



Impact of *Chlorella Vulgaris* Microalgae as Anti Stress on Hematological Parameters and Thermoregulation of Boer Goats in Arid Subtropical Regions

Ayman Y. Kassab*¹; Hatem A. Hamdon¹; Waleed Sanosy²; Esraa M. Wardy¹

¹Department of Animal Production, Faculty of Agriculture, Univ. of New Valley, Egypt

²Department of Theriogenology, Faculty of Veterinary Medicine, Univ. of New Valley, Egypt

ABSTRACT

This work analyzed the impact of *Chlorella vulgaris* (CV) microalgae administration on some hematological parameters and thermoregulation of Boer goats subjected to heat stress. Fifteen lactating Boer goats (35 ± 1.8 kg BW) were assigned to three groups (5 goats each), control diet, C V supplemented diet (7.5 g/head/ day) and C V supplemented diet (15 g/head/ day) that were subjected to heat stress for 90 days. AT and RH% data were recorded and used in calculate the current THI. Blood samples were obtained from all goats biweekly from jugular vein. Physiological parameters were measured and recorded during the experimental days. The results obtained showed that CV increases RBC count, HGB concentration, and HCT. Whereas RT, RR and PR decreased significantly ($P < 0.05$) in Boer goats. In this study, we can conclude that C V could improve some blood hematological parameters and thermoregulation of Boer goats in arid subtropical regions.

* Corresponding author
Ayman Y. Kassab



Received: 03/07/2023

Revised: 30/08/2023

Accepted: 28/09/2023

Published: 01/10/2023



©2023 by the authors.
Licensee NVJAS, Egypt.
This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<http://creativecommons.org/licenses/by/4.0/>).

Keywords: Microalgae; hematological parameters; Thermoregulation; Boer goats

1. INTRODUCTION

Climate change is altering the planet's ecosystems, threatening the well-being of current and future generations. Governments strive to keep the increases in global temperature below 2° C and avoid climate change. After a long tracking of the global average temperature (from 1901 to 2001), it was noted that the earth is warming. The global average AT has increased by about 0.8 °C over the past 100 years. The temperature increase began in 1910, and then highly increased in the 2000s (IPCC, 2013). Livestock production systems will be affected by the projected increase in global temperature of 1.5°C between 2030 and 2050 (Soliman et al., 2022). The New Valley Governorate is in the Western Desert between 25°; 42& 30°; 47 E longitude, 22° 30' & 29° 30' N latitude and is located at an altitude of 77.8 meters above sea level. This area is dry and arid. The AT ranges between 43-48°C during summer days to 8 °C during winter nights (Nasreldin, et al, 2020; Kassab et al., 2020; Soliman et al., 2022).

Microalgae are photosynthetic microorganisms that convert carbon dioxide (CO₂) and sunlight into extremely valuable biomass. CV are single-celled algae that grow in fresh water and contains a large protein content 60% of DM, with a large amount of chlorophyll, 18 amino acids plus vitamins and minerals (Tsiplakou et al., 2017; Kholif and Olafadehan, 2021).

Adding CV in animal rations as a feed additive led to improved animal performance and production. This is due to the higher biological value which can stimulate the immune system, increase DMI, and enhance reproduction (Madeira et al., 2017; Ru et al., 2020; Kholif and Olafadehan, 2021). Because of their contents of fatty acids, essential amino acids, and other health-enhancing nutrients, CV are a feed additive or supplement for livestock.

This trial aimed to compare the impacts of adding two levels of CV microalgae on blood hematological parameters and thermoregulation of Boer goats under New Valley climatic conditions.

2. Materials and Methods

This trial was conducted at the farm in the animal production department, Faculty of Agriculture, New Valley University from 21st June till 21st September 2022.

3. Experimental Design and Diets

Fifteen lactating Boer goats (35 ± 1.8 kg BW) were randomly assigned to three groups (5 goats each), control diet according to NRC (2007), CV supplemented diet (7.5 g/head/day) and CV supplemented diet (15 g/head/day) that were subjected to heat stress for 90 days. The goat diet contained concentrate feed mixture (CFM) and Wheat straw WS. Ingredients of CFM presented in Table (1). The chemical composition of CFM and WS are presented in Table (2).

Table (1): Ingredients of CFM

Items	%
Corn%	55
Wheat bran%	21.5
Soya bean meal%	20
Limestone %	1.5
Di - calcium phosphate%	0.5
Yeast %	0.2
Bicarbonate %	0.3
Sodium chloride %	1

Table (2): Chemical composition of CFM and WS (on DM basis).

Item	DM	OM	CP	CF	Fat	Ash	NFE
CFM	89.76	93.72	14.76	14.65	3.39	6.28	60.92
WS	92.36	89.42	1.88	37.81	1.09	10.58	48.64

Measurement of Temperature–Humidity Index (THI)

AT and RH% were measured and recorded using (Temperature /Humidity Temperature thermometer) at 8:00 h and 14:00 h.

The formula: $THI = (0.8 \times Ta) + [(RH/100) \times (Ta - 14.4)] + 46.4$ (Mader et al., 2006) was used, where; RH: relative humidity and Ta °C: ambient temperature

Blood Sampling

Blood samples (2 mL / goat) were collected from the jugular-vein at 08:00 h biweekly into a tube containing K3 EDTA for the hematological analysis. Samples were analyzed for WBC, RBC, HGB, HCT, MCH, MCHC, MCV, and PLT by an automatic hematology cell counter (Dirui Bcc-3600).

Body temperature measurements

The thermoregulatory responses, RR and PR were determined biweekly before measuring the body temperature, RR was measured by counting flank movements at 15 s and multiplying by 4 to obtain breaths/min (Al-Qaisi et al., 2020). RT, °C was recorded using a digital thermometer. Skin, hair, and ear

temperature °C were measured by portable infrared thermometer (IRT).

Statistical Analysis

Data were statistically analyzed using SAS v.9.1 (SAS, 2004).

The differences among treatments were tested using Duncan's Multiple Range Test (Duncan, 1955).

The model used was $Y_{ij} = \mu + A_j + \xi_{ij}$

Y_{ij} = Observation traits,

μ = Overall mean,

A_j = Experimental treatment

ξ_{ij} = Random error

4. Results

Environmental measurements

AT °C, RH percent and THI through the trial period were shown in Table (3) and Figure (1). The average AT °C through the trial period ranged from 24.2o C to 27.3o C and at 08:00 hrs. 37.4o C to 42.5o C at 14:00 hrs. Moreover, the average RH% was 11% and 22%. Furthermore, the THI values were 67.4 to 71.1 at 08:00 hrs. and 76.5 to 80.53 at 14:00 hrs. through the trial period.

Table (3): Means of ambient temperature (°C) and relative humidity (%)

day	08:00		14:00	
	AT, °C	RH, %	AT, °C	RH, %
21/6/2022	26.3	18	38.3	15
1/7/2022	25.6	17	38.7	12
10/7/2022	24.8	16	42.5	13
21/7/2022	26.5	13	41.7	15
1/8/2022	24.4	18	40.8	11
10/8/2022	27.3	22	40.5	12
21/8/2022	25.8	15	39.7	11
1/9/2022	26.8	18	39.4	13
10/9/2022	24.5	17	38.5	16
21/9/2022	24.2	17	37.4	18

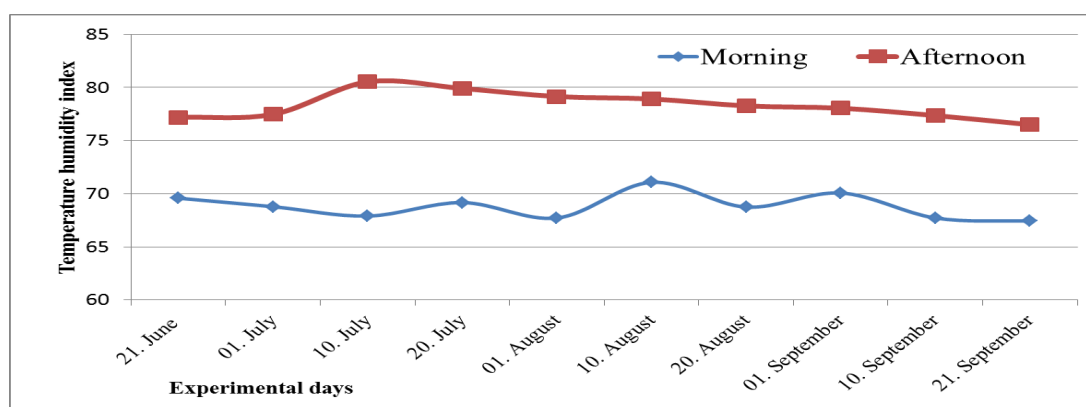


Figure 1: Temperature humidity index (THI)

Hematological parameters

Circulating leukocytes, HGB concentration,

erythrocytes, HCT, MCHC, MCH, MCV, and platelets are presented in Table (4).

Table (4): impact of CV on blood hematological parameters

Parameter	Control Means \pm SEM	CV 7.5 Means \pm SEM	CV 15 Means \pm SEM	P-values
WBC ($10^3/\mu\text{l}$)	11.83 \pm 0.41	10.08 \pm 0.66	11.01 \pm 0.81	0.203
RBC ($10^6/\mu\text{l}$)	9.61 ^b \pm 0.16	10.35 ^a \pm 0.24	10.17 ^{ab} \pm 0.17	0.039
HGB (g/dl)	11.27 ^b \pm 0.06	11.72 ^a \pm 0.10	11.64 ^a \pm 0.05	0.001
HCT %	31.98 ^b \pm 0.62	34.43 ^a \pm 0.45	33.35 ^{ab} \pm 0.68	0.030
MCV (fl)	47.93 \pm 0.94	47.55 \pm 0.79	47.71 \pm 0.89	0.957
MCH (pg)	17.43 \pm 0.46	17.12 \pm 0.38	17.02 \pm 0.33	0.756
MCHC (g/dl)	35.37 \pm 0.80	36.28 \pm 0.31	36.85 \pm 0.43	0.201
PLT	386.17 \pm 19.78	369.67 \pm 9.13	378.17 \pm 16.27	0.778

The number of RBCs, HGB concentration, HCT, in the CV 7.5 group goats were statistically higher ($P < 0.05$) than control. Whereas there was no difference observed between treatments on MCV, MCH, MCHC values, WBCs, and platelets. Dietary CV at level of 15g/day/head had no statistically significant differences on hematological parameters vs control group.

Body temperature variables

Table (5) illustrates that supplementation of CV microalgae led to decrease ST, HT, ET, whereas the differences were not significant. On the other hand, RT, RR and PR were decrease ($P < 0.05$) significantly in goats that treated with CV. Our results show that cv microalgae can improve thermoregulation in Boer goats subjected to heat stress.

Table (5): impact of CV on thermoregulatory responses

Parameter	Control Means \pm SEM	CV 7.5 Means \pm SEM	CV 15 Means \pm SEM	P-values
RT ($^{\circ}\text{C}$)	38.9 ^a \pm 0.09	38.64 ^b \pm 0.08	38.6 ^b \pm 0.05	0.021
RR (breath/min)	38.08 ^a \pm 0.46	36.08 ^b \pm 0.28	35.83 ^b \pm 0.52	0.002
PR (beat/min.)	66.25 ^a \pm 0.80	62.08 ^b \pm 1.02	62.33 ^b \pm 1.25	0.001
ST ($^{\circ}\text{C}$)	35.13 \pm 0.13	35.08 \pm 0.08	34.97 \pm 0.13	0.621
HT ($^{\circ}\text{C}$)	35.58 \pm 0.12	35.49 \pm 0.10	35.36 \pm 0.07	0.342
ET ($^{\circ}\text{C}$)	35.25 \pm 0.12	34.93 \pm 0.14	35.02 \pm 0.13	0.256

5. Discussion

Meteorological measures:

The thermal stress conditions were classifications as follows; Normal $THI \leq 74$; Alert $74 < THI < 79$; Danger $79 \leq THI < 84$; and Emergency $THI \geq 84$ (Mader et al., 2006). The THI values that were recorded in our study are in conformity with Kassab et al. (2017) who found that the values of THI ranged between 75.39 to 82.12 during hot season in New Valley. Likewise, The THI values were between 70.08 to 74.78 at 08:00 hrs and 75.33 to 83.01 at 14:00 hrs (Kassab et al., 2021). Under the same condition in New Valley, Soliman et al., (2022) noted the THI values were ranged from 79.43 to 85.46 at 14:00 hrs and 69.04 to 74.14 at 22:00 hrs indicating that the animals in New Valley were under heat stress, especially at 2 pm. Our data showed that goats were under the normal conditions at 8:00 hrs and under thermal stress at 14:00 hrs according to (Mader et al., 2006).

Hematological parameters:

Hematological parameters are one of the most important indicators to find the interaction between the environment impacts and blood characteristic from a physiolo-environmental standpoint (Ovuru and Ekweozor, 2004). Red blood cells (RBCs) serve as a carrier of HGB. that reacts with oxygen carried in the blood to form oxyhaemoglobin during respiration (Helms et al., 2018). Also, RBCs are involved in transportation of carbon dioxide and oxygen in the body (Mmereole, 2008; Isaac et al., 2013).

Through our results, we noticed the following, some hematological parameters were increased because of CV treatment such as, RBCs count, HGB concentration and HCT. According to many studies, feeding lactating cows and heifers with CV contributed to increase in HGB and increase in the RBCs and some forms of leukocytes (Bogdanova and Flerova, 2018). Also, El-Ratel (2017) reported that HGB, RBCs and HCT increased ($P < 0.05$) significantly in does feed on spirulina platensis microalgae. This Increase in HGB and RBCs within the

physiological norm indicates the improvement of redox reactions speed and oxygen availability for the tissues, thus strengthening the metabolic rate in the organs of animals.

Our results indicated that CV microalgae had no effect on WBC and platelet count of Boer goats. The betterment of some hematological parameters with CV supplementation will be reflected on improves the animal's health status and nutrient utilization.

Thermoregulatory responses:

The effect of CV microalgae to heat-stressed goats was clear when the AT was increased, which reducing the RT, RR, and PR. Hence, we accepted the hypothesis that microalgae in CV could reduce some of the harmful effects of heat stress on animal physiological responses and performance. The main homeostatic responses to heat stress in the animals included an increase in RT, PR, RR, drooling, panting, and sweating, with a decrease in DMI (Silanikove, 1992). Our results indicate there was significantly reduced in RT, RR and PR by CV microalgae. In addition, RT was significantly increased in the control group due to the inability to dissipate heat efficiently due to the elevation of THI.

The mechanism by which CV was reduced RT in treated goats was not investigated in our results, it may be through an antioxidant mechanism by enhancement of superoxide dismutase and catalase synthesis (Bogdanova and Flerova, 2018; Tsiplakou et al., 2018), a Powerful endogenous antioxidant molecule.

Interestingly, the RR and PR were significantly reduced by CV, which may be explained by effects of antioxidant that present in *Chlorella vulgaris* on the cardiac autonomic nervous system. Whereas goats supplemented with CV had decreased ST, HT, and ET which reduces the severity of heat stress.

6. Conclusions

In conclusion, the present results declare that supplementation with *Chlorella vulgaris* microalgae had beneficial effect on hematological parameters and thermoregulatory responses especially RT, RR,

and PR of Boer goats under hot climatic conditions. In addition, *Chlorella vulgaris* microalgae supplementation was effective to ameliorate the harmful effects of thermal stress.

7. REFERENCE

- Al-Qaisi, M., Horst, E. A., Mayorga, E. J., Goetz, B. M., Abeyta, M. A., Yoon, I., ... & Baumgard, L. H. (2020).** Effects of a *Saccharomyces cerevisiae* fermentation product on heat-stressed dairy cows. *Journal of dairy science*, *103*(10), 9634-9645.
- Bogdanova, A. A., & Flerova, E. A. (2018).** Bi^oC hemical and hematological composition of blood of cattle fed with *Chlorella*. *Regulatory Mechanisms in Biosystems*, *9*(2), 244- 249
- Duncan, D. B. (1955).** Multiple rang and multiple F test. *Biometrics* *11*: 1- 42
- El-Ratel, I.T. (2017)** Reproductive Performance, Oxidative Status and Blood Metabolites of Doe Rabbits Administrated with *Spirulina* Alga. *Egypt. Poult. Sci. Vol 37*(IV) 11531172
- Helms, C. C., Gladwin, M. T., & Kim-Shapiro, D. B. (2018).** Erythr^oC ytes and vascular function: oxygen and nitric oxide. *Frontiers in physiology*, *9*, 125, 1-9.
- IPCC. (2013).** Summary for policymakers. In *Climate Change 2013: The Physical Science Basis. Contribution of Working Group to the Fifth Assessment Report of the Intergovernmental Panel on Climate Change*. Cambridge University Press, Cambridge, UK and New York, NY, USA.
- Kassab, A. Y., Hamdon, H. A., and Mohammed, A. A. (2017).** Impact of probiotics supplementation on some productive performance, digestibility coefficient and physiological responses of beef bulls under heat stress condations. *Egyptian Journal of Nutrition and Feeds*, *20*(1), 29-39.
- Kassab, A. Y., Senosy, W., Hamdon, H., Daghash, H., and Soliman, A. (2020).** Impact of antioxidant supplementation on metabolic status and reproductive performance of Aberdeen Angus cows during seasonal thermal stress in arid subtropical regions. *Egyptian J. Anim. Prod*, *57*(1), 1-11.
- Kassab, A., Hamdon, H., Daghash, H., and Soliman, A. (2021).** Impact of Betaine Supplementation as Anti Stress on Some Hematological Parameters and Thermoregulatory Responses of Aberdeen Angus Cows in Arid Subtropical Regions. *New Valley Journal of Agricultural Science*, *1*(2), 89-97.
- Kholif, A. E., & Olafadehan, O. A. (2021).** microalgae in Ruminant Nutrition: a Review of the Chemical Composition and Nutritive Value. *Annals of Animal Science*, *21*(3), 789.608
- Kholif, A. E., Morsy, T. A., Matloup, O. H., Anele, U. Y., Mohamed, A. G., & El-Sayed, A. B. (2017).** Dietary *Chlorella vulgaris* microalgae improves feed utilization, milk production and concentrations of conjugated linoleic acids in the milk of Damascus goats. *The Journal of Agricultural Science*, *155*(3), 508-518.
- Madeira, M. S., Cardoso, C., Lopes, P. A., Coelho, D., Afonso, C., Bandarra, N. M., & Prates, J. A. (2017).** Microalgae as feed ingredients for lives ^oC k production and meat quality: A review. *Lives ^oC k. Science*, *205*, 111-121
- Mader, T.L., M.S. Davis and T. Brown-Brandl (2006).** Environmental factors influencing heat stress in feedlot cattle. *J. Anim. Sci.*, *84*:712-719.
- Mmereole, F. U. C. (2008).** The effects of replacing ground nut cake with rubber seed meal on the haematological and serological indices of broilers. *International Journal of Poultry Science*, *7*(6), 622-624.
- Nasreldin, N., Ewida, R. M., Hamdon, H., and Elnaker, Y. F. (2020).** Molecular diagnosis and bi^oC hemical studies of

- tick-borne diseases (anaplasmosis and babesiosis) in Aberdeen Angus Cattle in New Valley, Egypt. *Veterinary World*, 13(9), 1884.
- National Research Council, (2010).** *Guide for the care and use of laboratory animals.*
- NRC (2007).** *Nutrient Requirements of Small Ruminants: Sheep, Goats, Cervids, and New World Camelids.* Washington, DC: National Academies Press.
- Ovuru, S. S., & Ekweozor, I. K. E. (2004).** Haematological changes associated with crude oil ingestion in experimental rabbits. *African Journal of Biotechnology*, 3(6), 346348.
- Ru, I. T. K., Sung, Y. Y., Jusoh, M., Wahid, M. E. A., & Nagappan, T. (2020).** Chlorella vulgaris: A perspective on its potential for combining high biomass with high value bioproducts. *Applied Phycology*, 1(1), 2-11.
- SAS (2004).** *SAS/STAT 9.1 User's Guide. Statistical Analysis System.* Institute, Inc. Cary., NC.
- Silanikove, N. (1992).** Effects of water scarcity and hot environment on appetite and digestion in ruminants – A review. *Livest. Prod. Sci.* 30:175–194
- Soliman, A. S. H., Kassab, A. Y., Hamdon, H. A., Senosy, W., & Daghash, H. A. (2022).** Effect of Antioxidant supplementation on Some Hematological Parameters and Thermoregulatory Responses of Aberdeen Angus Cows During Hot Season in Arid Subtropical Regions. *New Valley Journal of Agricultural Science*, 2 (6), 544-551.
- Tsiplakou, E., Abdullah, M. A. M., Mavrommatis, A., Chatzikonstantinou, M., Skliros, D., Sotirakoglou, K., ... & Zervas, G. (2018).** The effect of dietary Chlorella vulgaris inclusion on goat's milk chemical composition, fatty acids profile and enzymes activities related to oxidation. *Journal of animal physiology and animal nutrition*, 102(1), 142-151.
- Tsiplakou, E., Abdullah, M. A. M., Skliros, D., Chatzikonstantinou, M., Fliemetakis, E., Labrou, N., & Zervas, G. (2017).** The effect of dietary Chlorella vulgaris supplementation on microorganism community, enzyme activities and fatty acid profile in the rumen liquid of goats. *Journal of Animal Physiology and Animal Nutrition*, 101(2), 275-283.