



Improving Growth, Health, and Immunity of Goats Under Thermal Stress Through Nano-emulsified Cardamom Oil Supplementation



Mohamed Medhat¹, Sameh A. Abdelnour^{1*}, Ali S. A. Saleem², Usama M. Abdel Monem¹, Bakry Khalil¹, and Ali Ali El-Raghi²

¹ Department of Animal Production, Faculty of Agriculture, Zagazig University, Zagazig 44519, Egypt.

² Animal Production Department, Faculty of Agriculture, Sohag University, Sohag, Egypt.

³ Department of Animal, Poultry and Fish Production, Faculty of Agriculture, Damietta University, Damietta 34517, Egypt.

Abstract

NANOTECHNOLOGY has emerged as a promising strategy to enhance phytochemicals' solubility, bioavailability, and efficacy, addressing limitations encountered in biological systems and industrial applications. This study investigated the effects of nano-emulsified cardamom oil (NCEO) on growth performance, physiological parameters, haematological and biochemical profiles, immune function, and oxidative stress in growing goats during summer. Thirty-two healthy, 4-month-old goats were randomly assigned to four groups (n=8 per group) and fed diets supplemented with NCEO at 0 (NCEO0), 100 (NCEO100), 200 (NCEO200), and 400 (NCEO400) mg/kg, respectively. Results demonstrated that NCEO supplementation significantly improved growth performance ($P < 0.05$). Compared to NCEO0, NCEO treatments reduced considerably respiration rate and rectal temperature. Furthermore, NCEO at 200 and 400 mg/kg significantly enhanced erythrocyte and leukocyte counts, blood protein levels, kidney and liver function markers, and lipid profiles. Regarding cellular immunity, IgG and IgM levels exhibited a cubic increase in response to NCEO supplementation, with optimal doses estimated at 300 mg NCEO/kg. Conversely, caspase-3 levels showed a quadratic decrease, with an optimal dose of 275 mg NCEO/kg. NCEO administration resulted in a linear decrease in interleukin-2 (IL-2) levels and a cubic decrease in malondialdehyde (MDA) levels. The total antioxidant capacity (TAC) exhibited a significant linear increase with escalating NCEO concentrations ($P < 0.0001$). Furthermore, the activities of superoxide dismutase (SOD), glutathione peroxidase (GPx), and catalase (CAT) were significantly enhanced in the NCEO-treated groups ($P < 0.01$). These findings suggest that dietary supplementation with 200-300 mg/kg NCEO can significantly enhance growth performance, haematological and biochemical profiles, redox status, and immune function in growing goats during summer.

Keywords: Goat kids, Health status, Heat stress, Nano-emulsion essential oil.

Introduction

The Earth temperature is anticipated to rise by 4°C over the next century [1]. This increase in global temperatures is expected to have prejudicial effects on the livestock industry, including decreasing quantity and quality of feedstuff, heightened competition for natural resources, proliferation of livestock diseases, loss of biodiversity, and increased heat stress (HS) [2]. Among these challenges, HS poses the most significant threat to animal populations in recent periods. High temperatures (HS) are negatively impacting animal health, behavior, and physiology, ultimately leading to

detrimental consequences for survival and reproduction [1].

Several prior studies have explored potential mitigation tactics, with nutritional manipulation emerging as a proactive approach [3, 4]. Incorporating spices or herbs and their derivatives into animal diets as dietary feed additives has shown promise as an effective alternative to antibiotics in relieving the unfavourable impacts of HS in the summer months. These products have garnered consumer approval as natural feed supplements [5, 6]

Prior experiments have emphasized the beneficial impacts of incorporating medicinal herbs and spices,

*Corresponding authors: Sameh A. Abdelnour, E-mail:samehtimor86@gamil.com, Tel.: 01003808525

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known as phytogetic additives, into the diets of growing animals on their growth metrics and general health status [7, 8]. Among these feed additives, cardamom stands out as a popular spice extensively utilized for its culinary applications and flavor-enhancing properties worldwide. Cardamom comprises the dried seeds from *Elettaria cardamomum*. Likewise, cardamom fruit is rich in fats, phytosterols, sterols, phenolic acids, and essential oils. Notably, cardamom essential oil (CEO) is abundant in compounds such as α -terpinyl α -pinene, acetate, phellandrene, myrcene, limonene, α -terpineol, terpinolene, linalool, sabinene, and linalyl acetate [9]. In general, numerous published reports have elucidated that the CEO derived from its fruits exhibits antispasmodic, antimicrobial, and anti-inflammatory properties [10, 11]. The antioxidant and anti-inflammatory capabilities of CEO have been substantiated in several research studies [12, 13]. Despite the advantageous outcomes of CEO, challenges such as inferior solubility, permeability, low storage capacity instability and limited bioavailability, have constrained its application in certain pharmaceutical managements.

Nano-emulsions represent a nanotechnological approach aimed at enhancing the properties of essential oils. Recently published papers have validated that transforming phytochemicals into nanostructures can impart them with various biological purposes, making them more adaptable for industrial applications and facilitating their movements within biological systems, as approved by [14]. According to the antioxidant, anti-inflammatory, and antimicrobial attributes of CEO, we hypothesized that the nano-formulation of cardamom essential oil (NCEO) might have the potential to attenuate the undesirable properties of HS in heat stressed-growing kids. As a result, this study is proposed to explore the effect of incorporating NCEO into the diets on growth parameters, physiological reactions, hematological and biochemical parameters, redox balance, and immune responses of Zaraibi growing kids subjected to elevated ambient temperatures.

Material and Methods

Preparation of Nanoemulsion Cardamom Essential Oil (NCEO)

The cardamom essential oil (CEO) was obtained from the Elhawag company, Nasr city, Egypt, for preparing nano-emulsion cardamom essential oil (NCEO). A single layer of NCEO oil-in-water nano-emulsion was performed [15]. In brief, the nanoemulsion was prepared by mixing 2.5 mL of CEO with a surfactant (Tween 80) mixture and slowly adding up to 10 mL of water while stirring with a magnetic stirrer at 25°C. The ratio of water addition was maintained at approximately 1.0 mL/min. The emulsion was then sonicated for 30

minutes in an ultrasonic bath (Sonix, USA) and further homogenized using an ultrasonic probe (Model CV 334) attached to a homogenizer (Sonics Vibra-cell™, Model VC 505, Inc., USA) under the following requirements: amplitude: 60%, timer: 5 minutes, and pulser: 1 second ON/1 second OFF to produce nanoemulsions.

Physicochemical features of NCEO

The morphology aspects of freshly synthesized NCEO were envisioned via a TEM (transmission electron microscope; JEOL JEM-2100, Tokyo, Japan) at 160 kV. Digital Micrograph and Soft Imaging Viewer software were employed to optimize the image capture and assessment manner. The surface charge of the nano-emulsion particles (Z-potential) was evaluated applying a Zetasizer Nano ZS analyzer

Experimental location, Animals, Management Conditions, and Treatments

This experiment was conducted at the private farm in Awlad Saqr (E, 31° 37' 32.700, N 36.096, 47), Sharkia Province, Egypt. A total of 32 growing Damascus male goats (age, weight) were used in this experiment. The animals were purchased from the Serw Experimental Research, Damietta, Egypt. Animals were housed in identical environmental circumstances. The goats were housed individually in a pen during the experimental period. The experiment has lasted four months during the Egyptian summer conditions. After one week of acclimation, the animals were divided into four groups. Each group consisted of 8 growing goats. The first group (n=8) was a fed basal diet as severed as a control diet without supplementation (NCEO0 group). The other 2nd, 3rd, 4th treated groups, where the growing goats were fed diets enriched with NCEO at various doses 100 (NCEO100), 200 (NCEO200) and 400 (NCEO400) mg/kg diet, respectively. Freshwater was provided ad libitum. The animals were fed a total mixed ration (TMR) twice daily at 08:00 and 15:00. The feed additives were incorporated into the diets during the weekly preparation of TMR. The diet was prepared to meet the growth of animals during the experimental period according to [16].

Using a farm thermometer (Sagar Poultries, India), the humidity data and ambient temperature were documented every day (at 14.00) throughout the experiment. The THI (temperature humidity index) values were estimated corresponding to [17, 18] formula: $THI = (1.8 \times T_{db} + 32) - [(0.55 - 0.0055 \times RH) \times (1.8 \times T_{db} - 26.8)]$, where RH is the relative humidity (%) and represents the dry bulb temperature (°C).

Respiration rate and rectal temperature

Respiration rates and rectal temperatures were documented at 14:00. The rectal temperature was determined using a digital clinical thermometer (Model No.: YF-160A, TYPE-K). The thermometer was sanitized between each goat by applying liquid alcohol (70%) with a cotton scrub. The respiration rate was determined by recording the exhalations and inhalations for 60 seconds.

Growth parameters assessment

Growing goats were weighted every two weeks from the start of the experimental period. The initial (IBW), and final (FBW) body weights were calculated using an electronic scale (Tru-Test AG500 Digital Indicator, New Zealand; precision ± 5 g) to determine the change in body weight. Feed consumption (FI) was recorded to calculate the daily and monthly FI values. The body weight gain (BWG) = $\text{FBW (g)} - \text{IBW (g)}$. FCR (feed conversion ratio) = $\text{total feed intake (g)} / \text{BWG (g)} \times 100$.

Blood sampling

The blood samples ($n=6$, each group) were gathered from the jugular vein on the last day of experimental period. The samples were separated into two subsamples. The first one was transferred into EDTA tube for assessing the blood haematology. The second tube was used for separation the plasma. Plasma was separated by centrifugation the blood for 15 min at $1500 \times g$, and deposited at -20°C .

Haematology and biochemical assessments

The haematological variables were assessed using haematology analyser (HB 7021). These variables were RBCs (red blood cells), HCT (haematocrit), HGB, (haemoglobin), MCV (mean corpuscular volume), WBCs (white blood cells), MCH (mean corpuscular haemoglobin), MCHC (mean corpuscular haemoglobin concentration, PLAT (platelets), SEG (segments), LEU (lymphocytes) and MON (monocytes). The plasma biochemical assays including total protein, albumin, uric acid, creatinine was assessed using commercial kits. Total cholesterol, gamma-glutamyl transferase (GGT), total glycerides, low density lipoprotein (LDL), high density lipoprotein (HDL) were analysed were evaluated spectrophotometrically conferring to the producer's procedure using commercial kits purchased from Biodiagnostic Company (Giza, Egypt).

Immunity and apoptosis markers

The concentration of immunoglobulins G (IgG) and M (IgM) were measured using ELISA kits in accordance with manufacturer instructions. The IgG (Catalog # F210, Cygnus Technologies, USA) and

IgM (ab97226, were determined according to the method in detail by [19], and [20], respectively. The commercial specific kit (E-EL-R0160) for detecting caspase-3 plasma was acquired from Elabscience Company (Texas, 77079, USA) following the method by [21]. Interleukin 2 (IL-2) in goat plasma were determined via commercially available sandwich ELISA kits corresponding to the approach by [22].

Antioxidant/oxidative biomarkers

For redox status calculations, the actions of the enzymes of catalase (CAT), glutathione peroxidase (GPx; MBS9718983) and superoxide dismutase (SOD; MBS8807589), the total antioxidant capacity (TAC; MBS8807700) and malondialdehyde (MDA; MBS8806802), and C-reactive protein (CRP) levels in plasma were measured using specific quantitative sandwich ELISA kits conferring to the constructor's measures using commercial kits (My BioSource, San Diego, CA, USA). All laboratory biochemical and analyses measurements followed the ISO/IEC 17025 procedures (the last version in 2005).

Statistical Analysis

The Shapiro-Wilk and Levene tests were used to confirm homogeneity and normality of variance. The MIXED model procedure in the Statistical Analysis System (PROC MIXED; Institute, 2012) was employed to analyze various growth indices, blood health, and oxidative stress parameters. Duncan's multiple range test was used as a post-hoc analysis to examine specific group differences following the overall ANOVA result [23]. To determine a dose-response curve, the response of each dependent variable to different levels of NCEO (0, 100, 200, and 400 NCEO /kg diet) was analyzed using orthogonal contrast statements for linear and quadratic trends. Significant relationships were further analyzed by fitting dose-response curves using regression equations to identify the optimal dietary NCEO dose. A p-value of < 0.05 was considered statistically significant.

Results

Meteorological parameters

The consequences of meteorological variables observed during the months of investigational time are stated in Table 1. The average values of AT, RH, and THI were $33.47 \pm 0.39^\circ\text{C}$, $46.76 \pm 1.32\%$, and 82.44 ± 0.52 , respectively. The current values of THI show that the growing kids were subjected to severe heat stress conditions.

Growth indicators, feed efficiency, and physiological reactions

The effect of dietary administration of NCEO on the growth indicators and feed proficiency of growing kids are performed in Table 2. The values of BWG were notably greater in the NCEO-treated

groups than the basal group ($p < 0.05$), quadratic regression analysis implied that the best dosage of NCEO supplementation was at 250 mg / kg diet (Fig. 1A). However, the dietary treatment did not have a considerable effect on feed intake ($p > 0.05$). On the other hand, the dietary addition of NCEO exhibited significantly improved feed utilization, indicated by inferior FCR, compared to the HS group ($p < 0.05$), optimizing at 250 mg NCEO/ kg diet (Fig. 1B). The respiration rate and rectal temperature notably declined in all NCEO-treated groups in opposite to the HS group ($p < 0.05$; Fig. 2A and B).

Haematological attributes

The consequences of dietary treatment on haematological variables are presented in Table 3. All Erythrocyte and leucocyte count linearly improved as response to the dietary treatment except HGB, HCT and WBCs showed quadratic increase. Meanwhile, cubic relationship was observed between PLT and the dietary levels of NCEO. The optimum doses were established at 275 mg/ kg diet for WBCs (Fig. 3D) and at 300 mg/kg diet for HGB, PLT and HCT (Fig. 3A, B, and C).

Blood profile

As illustrated in Table 4, all of blood parameters were statistically ($p < 0.0001$) affected by the NCEO treatment. The values of total protein and albumin linearly boosted as response to the dietary treatment ($p < 0.0001$ and 0.0058). NCEO treatments significantly reduced the concentrations of TC, TG, and LDL and significantly increased the levels of HDL. The polynomial regression evaluation implied that the optimal concentrations were at 200 mg/kg diet for TC (Fig. 4A), 275 mg for LDL (Fig. 4C), and 250 mg NCEO/kg diet for TG and HDL (Fig. 4B and 4D). With regard to liver function, both GGT and CRP quadratically decreased due to the treatment, the optimal doses were established at 300 and 200 mg NCE/ kg diet, respectively (Fig. 5 A and 5B). For kidney function, the concentrations of uric acid and creatinine substantially decreased by dietary treatment, minimizing in the NCEO200 and NCEO400 treated groups.

Immunity response and redox balance

The effects of various levels of NCEO on cellular immune response and antioxidant capacity in growing kids are explained in Table 5. With regard to immunity status, the dietary treatment significantly affected IgG, IgG, caspase-3 and IL-2. Both of IgG and IgM cubically increased as response to the NCEO treatment, however caspase-3 quadratically decreased, the dose response curves displayed that the optimal doses were 275 mg/ kg diet for caspase-3 (Fig. 6 C) and 300 mg/kg diet for IgG and IgM (Fig. 6A and B). A linear relationship was monitored between the increased doses of NCEO and the concentrations of IL-2.

Regarding redox status, increasing the levels of NCEO caused a quadratic decrease in the levels of MDA ($p = 0.0003$). Corresponding dose–response curves showed that the optimal dose was at 225 mg NCEO /kg diet (Fig. 7D). However, the levels of TAC were in a linear increase ($p < 0.0001$). Regarding the antioxidant enzymes, the dietary treatment cubically affected both of SOD and GPX activities ($p < 0.0001$ and 0.0391 , respectively), and quadratically affected catalase activity, the optimal NCEO dose was at 300 mg NCEO/ kg diet for SOD and GPX (Fig. 7-A and C) and at 275 mg NCEO/ kg diet for CAT (Fig. 7B).

Discussion

Heat stress (HS) is a major environmental issue affecting the health, welfare and physiology of animal organisms, including livestock species. Goats are a significant source of various animal products in the livestock industry, making them important for improving productivity during environmental HS challenges. Dietary interventions could help the goats to improve their resilience to HS, thereby enhancing growth, productivity, and overall health.

In this study, we observed that adding NCEO significantly enhanced growth and feed efficiency, cellular immunity, and antioxidant markers. It also reduced oxidative stress, pro-inflammatory cytokines, and the thermoregulatory response (RR and RT) in growing kids during summer conditions. The THI can be a useful tool for evaluating the expose of HS on goats in hot situations [17, 18]. Despite the THI values indicating severe heat stress conditions for the growing goats in this study [24], the goats supplemented with NCEO showed better heat tolerance, improved growth performance, blood health, and higher immune ability.

Fortifying growing goats' diets with NCEO led to a substantial reduction in body temperature, suggesting that the bioactive molecules in NCEO, such as phenolic compounds and flavonoids molecules, have anti-HS modulatory impacts. To accomplish thermoregulation, there should be a homeostasis state between heat loss and heat gain in the animal body. Hence, under acute HS conditions, goats heighten their respiratory rate to dissipate heat through evaporation, explaining the higher breathing frequency in the HS group compared to the NCEO groups. The thermoregulatory effects of some phytochemicals have been verified in farm mammals by [14], owing to their strong antioxidant pathways. Feeding broiler diets enriched with CEO (100 or 50 mg/kg diet) significantly improved body weight and FCR [11]. Dietary inclusion of cardamom powder improved egg production and body weight in broilers [25] and quails [26]. Moreover, stressed rabbits that received NCEO exhibited significant increase in their body weight and feed efficiency, as evidenced by [15]. Studies have revealed that HS can reduce body

weight in animals, while broilers fed diets with 1,8-Cineole significantly enhanced the body weights of broilers [27]. This is due to the anti-inflammatory activity of 1,8-Cineole and its ability to modulate the intestinal microbiota. The microbial activity of CEO may be attributed to its higher content of 1,8-cineole. Dietary 1,8-cineole microcapsules restored the regular assembly of the upper ileum and changed the composition of intestine microbiota under HS [27]. They boosted the levels of *Escherichia* and *Lactobacillus*, while reducing the proportion of *Salmonella* in the ileum. *In vitro*, 1,8-cineole efficiently hampered the growing of *Salmonella*, as proven by inhibition zone tests. A study by [28] found that phytochemical supplementation in goats' diets significantly improved body weight during heat stress conditions. The main flavor components of CEO incorporate α -pinene, β -pinene, 1,8-Cineole, α -terpineol, allo-aromadendrene, limonene, α -terpinyl acetate, and 1,8-cineole promoting substantially to its flavor profile. CEO has demonstrated anti-inflammatory, antifungal, and antimicrobial properties. Research by [29], also identified α -Terpinyl acetate as the main constituent of CEO (55.99%), followed by 1,8-cineole (8.82%), Z-caryophyllene (3.82%), linalool (6.99%), E-nerolidol (3.07%), eugenol (2.31%), dihydrocarveol (6.06%), geraniol (4.46), and terpinen-4-ol (1.83%). Studies found that the NCEO has potent antimicrobial action against *E. coli* due to its small size (<150 nm) and great entrapment competence (>90%) [29, 30].

Haematological blood parameters are essential for assessing the health of animals. It is well accepted that HS can disrupt blood haematology by decreasing erythrocyte counts and increasing leukocytes in goats. This could lead to anaemia and disrupt the function of red blood cells, triggering anaemia [24]. Therefore, enriching diets to support blood health may be a beneficial strategy for maintaining healthy animals and improving overall biological function in the body. In this study, CEO can improve erythrocytes and decrease leukocytes. The outcomes of this investigation are consistent with prior research by [28] in goats and [15] in rabbits. Additionally, [31] demonstrated that melatonin supplementation enhanced the blood health of goats exposed to HS conditions. In contrast, [11] found that dietary fortifications with CEO had no impact on the blood haematology of broiler chickens.

HS can impair lipid metabolism and absorption by damaging the intestinal barrier or reducing the enzymes that transport lipids, leading to lipid accumulation [18]. The addition of NCEO to stressed goats' diets improved lipid metabolism by reducing the lipid profile in the blood. The authors suggested that this improvement may be attributed to CEO inhibiting enzymes involved in lipogenesis, such as fatty acid synthase and acetyl-CoA carboxylase,

based on *in vitro* trials [32]. The anti-hyperlipidemic activity of NCEO was reported by [33]. The lipid profile markers, including VLDL, TG, HDL, LDL, and total cholesterol, were significantly reduced with NCEO administration in rats [33] as clarified in our current research. Similarly, CEO significantly reduced the levels of LDL and cholesterol in broilers [11]. A study by [29] found that CEO decreased lipid levels in obese rats, indicating its ability to prevent dyslipidemia, oxidative stress, and liver impairment in rats fed a high-fat diet. Cardamom powder supplementation significantly decreased the elevated liver enzymes induced by high-fat diets in rats [29]. Kidney function plays a critical role during heat stress conditions. Further, HS can increase renal biomarker damage in the blood of goats. As shown in this study, HS can accumulate creatinine and uric acid, which reflects the slight loss of remove waste products induced by HS in goats. This phenomenon was also shown in rabbits by several authors [34, 35]. Dietary NCEO enhanced the kidney function during HS in rabbits [15]. Studies have revealed CEO has potency as a nephroprotective agent against many toxic elements [13, 36].

This study also demonstrated that HS leads to oxidative stress and an imbalance in antioxidants in rabbits. This finding is consistent with previous research that showed HS reduces the defensive system by increasing oxidative stress and decreasing antioxidative stress in goats [17, 19]. Various tools have been anticipated to give explanations of the development of oxidative stress and organ dysfunction during HS conditions in animals. Oxidative stress in adipose tissue is an early event in the negative effects of heat stress on animals. HS disrupts the defensive system by causing an imbalance between antioxidants and oxidative stress [6, 24, 35]. In this study, the use of CEO significantly improved the defensive system in growing goat kids by increasing levels of TAC, SOD, CAT, and GPx, while reducing MDA levels as a marker of lipid peroxidation. Feeding goats with CEO enhanced cellular antioxidant capacities and prevented the increase of oxidative stress induced by heat stress. These findings are consistent with prior experiments that have shown cardamom enhances antioxidant capacity and reduces MDA in goats. Cardamom rhizome ethanolic extract substantially raised SOD concentrations and diminished MDA, CRP, and IL-6 [37].

It is prominent to mention that there was an overproduction of OS under high-temperature conditions. The antioxidant protection approach, including enzymes like SOD and GSH, helps balance ROS production. Previous studies have shown that high ambient temperatures can increase oxidative stress by promoting lipid and protein oxidation and interrupting antioxidant enzyme activity [6, 35, 38]. In this study, dietary supplementation with NCEO

improved anti-oxidative stress markers. These results are consistent with prior experiments implying that essential oils can mitigate the harmful consequences of HS by hindering oxidation consequences and suspending lipid and protein oxidations [38, 39].

HS can compromise the immune function of newly weaned rabbits by affecting the synthesis of immunological factors like IgM, and IgG, making them more susceptible to pathogens. This immune imbalance can hinder the growth of growing goats. In this study, NCEO supplementation enhanced the synthesis of immunological factors, containing IgG and IgM, counteracting the negative effects of heat stress on newly weaned rabbits through warmth conditions. The beneficial effects of NCEO may be ascribed to its antimicrobial, antioxidant, and anti-inflammatory actions [18], which can reduce bacterial colonization in the gut and promote a balanced gut microbiota, leading to improved feed efficiency and metabolism.

In this study, stressed goats showed a massive employment of inflammatory cells in the hepatic tissues, followed by an increase in extracellular matrix deposition, supporting the notion of hepatic harm [40]. CEO administration avoided the employment of extracellular matrix deposition and inflammatory cells in stressed goats. The defensive result is intervened by the rebuilding of antioxidant function and the lipid-lowering effect of cardamom administration [40]. It has been observed that CEO can reduce proinflammatory cytokines in mice after infection [41]. Moreover, CEO lowered the levels of monocytes, macrophages, and T cells in the colon of rats after infection. These conclusions are consistent with prior in vitro reports viewing the anti-inflammatory results of CEO [41]. For example, cardamom treatment suppressed the production of Th1 cytokines like IFN- γ in murine splenocytes and reduced TNF- α and nitric oxide levels in mouse peritoneal macrophages. Moreover, oral administration of cardamom provided protection against lung damage in Swiss mice [42]. In our

study, treatment with NCEO reduced pro-inflammatory immune responses such as IL-2 and caspase-3 in stressed goats.

The liver is a vital organ for metabolism in the body. HS can induce damage to internal organs such as the rumen and liver, disrupting the absorption and metabolism of nutrients [43]. The present study found that adding NCEO to the diets of growing goats helped preserve the organ structures of the animals, including the liver and rumen. The results were consistent with previous results of [44], who found that herb oils can protect the organ damages induced by HS in sheep. This topic requires further clarification to support our results, especially at the molecular and transcriptional levels.

Conclusion

Taken together, incorporating phytogetic antioxidants derived from the nano-emulsion of NCEO into the diets of heat-stressed growing animals can effectively mitigate the adverse effects of heat stress on growth performance, feed efficiency, cell-mediated immunity, and antioxidant status. Dose-response evaluation revealed that a supplementation level of 200-300 mg NCEO/kg of diet is recommended for growing animals reared under hot climatic conditions.

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Declaration of Conflict of Interest

The authors declare that there is no conflict of interest.

Ethical of approval: All experimental procedures and animal handling were reviewed and approved under approval NO. ZU-IACUC/2/F/367/2022 by Animal Use in Research Committee (IACUC), Zagazig University, Egypt.

TABLE 1. The mean values of relative humidity (RH), ambient temperature (AT), and THI throughout the investigational time

Parameters	Months				
	May	June	July	August	Overall
RH (%)	43.75 $\pm$ 2.39	42.50 $\pm$ 1.44	47.50 $\pm$ 1.44	52.00 $\pm$ 2.23	46.76 $\pm$ 1.32
AT ($^{\circ}$ C)	33.50 $\pm$ 0.50	33.25 $\pm$ 1.10	33.50 $\pm$ 0.96	33.60 $\pm$ 0.83	33.47 $\pm$ 0.39
THI	81.93 $\pm$ 0.71	81.37 $\pm$ 1.22	82.66 $\pm$ 1.47	83.54 $\pm$ 0.76	82.44 $\pm$ 0.52

THI: temperature–humidity index; AT: ambient temperature; RH: relative humidity.

TABLE 2. Feed utilization Growth and performance of growing kids fed dietary supplemented with nano-emulsion of cardamom essential oil

Items	Treatments (TRTs)				SEM	p-values			
	NCEO0	NCEO100	NCEO200	NCEO400		TRTs.	Lin.	Quad.	Cub.
IBW, kg	14.29	14.39	14.38	14.14	0.14	0.558	0.937	0.983	0.177
FBW, kg	23.59 ^b	24.04 ^a	24.13 ^a	23.93 ^a	0.15	0.021	0.059	0.043	0.082
BWG, kg	9.30 ^b	9.65 ^a	9.75 ^a	9.79 ^a	0.11	0.007	0.021	0.010	0.463
TFI, kg	52.91	53.33	50.66	53.06	0.63	0.465	0.888	0.136	0.969
Monthly FI, kg	13.00	13.01	13.21	13.26	0.15	0.463	0.899	0.135	0.975
Daily FI, kg	0.43	0.43	0.44	0.44	0.01	0.454	0.633	0.155	0.887
FCR	5.69 ^a	5.47 ^{ab}	5.25 ^b	5.42 ^{ab}	0.07	0.034	0.190	0.046	0.073

Lin, linear regression; quad, quadratic; cub, cubic regression. ^{a,b} means within a row without a common letters differ at $p < 0.05$. IBW: initial body weight; FBW: final body weight; BWG: body weight gain; TFI: total feed intake; FCR: feed conversion ratio.

TABLE 3. Haematological indices of growing kids fed dietary supplemented with nano-emulsion of cardamom essential oil

Items	Treatments (TRTs)				SEM	p-values			
	NCEO0	NCEO100	NCEO200	NCEO400		TRTs.	Lin.	Quad.	Cub.
RBCs ($\times 10^6/\text{mm}^3$)	3.80 ^b	4.36 ^b	5.78 ^a	6.57 ^a	0.32	0.001	0.0001	0.723	0.331
HGB (g/dL)	10.30 ^c	13.22 ^b	15.66 ^a	16.38 ^a	0.26	<.001	<.0001	0.005	0.242
HCT (%)	34.28 ^b	37.60 ^a	39.86 ^a	39.84 ^a	0.68	0.001	0.0003	0.042	0.686
MCV (μm^3)	73.69 ^b	81.48 ^a	82.91 ^a	85.43 ^a	1.48	0.002	0.0006	0.114	0.295
MCH (pg)	30.21 ^b	31.29 ^b	32.22 ^b	35.35 ^a	0.62	0.002	0.0004	0.140	0.428
MCHC (g/dL)	33.85 ^b	35.14 ^b	39.24 ^a	41.05 ^a	1.07	0.004	0.0007	0.816	0.321
PLAT ($\times 10^3/\text{mm}^3$)	20.33 ^c	30.33 ^b	49.67 ^a	48.67 ^a	0.97	<.0001	<.0001	0.001	0.0004
WBCs ($\times 10^3/\text{mm}^3$)	14.07 ^a	11.31 ^b	9.24 ^c	9.73 ^c	0.35	0.0001	<.0001	0.005	0.718

RBCs, red blood corpuscles; HGB, hemoglobin; HCT, hematocrit; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin; PLT, platelet count; MCHC, mean corpuscular hemoglobin concentration; WBCs, white blood cells count. Lin, linear regression; quad, quadratic; cub, cubic regression. ^{a,b} means within a row without a common letters differ at $p < 0.05$.

TABLE 4. Blood profile of growing kids fed dietary supplemented with nanoemulsion of cardamom essential oil

Items	Treatments (TRTs)				SEM	p-values			
	NCEO0	NCEO100	NCEO200	NCEO400		TRTs.	Lin.	Quad.	Cub.
TP (g/dL)	3.17 ^b	3.37 ^b	4.07 ^a	4.17 ^a	0.07	<.0001	<.0001	0.545	0.086
Alb (g/dL)	1.53 ^c	1.73 ^{bc}	2.06 ^a	1.94 ^{ab}	0.09	0.017	0.005	0.130	0.199
UA (mg/dL)	6.44 ^a	5.75 ^b	2.78 ^c	2.65 ^c	0.10	<.0001	<.0001	0.085	0.074
Creat (mg/dL)	1.16 ^a	0.99 ^b	0.71 ^c	0.70 ^c	0.01	<.0001	<.0001	0.240	0.084
GGT (U/L)	3.56 ^a	1.86 ^b	1.13 ^c	1.18 ^c	0.06	<.0001	<.0001	<.0001	0.448
CRP (ng/mL)	44.89 ^a	29.62 ^c	25.01 ^c	37.41 ^b	1.89	<.0001	<.0001	<.0001	0.705
TBil (mg/dL)	0.39 ^a	0.27 ^b	0.17 ^c	0.15 ^c	0.02	0.0001	<.0001	0.644	0.458
TC (mg/dL)	79.93 ^a	61.86 ^b	55.28 ^c	76.53 ^a	1.65	<.0001	0.853	<.0001	0.144
TG (mg/dL)	72.89 ^a	60.81 ^b	49.79 ^c	60.88 ^b	0.77	<.0001	<.0001	0.0002	0.086
LDL (mg/dL)	77.82 ^a	61.27 ^b	41.73 ^d	49.16 ^c	1.02	<.0001	<.0001	0.0065	0.690
HDL (mg/dL)	25.34 ^b	46.92 ^a	46.47 ^a	43.80 ^a	1.47	<.0001	<.0001	0.0001	0.051

TP, total protein; Alb, albumin; UA, uric acid; Creat, creatinine; GGT, gamma-glutamyl transferase; CRP, c-reactive protein; TBil, total bilirubin; LDL, low-density lipoprotein; TG, triglycerides; HDL, high-density lipoprotein; TC, total cholesterol. Lin, linear regression; quad, quadratic; cub, cubic regression. ^{a,b} means within a row without a common letters differ at $p < 0.05$.

TABLE 5. Immunity response and redox status of growing kids fed dietary supplemented with nanoemulsion of cardamom essential oil

Items	Treatments (TRTs)				SEM	p-values			
	NCEO0	NCEO100	NCEO200	NCEO400		TRTs.	Lin.	Quad.	Cub.
Immunity status									
IgG (ng/mL)	150.97 ^c	287.80 ^b	547.33 ^a	560.33 ^a	20.94	<.0001	<.0001	0.018	0.004
IgM (ng/mL)	218.85 ^d	353.07 ^c	519.00 ^a	498.67 ^b	5.02	<.0001	<.0001	<.0001	<.0001
CASP3 (ng/mL)	8.11 ^a	5.24 ^b	4.920 ^b	5.19 ^b	0.35	<.0001	<.0001	<.0001	0.124
IL-2 (pg/mL)	808.65 ^a	552.67 ^b	330.00 ^{cd}	302.00 ^d	37.26	<.0001	<.0001	0.2716	0.9012
Redox status									
TAC (ng/mL)	2.40 ^d	3.61 ^c	5.53 ^b	8.20 ^a	0.29	<.0001	<.0001	0.451	0.487
SOD (U/mL)	23.06 ^c	25.58 ^a	32.04 ^b	30.31 ^b	1.34	<.0001	0.0002	<.0001	<.0001
CAT (ng/mL)	9.13 ^d	10.10 ^a	14.72 ^c	12.50 ^b	0.32	<.0001	<.0001	0.0014	0.105
GPX (U/mL)	3.08 ^b	3.40 ^b	5.96 ^a	4.90 ^a	0.48	<.0001	<.0001	0.0002	0.039
MDA (nmol/mL)	12.11 ^a	8.23 ^b	6.34 ^c	10.01 ^{ab}	0.45	<.0001	<.0001	0.0003	0.130

IgG, immunoglobulin G; IgM, immunoglobulin M; IL-2, Interlukin-2; TAC, total antioxidant capacity; SOD, super oxide dismutase; CAT, catalase, GPX, glutathione peroxidase, MDA, malondialdehyde. Lin, linear regression; quad, quadratic; cub, cubic regression. ^{a,b,c,d} means within a row without common letters differ at $p < 0.05$.

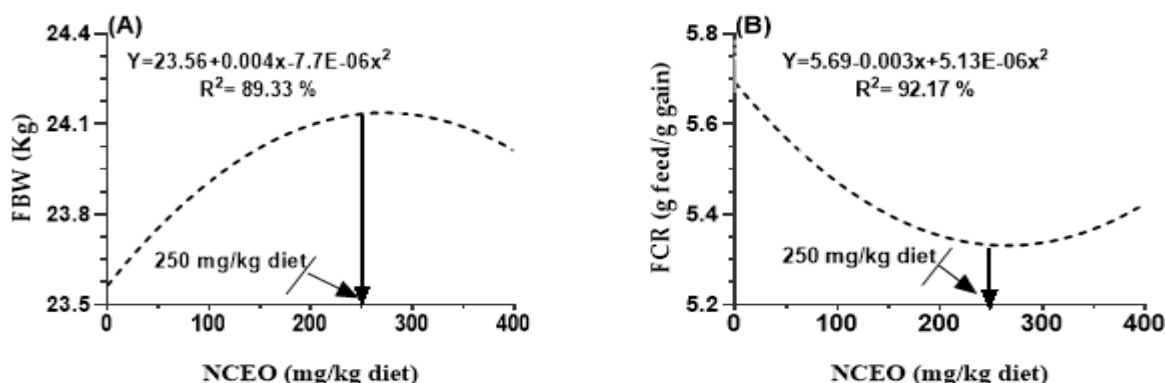


Fig. 1. A polynomial regression analysis between dietary levels of NCEO and final body weight (A – FBW), and feed conversion ratio (B – FCR)

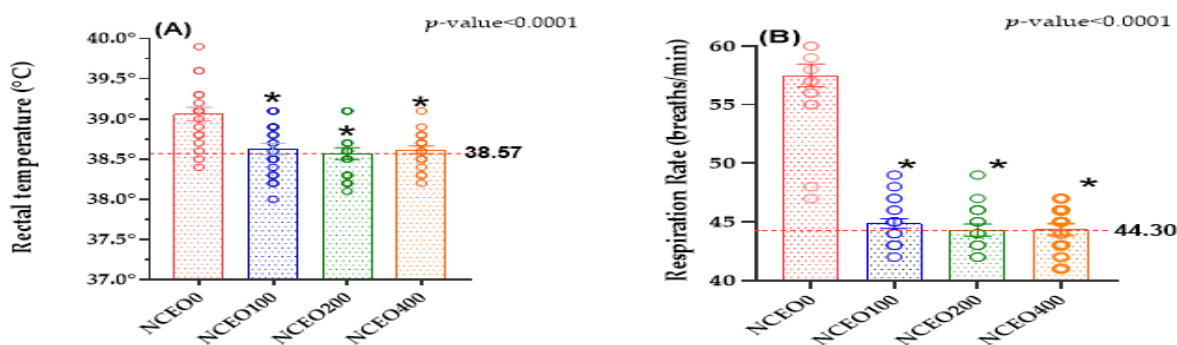


Fig. 2. The values of rectal temperature (A) and respiration rate (B) of heat-stressed kids fed dietary fortified with 100 (NCEO100), 200(NCEO200), and 400 (NCEO400), mg/kg of nano-emulsion of cardamom essential oil related to the basal diet. * $p < 0.05$: differ significantly with control.

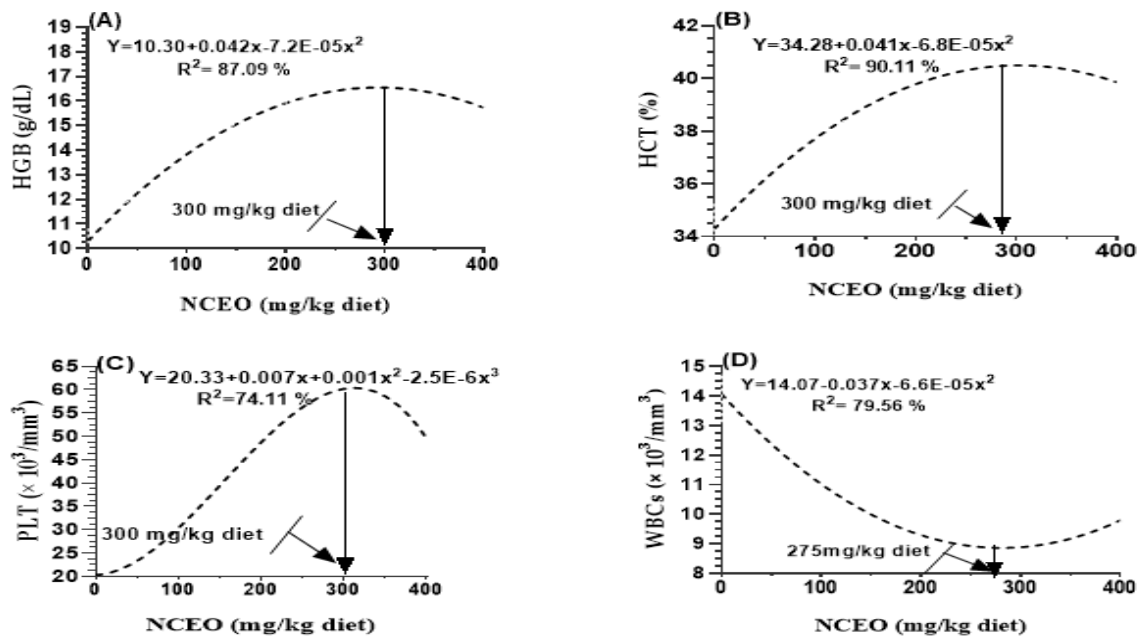


Fig. 3. A polynomial regression analysis between dietary levels of NCEO and haemoglobin concentration (A – HGB), haematocrit (B-HCT), platelet count (C-PLT) and white blood cells count (D – WBCs).

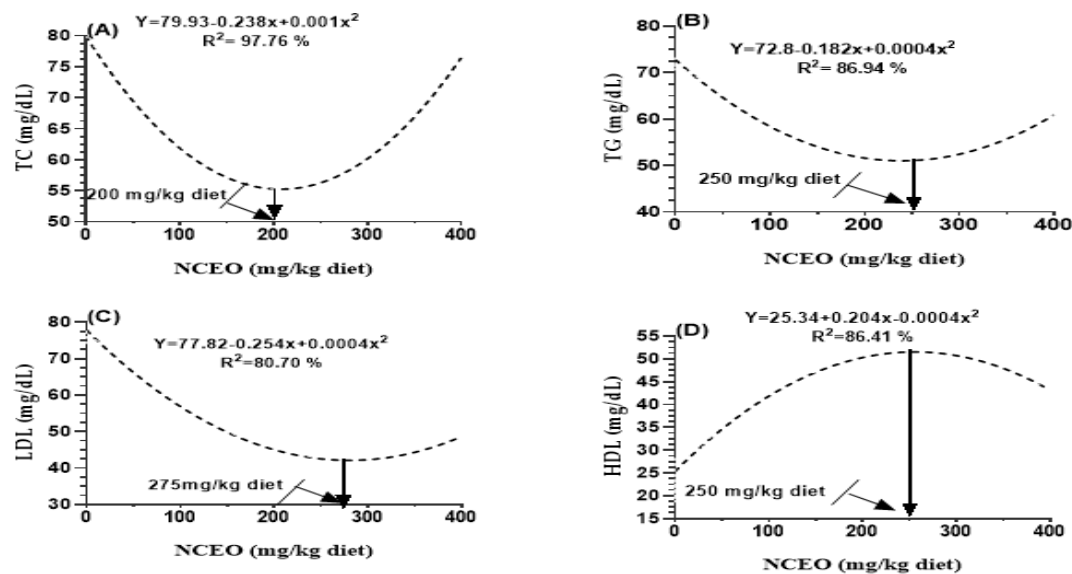


Fig. 4. A polynomial regression analysis between dietary levels of NCEO and total cholesterol (A – TC), triglycerides (B-TG), low density lipoprotein (C-LDL) and high-density lipoprotein (D – HDL).

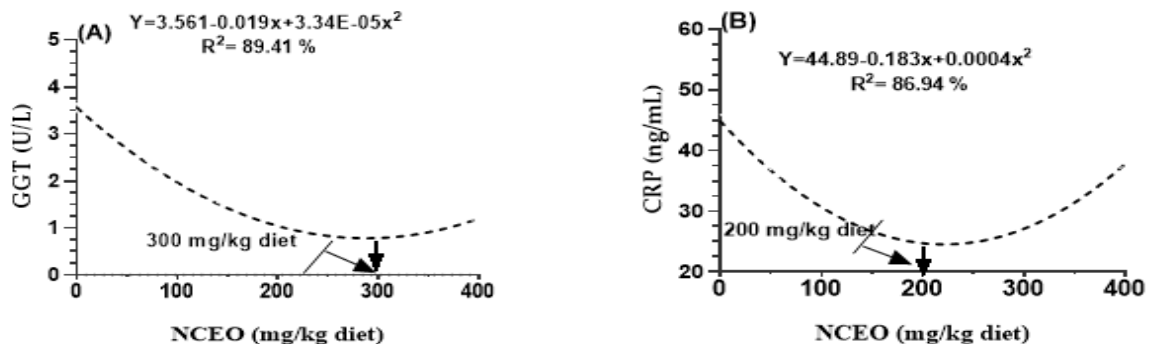


Fig. 5. A polynomial regression analysis between dietary levels of NCEO and gamma-glutamyl transferase (A – GGT) and C - reactive protein (B-CRP).

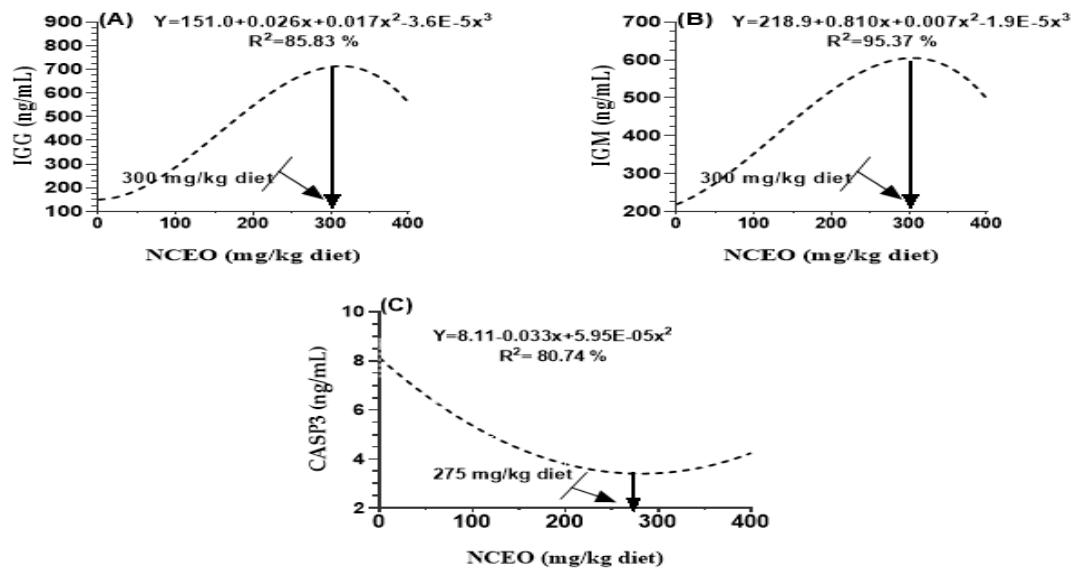


Fig. 6. A polynomial regression analysis between dietary levels of NCEO and immunoglobulin G (A – IgG), immunoglobulin M (B – IgM) and caspase-3 (C-CASP).

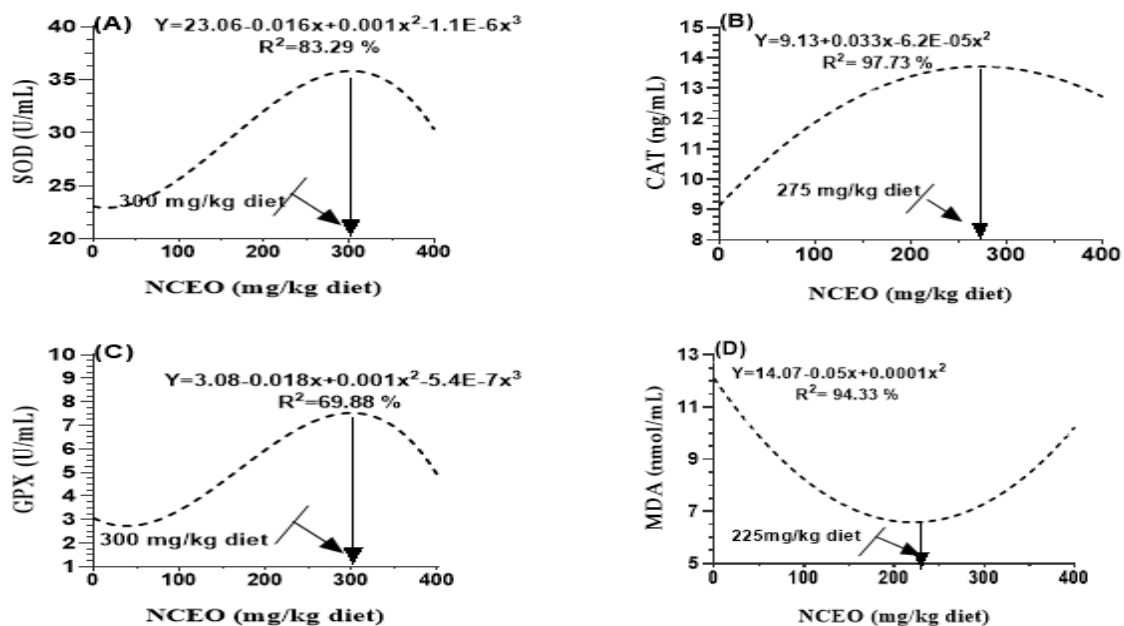


Fig. 7. A polynomial regression analysis between dietary levels of NCEO and super oxide dismutase (A –SOD), catalase (B – CAT), glutathione peroxidase (C-GPX) and malondialdehyde (D-MDA).

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تحسين النمو، الصحة والمناخ للماعز تحت ظروف الإجهاد الحراري باستخدام

مستحلب النانو من زيت الهيل

محمد مدحت¹، سامح عبدالنور¹، علي سليم علي سليم²، اسامه عبد المنعم¹، بكري عبد الغني خليل¹ وعلي الراجحي³

¹ قسم الإنتاج الحيواني، كلية الزراعة، جامعة الزقازيق، مصر.

² قسم الإنتاج الحيواني، كلية الزراعة، جامعه سوهاج، مصر

³ قسم الإنتاج الحيواني-والسمكي والداجني، كلية الزراعة، جامعة دمياط، مصر

الملخص

برزت تقنية النانو كاستراتيجية واعدة لتعزيز قابلية ذوبان المواد الكيميائية النباتية وتوافرها الحيوي وفعاليتها، ومعالجة القيود التي تواجهها محدوديتها وتطبيقاتها في الأنظمة البيولوجية الصناعية. هدفت هذه الدراسة الي توضيح دور زيت الهيل النانوي المستحلب (NCEO) على أداء النمو، والمعايير الفسيولوجية، قياسات الدم، والوظيفة المناعية، والإجهاد التأكسدي لدى الماعز النامية خلال فصل الصيف. قسمت اثنان وثلاثون ماعزًا بعمر أربعة أشهر عشوائيًا إلى أربع مجموعات (8 حيوان لكل مجموعة) تم تغذيتها علي عليه مدعمة بـ NCEO بجرعات 0 (NCEO0)، و100 (NCEO100)، و200 (NCEO200)، و400 (NCEO400) ملجم/كغم على التوالي. أظهرت النتائج أن إضافة NCEO حسنت أداء النمو بشكل ملحوظ ($P < 0.05$) وبالمقارنة مع NCEO0. لاحظ انخفاض معنوي في معدلات التنفس ودرجة حرارة المستقيم عند التغذية علي مستويات مختلفة من NCEO0. إضافة 200 او 400 ملجم حسنت بشكل معنوي كل من تعداد كريات الدم الحمراء والبيضاء ومستويات بورتين الدم، وانخفاض وظائف الكلى والكبد، ومستويات الدهون. وفيما يتعلق بالمناعة الخلوية، أظهرت مستويات IgG وIgM زيادة مكعبة استجابة لمكملات NCEO، مع تقدير الجرعات المثلى بـ 300 ملغ/كغم من NCEO. وعلى العكس، أظهرت مستويات الموت المبرمج للخلايا (Caspase-3) انخفاضًا معنويًا، مع جرعة مثالية قدرها 275 ملغ/كغم من NCEO. وأظهرت مستويات إنترلوكين-2 (IL-2) زيادة خطية مع زيادة تركيزات NCEO. وفيما يتعلق بالإجهاد التأكسدي، أظهرت مستويات MDA انخفاضًا معنويًا. وأظهرت TAC زيادة خطية مع زيادة مستويات الزيت النانو. أظهرت أنشطة SOD وGPX استجابة مكعبة لـ NCEO، مع جرعات مثالية عند 300 ملغم/كغم، بينما أظهر نشاط CAT استجابة تربيعية، مع جرعة مثالية عند 275 ملغم/كغم. تشير هذه النتائج إلى أن المكملات الغذائية التي تحتوي على 200-300 ملغم/كغم من NCEO يمكن أن تعزز بشكل كبير أداء النمو، قياسات الدم والكيميائية الحيوية، وحالة الأكسدة والاختزال، والوظيفة المناعية في الماعز النامية خلال فصل الصيف.

الكلمات الدالة: زيت النانو الهيل، الماعز، المناخ والنمو.