a] | 78.31[b] | 80.27[b] | 1.84 | 0.0429 |
| Triglycerides mg/dl | 116.22 | 133.53 | 126.28 | 6.90 | 0.597 |
| Glucose mg/dl | 122.12 | 116.30 | 112.41 | 4.03 | 0.6212 |

[a, and b ….] Values within the same row followed by different superscript letters indicate statistically significant differences ($p < 0.05$).

The current results findings demonstrate that blood constituents are strongly associated with metabolic processes and are affected by external factors, including nutrition, climate, and management practices. Serum total protein indicates the animal's nutritional status and positively correlates with dietary protein availability **[46]**; this may result from enhanced protein absorption and improved synthesis of ruminal microbial protein. At that, globulin is enhanced due to this blend of essential oils and their synergistic influence, including steroidal flavonoid terpenes that promote appropriate cortisol secretion. Under basal conditions, globulin binds to 80% of the circulating cortisol. Furthermore, cortisol has pleiotropic actions on metabolic, cardiovascular health, neurocognitive processes, and immunomodulation **[47, 48, and 49]**. The reduction in blood serum cholesterol concentration of growing Saidi lamb-fed rations treated with LMBOM and RTPOM may be linked to a reduction in hepatic lipid buildup or the suppression of hepatic cholesterol biosynthesis. Additionally, several studies have elucidated the reduction in serum cholesterol status by adding medicinal plant essential oils, potentially attributable to the regulatory influence of terpene derivatives on sterol regulatory element-binding protein-1c, leading to reduced transcription and increased degradation of hepatic 3-hydroxy-3-methylglutaryl coenzyme A. reductase (statins) serves as the principal mechanism for cholesterol synthesis **[50, 51, and 52]**.

The results obtained are congruent with our earlier results **[12]**, which showed that supplementation of a 1:1:1 mixture of dried Rosemary, Thyme, and Peppermint leaves significantly influenced serum levels of total protein and globulin. Moreover, there was no effect on albumin, glucose, kidney, and liver function in Saidi lambs. Recently, adding a 1:1 Peppermint and Rosemary essential oil mixture significantly improved plasma total protein and globulin in diet-fortified calcium soap from the soybean oil of sheep **[53]**. In the same way, oral supplementation of a 1:1 Thyme and Garlic oil blend enhanced serum total protein, globulin, kidney, and liver functions. Also, they decreased serum cholesterol compared to the control diet of Damascus goats **[54]**.

**Meteorological data and physiological parameters:**

In the current study, growing Saidi lambs were raised during hot and cool months under hot desert climate conditions (Table 5). Indeed, the most significant increases in air temperature (AT) data inside the building occurred during the first and third periods of the experiment (November, December, May, and April), while it dropped to the lowest level during the second period (January and February). The relative humidity (RH%) rose during the first and second periods but fell to a minimum in the third period. Based on the above, the temperature and humidity index (THI) was determined to peak during the third period, followed by the first and, subsequently, the second period. Additionally, it is noteworthy that the addition of RTPOM and LMBOM orally administered to the lambs significantly reduced the rectal temperature in the morning and afternoon throughout the experiment (Table 6).

Observations reveal that the lambs experienced severe heat stress in the third period (March and April), with THI values of 25.6 categorized as extreme heat stress. However, the growing Saidi lambs experienced moderate and absent heat stress during the first and second periods (November to February), respectively **[28].**

The Temperature-Humidity Index (THI) is a commonly used metric to assess the impact of heat stress. It combines temperature and relative humidity **[55]**. It is often divided into various ranges to represent the severity of heat stress experienced by small ruminants **[56].**

**Table 5. Air temperature (AT), relative humidity (RH) and temperature humidity index (THI) throughout the trial duration.**

| Item | Time | Period 1 | Period 2 | Period 3 | SEM | *p value* |
|---|---|---|---|---|---|---|
| AT | 08:00 AM | 20.52$^b$ | 15.73$^c$ | 23.87$^a$ | 0.30 | <0.0001 |
| | 12:00 PM | 26.00$^b$ | 22.98$^c$ | 33.17$^a$ | 0.58 | <0.0001 |
| | 03:00 PM | 27.97$^b$ | 25.25$^c$ | 35.35$^a$ | 0.39 | <0.0001 |
| | Means | 24.83$^b$ | 21.32$^c$ | 30.79$^a$ | 0.42 | <0.0001 |
| RH | 08:00 AM | 57.28$^a$ | 57.28$^a$ | 44.73$^b$ | 0.66 | <0.0001 |
| | 12:00 PM | 50.53$^a$ | 49.93$^a$ | 36.27$^b$ | 0.38 | <0.0001 |
| | 03:00 PM | 46.35$^a$ | 42.75$^b$ | 30.23$^c$ | 0.67 | <0.0001 |
| | Means | 51.39$^a$ | 49.99$^a$ | 37.08$^b$ | 0.57 | <0.0001 |
| THI | 08:00 AM | 19.69$^b$ | 15.55$^c$ | 22.23$^a$ | 0.25 | <0.0001 |
| | 12:00 PM | 24.20$^b$ | 21.63$^c$ | 29.44$^a$ | 0.30 | <0.0001 |
| | 03:00 PM | 25.69$^b$ | 23.30$^c$ | 30.81$^a$ | 0.29 | <0.0001 |
| | Means | 23.19$^b$ | 20.16$^c$ | 27.49$^a$ | 0.28 | <0.0001 |

$^{a, b, and c ....}$ Values within the same row followed by different superscript letters indicate statistically significant differences ($p < 0.05$).

**Table 6. Influence of administration of essential oil blends on rectal temperatures of lambs throughout the trial duration.**

| Period | Timing | Experimental rations | | | SEM | *p value* |
|---|---|---|---|---|---|---|
| | | Control | RTPOM | LMBOM | | |
| Period 1 | Morning | 39.73[a] | 39.25[b] | 39.37[b] | 0.05 | 0.0001 |
| | Afternoon | 39.84[a] | 39.57[b] | 39.58[b] | 0.05 | 0.0389 |
| Period 2 | Morning | 39.46[a] | 39.18[b] | 39.11[b] | 0.04 | 0.0004 |
| | Afternoon | 39.53[a] | 39.21[b] | 39.25[b] | 0.04 | 0.0006 |
| Period 3 | Morning | 39.57[a] | 39.03[b] | 38.87[c] | 0.05 | <0.0001 |
| | Afternoon | 39.59[a] | 39.12[b] | 39.03[b] | 0.05 | <0.0001 |

a, b, and c …. Values within the same row followed by different superscript letters indicate statistically significant differences ($p < 0.05$).

Significant differences were seen in the rectal temperature responses of Saidi lambs subjected to various treatments during hot and cool months. Throughout the experimental period (absence, moderate, and extreme-severe heat stress), the rectal temperature of lambs in the LMBOM and RTPOM groups in both the morning and afternoon exhibited a substantial reduction compared to the control group. Previous studies have shown that incorporating medicinal plant mixtures or their essential oils in ruminant diets mitigates the negative impacts of heat stress **[12, 16, and 57]**.

## Conclusion

Taking together, our results demonstrate that essential oil blends (RTPOM or LMBOM) and their active substances underscored the significant synergistic influence on performance of Saidi lambs. In particular, the incorporation of the RTPOM or LMBOM enhanced blood serum biochemical parameters and mitigated heat stress danger.

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# التأثيرات التآزرية لمخاليط الزيوت العطرية والمواد الفعالة لها على مكونات الدم البيوكيميائية في ظل تغير المناخ للحملان الصعيدي النامية

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**الملخص**

هدفت هذه التجربة الي دراسة إلى التحقيق في التأثير التآزري لمزيجين من الزيوت العطرية المستخلصة من النباتات الطبية كإضافة نباتية. تم تحليل المواد الفعالة وتأثيرها على معايير مختلفة لمصل الدم. تتمثل المكمل الأول خليطًا بنسبة 1:1:1 من زيوت الروزماري، الزعتر، والنعناع. بينما المكمل الثاني خليطًا بنسبة 1:1:1 من زيوت حشيشة الليمون، البردقوش، وحبة البركة. تم إعطاء المكملات للمجموعتين من ذكور حملان أغنام الصعيدي النامية بعدد 5 في كل مجموعة باستخدام محقنة عن طريق الفم. تضمنت المجموعة الأولى نظامًا غذائيًا ضابطًا خاليًا من خليط الزيوت العطرية (بنسبة خليط العلف المركز إلى العلف الخشن 70:30). تضمنت المجموعة الثانية نظامًا غذائيًا ضابطًا مدعمًا بـ 0.03% من مخلوط زيوت الروزماري، الزعتر، والنعناع على أساس المادة الجافة، بينما تضمنت المجموعة الثالثة نظامًا غذائيًا ضابطًا مدعمًا بـ 0.03% من مخلوط زيوت الروزماري، الزعتر، والنعناع على أساس المادة الجافة. استمرت التجربة لمدة 180 يومًا. تم تحليل جميع الزيوت العطرية للنباتات الطبية باستخدام تقنية الكروماتوجرافيا الغازية-مطياف الكتلة لتحديد المواد الفعالة. أظهرت النتائج أن المواد الفعالة الرئيسية لمخلوط زيوت حشيشة الليمون، البردقوش، وحبة البركة هي يوكاليبتول وثيمول للروزماري، وثيمول وب-سيمين للزعتر، و(-)-كارفون والليمونين للنعناع. أما بالنسبة لمخلوط زيوت حشيشة الليمون، البردقوش، وحبة البركة، فكانت المواد الفعالة الرئيسية هي السيترال و1,3,8-ب-مينثاترين لحشيشة الليمون، وتربينين-4-أول وجاما-تربينين للبردقوش، والهيبتان، 2,2,4,6,6-بنتاميثيل، ثيموكوينون، والبنزين 1-ميثيل-3-(1-ميثيل إيثيل)- لحبة البركة. أظهرت النتائج ارتفاعًا كبيرًا في إجمالي بروتين المصل (P=0.0069) والجلوبيولين (P=0.0453) وانخفاضًا في مستويات الكوليسترول (P=0.0429) مع تقديم مخلوط (زيوت الروزماري، الزعتر، والنعناع) ومخلوط (زيوت حشيشة الليمون، البردقوش، وحبة البركة). ومع ذلك، لم تتغير مستويات ألبومين المصل، الكرياتينين، اليوريا، حمض اليوريك، AST، ALT، الدهون الثلاثية، والجلوكوز بشكل كبير. كانت جميع قياسات مكونات مصل الدم ضمن المستويات الطبيعية المعروفة. علاوة على ذلك، أدى تضمين خليط الزيوت العطرية إلى انخفاض ملحوظ في درجة حرارة المستقيم (P<0.0001) في بيئات حارة وباردة مقارنة بالأغنام المغذاه على عليقة المقارنة.