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Assessment of the Effect of Thyme Essential Oil on Biochemical Parameters in Endurance Horses

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ABSTRACT

Essential oils as natural plant extracts have received significant attention in recent time. The extracts of thyme play vital roles in the metabolic and regulatory functions of the body. The objective of this study was to evaluate the effect of thyme essential oil on the metabolic functions in endurance horses and to determine the dose for the optimal levels of major biochemical parameters. Thyme essential oils were administered orally to the horses at doses of 150 mg/kg and 600 mg/kg body weight by mixing the oil with their diet. Serum biochemical parameters such as total cholesterol, triglycerides, creatinine, urea, uric acid, total proteins, aspartate aminotransferase, alanine aminotransferase were evaluated according to standard methods. The results showed that the addition of thyme to the horses' diets seems to have a beneficial effect with decrease in key biochemical parameter like total cholesterol (0.68 ± 0.02 g/L), uric acid (0.006 ± 0. 0005 g/L), and alanine aminotransferase (7.90 ± 5.28 g/L), and aspartate aminotransferase (213.00 ± 47.46 g/L) at 150 mg/kg, and decrease in triglycerides (0.27 ± 0.28 g/L), creatinine (0.013 ± 0.004 mg/L), and urea (0.28 ± 0.09 mg/L) at 600 mg/kg. Thyme-based diet could help improve the fitness, performance, and overall wellbeing of race horses through optimization of their serum biochemical profile.

***Keywords*:** Endurance horses, Essential oil, Biochemical parameters, Thyme.

Introduction

Following the European Union (EU) ban on the use of antibiotics in animal feed in 2006,[1] the use of natural plant extracts, in particular essential oils (EOs), has increased significantly. Aromatic plant extracts such as herbs, spices, and volatile oils are frequently used as feed additives due to their anti-hypercholesterolemic and anti-inflammatory activity.[2-5] Their role in supporting metabolic functions, their action on biological functions and regulatory mechanisms in severe and chronic diseases has been proven by several studies.[6-8] Laboratory experiments have shown the antimicrobial activity of essential oils, including thymol.[9,10] Thymol is considered the main component of thyme and oregano essential oil and has also been shown to have an antibacterial effect on several bacterial species.[11, 12] In human and animal health, studies have attempted to demonstrate the efficacy of EOs for the treatment of certain diseases.[12] Several studies have shown the therapeutic effects of *Thymus vulgaris* (thyme) on skin ulcerations due to leishmaniasis in rats, and its essential oil have been proven to be very effective in controlling insects and have also been suggested as a treatment for cutaneous leishmaniasis. [13,14, 15,16] However, Nardelli *et al* (2009) asserted that the use of thyme essential oil should follow aromatherapy practices to avoid possible intoxication as thyme oil can cause allergic reactions, in the form of dermatitis or skin inflammation.[17]

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Although, veterinary studies have proven the beneficial doses of thyme in animal species, such as rats,[18] and poultry,[19] studies to evaluate the effects of thyme essential oil on the biochemical parameters in endurance horses is scanty. Few studies have focused on determining the dose to be recommended to optimize the biological functions of horses, while some have determined the toxic dose of thyme essential oils in horses. One study showed that 880 mg/kg body weight is considered the toxic dose of thyme in horses.[20] Another study has shown that the beneficial effect of thyme on the digestive function of horses starts at 300 mg/kg.[21] To the best of our knowledge, no study has been conducted to determine the optimal dose of the beneficial effect of thyme on serum biochemical parameters in horses.

Therefore, the present study was conducted to evaluate the effect of thyme essential oil on the key biochemical parameters including total cholesterol (TC), triglycerides (TG), creatinine (CRT), urea (UREE), uric acid (UA), total protein (TP), aspartate aminotransferase (AST), and alanine aminotransferase (ALT) in endurance horse and to determine the optimal dose for better performance. The results obtained from this study will be of great importance in the field of aromatherapy in terms of rational use of these natural products and in the field of veterinary medicine to improve the overall well-being of horses.

Materials and Methods

Animals

Adult male horses (Age 12 - 20 years, with an average weight of 300 – 450 kg) used in this study were the property of the Hassan II Agronomic and Veterinary Institute in Rabat, Morocco, and their handling and monitoring were the responsibility of an experienced veterinarian working at the Institute.

Ethical considerations

The study was authorized by the Hassan II Agronomic and Veterinary Institute, Rabat. All ethical rules were respected by following the ethical standards and recommendations of the Moroccan Ministry of

Agriculture in force at the institute, the international ethical standards (European Union Directive 2010/63/EU) and the ARRIVE (Animal Research Reporting of *In Vivo* Experiments) guidelines.

Essential oil of thyme
Thyme-based Essential Oil/Drinkable Substance (EO/DS) product was donated by the Laboratory of Molecular Biology of Bacterial and Fungal Diseases of the Faculty of Sciences of Fez. The certification of the different constituents of the essential oil shows thymol as the major component of thyme and oregano essential oil.[11]

Experimental procedure
Sampling
Five horses were randomly selected from the breeding of male horses of Arabian beard breed aged between 12 and 20 years, with an average weight between 300 kg and 450 kg, free of any apparent pathology and in good condition. The selected horses were divided into two groups. The first group (Group 1) consisting of three horses received a dose of 150 mg/kg live weight (LW) of EO/DS and the serum concentrations of the studied biochemical parameters were measured on day 0 (D0) before administration of the product, on day 2 (D2), on day 6 (D6) and day 10 (D10) after administration of the product. The second group (Group 2) of two horses was dosed at 600 mg/kg body weight (BW) and serum concentrations of the biochemical parameters were measured at day 0 (D0) and day 2 (D2). It is important to state that all the manipulations carried out; the administration of thyme essential oil, and the follow-up of the horses were carried out by an experienced veterinarian of the Hassan II Agronomic and Veterinary Institute of Rabat. The horses were in good health condition throughout the study. Blood samples for biochemical studies were taken by the experienced veterinarian, and were sent to the laboratory for analysis of the different biochemical parameters.

Biochemical analysis
Blood samples were collected just before EO administration at D0 and after EO administration at D2, D6, and D10 depending on the EO doses (150 mg/kg) and (600 mg/kg). The blood samples collected were placed in 10 mL dry glass tubes, centrifuged, and the supernatants (serum) were collected and stored at -20°C until ready for use. Biochemical analyses were done using commercial kits (R.S.A. biosystem). The following parameter were determined: Total cholesterol (TC), Triglycerides (TG), Creatinine (CRT), Urea (UREE), Uric acid (URA), Total protein (TP), Aspartate aminotransferase (ASAT), and Alanine aminotransferase (ALT). The serum concentration of each of these parameters was determined following the manufacturer's protocol (BIOSYSTEMS ® S.A.).

Determination of total protein
The determination of total serum protein was performed according to the Biuret reaction. Proteins present in the sample react with copper (II) ions (Cu^{2+}) in an alkaline medium to give a violet complex that can be quantified spectrophotometrically at 545 nm. Albumin in the sample reacts with bromocresol green in acidic medium to form a coloured complex (blue-green) that can be measured spectrophotometrically.

Urea determination
This was done using the urease method. Urea in the blood is decarboxylated with the help of an enzyme specific to urea in aqueous medium, called urease. The action of the mixture of salicylate and sodium hypochlorite on the ammonium ion formed in the presence of nitroprusside leads to a green coloured indophenol that can be quantified spectrophotometrically at 630 nm.

Determination of creatinine
The creatinine concentration in the sample was measured by the Jaffé reaction method. In this method, creatinine reacts with picrate in alkaline medium to give a coloured complex quantifiable by spectrophotometry at 492 nm.

Determination of liver enzymes
To determine the activity of an enzyme, the reaction catalyzed by the enzyme is used and the amount of product formed or the amount of substrate destroyed in a given time is measured. The enzyme activities of ALT and AST were determined using the coupled reactions of lactate dehydrogenase (LDH) and malate dehydrogenase (MDH), respectively, from the rate of disappearance of NADH, measured at 340 nm using a spectrophotometer.

Determination of cholesterol
Cholesterol concentration was determined by an enzymatic method using cholesterol esterase and cholesterol oxidase. Cholesterol esters are first hydrolyzed to free cholesterol by esterase and then the free cholesterol is oxidized to cholestenone with the production of hydrogen peroxide.

Determination of uric acid
The determination of uric acid is a colorimetric enzymatic assay. Uric acid is converted by uricase into allantoin and hydrogen peroxide. The latter, under the catalytic action of peroxidase, oxidizes 3, 5-dichloro-2-hydroxy benzene sulphonic acid and 4-aminophenol to form a red-violet quinone-imine complex; the sample is read at 546 nm.

Determination of triglycerides
The triglycerides present in the sample give a colored complex after enzymatic hydrolysis and oxidation which can be quantified spectrophotometrically. The indicator, quinone imine, is formed from hydrogen peroxide, 4-amino-antipyrine, and chlorophenol under the action of peroxidase. The reading is taken at 546 nm.

Data processing and statistical analysis
Statistical analysis of the obtained data was performed using the Statistical Package for the Social Sciences (SPSS) version 20 software. The Kolmogorov-Smirnov test was used to study the distribution of the variables. Quantitative variables with a Gaussian distribution were expressed as the mean and standard deviation. Quantitative variables with a non-Gaussian distribution were expressed as median and quartile. Categorical variables were expressed as numbers and percentages.

Results and Discussion

To the best of our knowledge, this is the first study to evaluate the effect of thyme essential oil on the metabolic functions of horses and to determine the optimal dose of thyme for a better performance of endurance horses. To achieve this objective, we studied the effect of thyme essential oil at 150 mg/kg and 600 mg/kg on the biochemical parameters of these animals.
The weights of the endurance horses used in this study and the amount (g) of thyme administered are presented in Table 1. The weight of the horses studied ranges from 420 to 492 kg, and the median weight was 458.40 ± 29.89 kg.
A total of 144 biochemical analyses were performed. The results obtained from the studied parameters, namely; lipid profile (total cholesterol and triglycerides), renal function (urea, creatinine), liver function (aspartate aminotransferase or AST, alanine aminotransferase or ALT), total proteins and uric acid were expressed as means and standard deviations with respect to the doses administered and the time of administration of thyme EO.
Biochemical parameters were measured before thyme EO administration at D0, after thyme administration at D2, D6, and D10 (Group 1; 150 mg/kg) and after thyme administration at D2 (Group 2; 600 mg/kg).

Effect of thyme essential oil on lipid profile in endurance horses
The results of the biochemical analysis with respect to the lipid profile before the administration of thyme EO (D0) to the horses in group 1, revealed a mean value of 0.83 ± 0.13 g/L for total cholesterol, and a mean value of 0.19 ± 0.11 g/L for triglycerides. After administration of thyme EO (150 mg/kg), total cholesterol and triglyceride levels were measured as a function of time on day 2 (D2), day 6 (D6), and day 10 (D10). The results of the time course analysis of the mean total

cholesterol levels in the population of endurance horses in group 1 revealed a slightly elevated cholesterol level at D2 (0.85 ± 0.06 g/L) and a very remarkable decrease at D6 (0.69 ± 0.15 g/L) and D10 (0.69 ± 0.02 g/L) (Table 2). On the other hand, triglycerides levels were found to increase at D2 (0.34 ± 0.11 g/L), D6 (0.52 ± 0.14 g/L) and D10 (0.33 ± 0.01 g/L) post administration of thyme EO at 150 mg/kg (Table 2).

Similarly, the analysis of lipid profile before the administration of thyme EO (D0) to the population of endurance horses in group 2 (600 mg/kg), showed mean levels of total cholesterol of 0.71 ± 0.23 g/L, while post administration at D2 showed slightly elevated total cholesterol levels of 0.81 ± 0.18 g/L. For triglycerides measurement, serum triglycerides was found to be significantly reduced at D2 with a value of 0.27 ± 0.29 mg/L compared to D0 (0.47 ± 0.11 mg/L) (Table 2).

The results showed that administration of thyme EO altered serum lipid profile (total cholesterol and triglycerides). While serum cholesterol level is reduced on days 6 and 10 (D6 and D10) after administration of EO at 150 mg/kg, serum triglycerides level is remarkable decrease at a dose of 600 mg/kg compared to the control (D0). This result suggests that the addition of thyme EO to the diet of endurance horses could protect the vessels from arteriosclerosis due to hyperlipidemia. Studies have shown this beneficial effect of essential oils in reducing total cholesterol and triglycerides in the blood of broilers.[22] In addition, other studies have reported that the administration of certain essential oils-rich diet to growing broilers had significant cholesterol-lowering effect.[19,23]

Effect of thyme essential oil on renal function in endurance horses

The results of the analysis of the biochemical parameters for renal function before the administration of thyme essential oil, that is at day zero (D0) revealed a mean creatinine value of 0.016 ± 0.002 g/L, and mean urea value of 0.24 ± 0.15 g/L. However, after a dose of 150 mg/kg of thyme OE (Group 1), the mean levels of creatinine and urea were measured as a function of time on day 2 (D2), day 6 (D6) and day 10 (D10). The results of the time course analysis of mean creatinine levels in the population of endurance horses in group 1 showed no difference in creatinine levels at D2 (0.015 ± 0.002 g/L), D6 (0.015 ± 0.005 g/L) and D10 (0.012 ± 0.004 g/L). While the measurement of urea level showed an increase in urea with values of 0.40 ± 0.015 g/L, 0.38 ± 0.09 g/L and 0.43 ± 0.22 g/L at D2, D6 and D10, respectively (Table 3).

For the group 2 horses, prior to the administration of thyme EO (D0), the creatinine was measured and found to be 0.016 ± 0.001 g/L, while the administration of thyme EO at the dose of 600 mg/kg to the endurance horse population resulted in a decrease in the level of creatinine (0.013 ± 0.004 g/L) on day 2 (D2). For urea, serum urea was found to be reduced with a value of 0.28 ± 0.091 g/L at D2 compared to the control (D0) with mean urea value of 0.33 ± 0.028 g/L (Table 3).

Although, at a dose of 150 mg/kg, there was no difference in serum creatinine, but a slight increase in urea on day 10 (D10) compared to the control. However, at 600 mg/kg dose of thyme EO, a decrease in these two parameters (creatinine and urea) was observed at D2 compared to the control (D0). The results obtained in the renal function test suggest that the addition of thyme EO to the diet of horses at a dose of 600 mg/kg has beneficial effect by decreasing creatinine levels. Knowing that creatinine is a good marker of renal function, it is routinely performed by veterinarians, particularly in the diagnosis, prognosis, and monitoring of renal failure. In renal failure, the two biochemical parameters (creatinine and urea) increase in parallel in a staggered manner, the increase in urea being earlier.[24] Indeed, the isolated increase in urea is due to a decrease in renal perfusion and is often secondary to hypovolaemia.[24]

Effect of thyme essential oil on liver function in endurance horses

The results of the biochemical analysis with respect to liver function enzymes before the administration of thyme EO (D0) in the group 1 horses revealed a mean AST value of 202.00 ± 42.14 g/L, and ALT value of 13.37 ± 4.36 g/L (Table 4). After the administration of thyme EO at a dose of 150 mg/kg there was an increase in serum AST at D2 (233.70 ± 16.65 g/L), followed by a decrease at D6 (198.00 ± 28.36 g/L) and a slight increase at D10 (213.00 ± 47.47 g/L). For serum ALT, the analysis of the results showed an increase at D2 (83.93 ± 3.43 g/L), and at D6 (23.17 ± 15.05 g/L) followed by a very remarkable decrease at D10 (7.90 ± 5.29 g/L) (Table 4).

Table 1: Body weight of endurance horses included in the study and doses of thyme essential oil (EO) administered

Group	**1**			**2**		**Average**	**SD****
Horse number (H*)	H1	H2	H3	H4	H5		
Weight (kg)	441	492	420	455	484	458.40	29.89
Dose of thyme EO (g)	66	74	63	273	290	153.20	117.34

*H: Horse; ** SD: Standard deviation

Table 2: Effect of thyme essential oil (EO) on lipid profile in endurance horses

Biochemical parameter	**Cholesterol (g/L)**				**Triglyceride (g/L)**			
Time of administration of thyme EO	**Before**	**After**			**Before**	**After**		
Group 1*	**D0**	**D2**	**D6**	**D10**	**D0**	**D2**	**D6**	**D10**
H1*	0.94	0.91	0.77	0.71	0.15	0.28	0.68	0.32
H2	0.68	0.85	0.52	0.67	0.31	0.46	0.41	0.34
H3	0.86	0.8	0.78	0.68	0.11	0.27	0.48	0.32
Average	0.83	0.85	0.69	0.69	0.19	0.34	0.52	0.33
SD	0.13	0.06	0.15	0.02	0.11	0,11	0.14	0.01
Group 2**	**D0**	**D2**			**D0**	**D2**		
H4	0.55	0.68			0.4	0.07		
H5	0.87	0.93			0.55	0.48		
Average	0.71	0.81			0.475	0.275		
SD	0.23	0.18			0.11	0.29		

* Dose of 150 mg/kg of thyme EO, ** Dose of 600 mg/kg of thyme EO.

On the other hand, the analysis of biochemical parameters for liver function before the administration of thyme EO (D0) in group 2 horses showed a mean AST value of 163.00 ± 5.66 g/L, and a mean ALT value of 21.30 ± 10.32 g/L. While the results of the biochemical analysis after administration of thyme EO at 600 mg/kg showed a decrease in the AST value (129.00 ± 9.90 g/L) on the second day (D2), there was an increase in ALT value (25.35 ± 2.76 g/L) (Table 4).

Compared to other essential oils such as sunflower and soybean, studies have shown that horses fed sunflower oil as the main energy source had higher levels of aspartate aminotransferase after two hours of water training.[25] Aspartate aminotransferase (AST) is a liver enzyme that is used as a marker for the early detection of liver damage. The liver has a large functional reserve; 60-70% of its parenchyma must be damaged for its function to be compromised. When clinical signs of liver failure appear (weight loss, jaundice, photosensitization, etc.), the prognosis becomes more difficult. Routine biochemical blood tests often include at least one liver enzyme such as AST, gamma-glutamyl transferase (GGT), alkaline phosphatases (ALP).[26]

Although AST has both a hepatic and a muscular origin,[27] AST and creatinine values are higher in sports horses. The higher values of AST in sport horses could be explained by the higher muscle activity in these horses due to the rigorous training imposed on them,[28] and the higher creatinine concentration is due to their higher muscle mass.[29,30] However, the average live weight of sport horses in this study is 451 kg, a study in Senegal shows that the live weight reaches 460 kg in the same case.[28]

For alanine aminotransferase, there was a remarkable decrease at D10 at 150 mg/kg dose of thyme EO. This observation could suggest that the addition of thyme EO to the diet of endurance horses at a dose that decreased the level of this parameter will have a beneficial effect by preventing the horses from pathologies related to the increase of ALT. Studies have shown that an increase in serum ALT concentration can be a sign of liver disease; the major ones being liver cirrhosis, cholestasis, and hepatocellular failure.[31,32] The results obtained from the liver function tests, which was characterised by an increase in AST concentration with a concomitant decrease in ALT concentration allowed for the assessment of the performance and well-being of the horses.

Effect of thyme essential oil on total protein and uric acid in endurance horses

The results obtained for the total protein test before administration of thyme EO (D0) in the group 1 horses showed a mean value of 68.70 ± 9.44 g/L (Table 5), while after administration of thyme EO at the dose of 150 mg/kg there was a slight increase in total protein level at D2 (77.13 ± 24.74 g/L), followed by a remarkable increase at D6 (93.40 ± 44.74 g/L) and D10 (114.37 ± 55.44 g/L) (Table 5). However, for the group 2 horses, the total protein concentration prior to thyme EO administration was 72.8 ± 8.70 g/L, and no change was observed after administration of thyme EO at 600 mg/kg (D2) (72.9 ± 5.65 g/L) (Table 5).

Table 3: Effect of thyme essential oil (EO) on renal function in endurance horses

Biochemical parameters	**Creatinine (g/L)**				**Urea (g/L)**			
Time of administration of thyme EO	Before	After			Before	After		
Group 1*	**D0**	**D2**	**D6**	**D10**	**D0**	**D2**	**D6**	**D10**
H1*	0.014	0.013	0.014	0.008	0.3	0.39	0.42	0.69
H2	0.017	0.016	0.02	0.016	0.36	0.42	0.28	0.34
H3	0.016	0.015	0.01	0.012	0.07	0.4	0.45	0.26
Average	0.016	0.015	0.015	0.012	0.24	0.4	0.38	0.43
SD	0.002	0.002	0.005	0.004	0.153	0.015	0.091	0.229
Group 2**	**D0**	**D2**			**D0**	**D2**		
H4	0.017	0.011			0.31	0.22		
H5	0.015	0.016			0.35	0.35		
Average	0.016	0.0135			0.33	0.285		
SD	0.001	0.004			0.028	0.092		

* Dose of 150 mg/kg of thyme EO, ** Dose of 600 mg/kg of thyme EO.

Table 4: Effect of thyme essential oil (EO) on liver function in endurance horses

Biochemical parameters	**AST (g/L)**				**ALT (g/L)**			
Time of administration of thyme EO	**Before**	**After**			**Before**	**After**		
Group 1*	**D0**	**D2**	**D6**	**D10**	**D0**	**D2**	**D6**	**D10**
H1	198	239	188	265	13.6	17.8	13.4	1.8
H2	246	247	230	202	17.6	13.6	15.6	10.8
H3	162	215	176	172	8.9	20.4	40.5	11.1
Average	202	233.7	198	213	13.37	83.93	23.17	7.9
SD	42.14	16.65	28.36	47.47	4.36	3.43	15.05	5.29
Group 2**	**D0**	**D2**			**D0**	**D2**		
H1	159	136			14	27.3		
H2	167	122			28.6	23.4		
Average	163	129			21.3	25.35		
SD	5.66	9.90			10.32	2.76		

* Dose of 150 mg/kg of thyme EO, ** Dose of 600 mg/kg of thyme EO

Table 5: Effect of thyme essential oil (EO) on total protein and uric acid in endurance horses

Biochemical parameters	**Total protein (g/L)**				**Uric acid (g/L)**			
Time of administration of thyme EO	**Before**	**After**			**Before**	**After**		
Group 1*	**D0**	**D2**	**D6**	**D10**	**D0**	**D2**	**D6**	**D10**
H1	57.9	53.7	145	178	0.003	0.01	0.01	0.006
H2	75.4	103	65.4	88.6	0.02	0.01	0.008	0.006
H3	72.8	74.7	69.8	76.5	0.008	0.02	0.04	0.007
Average	68.7	77.13	93.4	114.37	0.011	0.013	0.019	0.006
SD	9.44	24.74	44.74	55.44	0.01	0.01	0.02	0
Group 2**	**D0**	**D2**			**D0**	**D2**		
H1	78.9	76.9			0.01	0,02		
H2	66.6	68.9			0.02	0.01		
Average	72.8	72.9			0.015	0.015		
SD	8.70	5.66			0.01	0.01		

* Dose of 150 mg/kg of thyme EO, ** Dose of 600 mg/kg of thyme EO.

For uric acid determination, the results showed a uric acid concentration of 0.011 ± 0.010 g/L before thyme EO administration (D0) to the endurance horses in group 1. Following the administration of thyme EO at a dose of 150 mg/kg, serum uric acid levels were measured on day (D2), day 6 (D6), and day 10 (D10). The results obtained showed a stable uric acid level at D2 (0.013 ± 0.001 g/L), a slight increase at D6 (0.019 ± 0.020 g/L) followed by a remarkable decrease at D10 (0.006 ± 0.000 g/L) (Table 5). On the other hand, measurement of the uric acid level in the group 2 horses before thyme EO administration showed a mean value of 0.015 ± 0.001 g/L, which remained unchanged (0.015 ± 0.007 g/L) at D2 after the administration of thyme EO at the dose of 600 mg/kg (Table 5).

The remarkable decrease in the uric acid concentration at D10 after administration of thyme EO at a dose of 150 mg/kg suggest that the addition of thyme EO to the diet of endurance horses has beneficial effect by decreasing uric acid level, thereby preventing uric acid-induced pathologies in endurance horses.

Furthermore, the in-depth analysis of the effect of thyme EO on these biochemical parameters by comparing the administered doses (150 mg/kg and 600 mg/kg) in endurance horses in order to determine the optimal dose revealed a decrease in the concentration of most of the biochemical parameters including creatinine, cholesterol, uric acid, AST and ALT at the 150 mg/kg dose on day 10 (D10). However, for the 600 mg/kg dose on day 2 (D2), only triglycerides, urea, creatinine, and AST levels were improved with a remarkable decrease in their values.

In summary, the best results for liver function parameters (AST and ALT), cholesterol, and uric acid were obtained after the 150 mg/kg dose. On the other hand, the 600 mg/kg dose of thyme EO was beneficial for renal function parameters (creatinine and urea), and triglycerides as they were elevated at the 150 mg/kg dose. Thus, these results allowed us to determine the optimal dose of thyme EO that could help sports medicine veterinarians to improve the fitness, performance, and well-being of race horses through good aromatherapy practice.

It should be noted that a significant number of studies have been conducted in the field of herbal medicine on the effects of thyme EO on small animals such as chickens, rats, and mice, so this work could be considered a complement to these studies. However, the paucity of studies on the effects of thyme EO in endurance horses necessitated this study as a pilot study that focuses on the application of good aromatherapy practices based on the use of thyme EO, and measurements of biochemical profiles in endurance horses. Nevertheless, the technical aspect of this work with respect to the population of horses studied, their handling, administration of the extract (thyme EO) and follow-up of the horses was the sole responsibility of the veterinarian in charge, and this was a major limitation of the study and this did not allow a thorough clinical evaluation of these horses.

Conclusion

To the best of our knowledge, the present study is the first to evaluate the effect of thyme essential oil on the main metabolic functions of endurance horses. The results obtained showed that most of the biochemical parameters of the horses were significantly improved after 150 mg/kg dose of thyme essential oil compared to the 600 mg/kg dose. This study suggests that the dose of 150 mg/kg is an optimal dose for better improvement of biochemical parameters in endurance horses.

These findings will therefore be of great importance in the field of aromatherapy and animal health, as they could help sports medicine veterinarians to improve the fitness, performance, and well-being of race horses through appropriate clinical management on the basis of improved biochemical profile after thyme EO administration.

Conflict of Interest

The authors declare no conflict of interest.

Authors' Declaration

The authors hereby declare that the work presented in this article is original and that any liability for claims relating to the content of this article will be borne by them.

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