INTRODUCTION

Exposure of animals to heat stress stimulates a sequence of drastic changes regarding biological functions. It is include a decrease in feed efficiency, utilization, disturbances in water, change in protein, energy and mineral balances, enzymatic activities, hormonal secretions and blood metabolites. This ending by impairment both productive and reproductive performance. Heat stress also, altering natural immunity and making animals more vulnerable to disease (Habeeb et al., 2008 and 2018).

Rabbits are highly sensitive to heat stress due to the absence of sweat glands or any other means of eliminating excess body heat (Marai et al., 2008 and Morera et al., 2012). Several researches claimed that such adverse impacts are mainly linked to an extreme generation of free radicals and active oxygen species with decrease in antioxidant resistance (Ganaie et al., 2013 and Nisar et al., 2013). Hence, several in vivo and in vitro trials confirmed the importance of antioxidant supplementation in ameliorating heat stress effect (Alhidary et al., 2012 and McKee and Harrison, 2013). Vitamins, essential oils, fats, and amino acids are the main dietary supplements with marked antioxidant properties. But, for the time being, the use of natural antioxidants has paid great interest from both livestock producers and nutritionists (Tawfeek et al., 2014). Thus, recently many researchers are interested in finding safe and effective natural antioxidants which can be substituted the synthetic commercial antioxidant supplements such as butylated hydroxyl anisole (BHA), butylated hydroxy toluene (BHT), α-tocopherol and propyl gallate (PG) that have been used in order to reduce oxidative damages (Lee et al., 2008).
Algae have attracted a great deal of interest as alternative sources of nutrients. It is logical to consider that algae could be a key resource containing rich source of functional metabolites such as polysaccharides, proteins, peptides, amino acids, lipids, polyphenols, and minerals (Brown et al. 2014). Otherwise, diatoms were considered as promising source of sustainable antioxidants because they have effective radical scavengers, ability to adapt and rapidly grow either in open or closed cultivation facilities (Banerjee et al. 2011). Diatoms contribute about 40% of the marine primary production, constituting half of entire organic material produced in the planet (Rousseaux and Gregg, 2014). Interestingly, under certain conditions, diatoms produce the highest amounts of Polyunsaturated fatty acids (PUFAs) among phytoplankton groups (Leflaive and Ten-Hage, 2009). Diatoms can produce inhibitory compounds against bacteria, constituting an alternative to the use of chemicals to control pathogenic bacterial growth. Antibacterial activity has been detected in co-cultures of microalgae (Qasem et al., 2016, Molina-Cardenas and Sánchez-Saavedra, 2017, Badr et al., 2017, El-Sayed et al., 2018a and Ayoub et al., 2019). Lee et al. (2009) studied A. coffeaeformis diatom extracts for their potential antioxidant effects and they found that A. coffeaeformis exhibited lipid peroxidation inhibitory activity significantly higher than that of α-tocopherol. In addition, A. coffeaeformis is rich in hydrophilic and hydrophobic anti oxidative compounds with different anti-oxidative properties. Ayoub et al. (2019) indicates that the oral administration of A. coffeaeformis at three concentration of (10, 20 and 30g /kg diet) in Nile tilapia diets leads to enhance the growth performance, feed efficiency, serum lysozyme activity and improved total protein, albumin and globulin. Khatoon et al. (2009) showed that isolated marine diatoms (Amphora, Navicula and Cymbella) grown on substrate could be used as feed supplement in enhancing the growth and survival of Penaeus monodon postlarvae. However, still there is not enough data or researches about the effects of A. coffeaeformis or diatoms on rabbits or poultry performance.

Therefore, the objective of this study was to evaluate the effect of A. coffeaeformis diatoms Alga extract supplementation in drinking water on growth performance, carcass, physiological and antioxidant status of growing rabbits under heat stress conditions.

MATERIALS AND METHODS

Experimental design and management

The present study was carried out in El-Semman Unit for development of Rabbit Research, Faculty of Agriculture, Cairo University. It was lasted six weeks from 18 July to August, 29 2018. A total number of 60 weaned V-line rabbits were used (5.5 weeks old and average body weight was 853.4±22.98 gm). The experimental rabbits were divided randomly into four equal groups (n=15 each). In 1st group (C), rabbits were kept as control (drank water without additives); in the 2nd group rabbits drank water supplemented with vitamins and minerals (1 ml/liter, T1), in the 3rd group rabbits drank water supplemented with 0.5ml A. coffeaeformis alga extract (0.5 ml ACE/liter, T2); while in the 4th group rabbits drank water supplemented with 1ml A. coffeaeformis alga extract (1 ml ACE/liter, T3).

Rabbits are healthy and diagnosed as clinically free from internal and external parasites. Rabbits were kept under the same management and environmental conditions and housed in standard dimensions wired metallic cages (30 x 40 x 25 cm) (5 rabbits/cage) and equipped with feeding hoppers. All rabbit groups were fed and drink ad libitum. Feed ingredients and chemical composition of the concentrate pelleted experimental diet that cover the requirements of growing rabbits (according to the Agriculture Ministry Decree 1996) are shown in Table (1).

Alga extracts preparation

The locally isolate Bacillariophyta alga, Amphora coffeaeformis (El-Sayed et al., 2018b) was massively produced (Algal Biotechnology Unit, National Research Centre, Dokki, Giza, Egypt) based on F2 nutrient solution (Gillard and Rutherford, 1962). Outdoor mass production was performed within semi-closed photobioreactor 1200L capacity of fully transparent Zigzag shape photobioreactor (El-Sayed et al., 2018b). Outdoor nutrient solution was made from commercial fertilizers compounds as suggested early by El-Sayed et al. (2001). Technical processes were performed as described by Hassan et al. (2015).

Alga biochemical analysis

Chlorophylls and carotenoids determination

The acetone algal extract was transferred to separating funnel containing petroleum ether following by drying with anhydrous sodium sulphate and spectrophotometrically assayed (Davies, 1976). Total
chlorophyll, chlorophyll a and chlorophyll b were spectrophotometrically determined at wavelengths 645 and 663 nm (Enwereuzoh and Onyeagoro, 2014). β-carotene was determined at 436 nm, while total carotenoids were determined at 450 nm (Mustapha and Babura 2009).

Table (1): Ingredient and calculated analysis of the grower diets during 15-28 d.

| Ingredient          | %    | Calculated analysis |
|---------------------|------|---------------------|
| Alfalfa hay         | 35.2 | CP % 17.0           |
| Yellow corn         | 12.6 | CF% 12.6            |
| Soybean meal (44%)  | 14.5 | DE Kcal/kg 2500     |
| Wheat bran          | 14.3 | Ca % 1.15           |
| Barley              | 17.0 | Total P % 0.8       |
| Molasses            | 3.0  | Lys. % 0.93         |
| Lime stone          | 1.0  | Meth. % 0.34        |
| Mono calcium phos.  | 1.6  | Meth + Cys % 0.60   |
| Vit.&Min. Premix*   | 0.30 |                     |
| DL-Methionin        | 0.06 |                     |
| L-Lysine-HCl        | 0.05 |                     |
| NaCl                | 0.35 |                     |
| Total               | 100  |                     |

*Supplied per kg of diet: 12000 IU vit.A; 2200 IU vit. D3; 10 mg vit.E; 2.0 mg vit.K3; 1.0 mg vit.B1; 4.0 mg vit.B2; 1.5 mg vit.B6; 0.0010 mg vit.B12; 6.7 mg vit.PP; 6.67 mg vit. B5; 0.07 mg B8; 1.67 mg B9; 400 mg Choline chloride; 133.4 mg Mg; 25.0 mg Fe; 22.3 mg Zn; 10.0 mg Mn; 1. 67 mg Cu; 0.25 mg I and 0.033 mg Se

**Total lipid extraction and determination**

The Soxhlet extraction procedure is a semi-continuous process, which allows the buildup of the solvent in the extraction chamber for 5 to 20 min. The solvent surrounds the sample and is then siphoned back into the boiling flask. Multi-extractor units are available for extraction of lipids from several different samples or replicate runs of the same material. The procedure provides a soaking effect and does not permit channeling. The fact that polar and bound lipids are not removed is a drawback to the procedure. Extraction was performed using n-hexane (60-80°C) and 500 ml flat-bottom flask in three replicates for 12 hours.

**Determination of fatty acids**

**Methylation of fatty acids**

The fatty acids of oil were converted to methyl esters using methyl alcohol. 3 % sulphoric acid in methanol was prepared (3ml H2SO4 + 97 ml methanol). Reflexed at 90°C for 3 hrs and re-extracted with n-hexane. Leave over night with sodium sulphate. Whole extract was concentrated by rotary evaporator and then Inject into G.C.

**Identification and determination of fatty acids by Gas Chromatography (GC)**

Perkin Elmer Auto System XL; equipped with flame ionization detector (FID); fused silica capillary column DB-5 (60 x 0.32 mm) was used. Oven temperature was maintained initially at 150°C and programmed from 150 to 240°C at rate 3°C.min⁻¹. Injector temperature (230°C); detector temperature (250°C) and helium (carrier gas) flow rate at 1 ml.min⁻¹ were maintained.

**Total phenolic, antioxidant**

Total phenolic (Singleton and Rossi, 1965); total reducing power Paget and Barnes, (1964) and total antioxidant activity (Prieto et al., 1999). Furthermore, percentage of the antioxidant activity was evaluated by method described by Brand-Williams et al. (1995) using DPPH (2,2-diphenyl-1-picryl-hydrazyl-hydrate) for initiation of the free radicals.Total antioxidant capacity (TAC) of Amphora acetone extract was spectrophotometrically estimated according to method described by Koracevic et al. (2001) at wavelength 505 nm.
Experimental measurements

Climate changes

Changes in maximum and minimum ambient air temperatures (AT, °C) and relative humidity (RH, %) were daily recorded inside the rabbitry using electronic digital thermo-hygrometer. Also, the relationship between AT and RH, which termed to temperature-humidity index (THI), was calculated according to the equation of Marai et al. (2002). The equation was as follow:

\[
THI = db °C - [(0.31 - 0.31 \times RH / 100) (db °C - 14.4)],
\]

where \( db °C \) represents the dry bulb temperature in degrees Celsius and \( RH \) is the relative humidity expressed as a percentage.

The THI values obtained were then classified as follows; (27.8 and less) absence of heat stress, (27.8 - 28.9) moderate heat stress, (28.9 – 30.0) severe heat stress and (30.0 and more) very severe heat stress (Marai et al., 2002).

### Table 2: Maximum and minimum air temperatures (AT, °C), and relative humidity (RH, %)

| Variable | Maximum       | Minimum       |
|----------|---------------|---------------|
| AT (°C)  | 35.79 ±0.18   | 27.88 ±0.16   |
| RH (%)   | 71.14 ±0.55   | 28.43 ±0.98   |
| THI (units) | 33.67 ±0.15 | 24.71 ±0.12   |

Relative humidity (RH, %). Calculated values for the temperature-humidity index (THI) inside the rabbitry during the experimental period (Mean ±SE).

Productive performance

Throughout the experimental period, body weight was recorded weekly and average body weight gain was calculated. Feed intake was determined precisely and given as grams per rabbit per week. From each cage, feed residuals were collected daily, weighed and taken into consideration for the calculation of feed intake and feed conversion ratio.

Slaughtering and carcass traits

At the end of the experimental period (12 weeks), from each group, randomly 6 rabbits were taken, fasted for 12 h, individually weighed and immediately slaughtered. After complete bleeding, pelt, viscera and tail were removed, and then the carcass and its components were weighed as edible parts. The non-edible parts containing lung, liver, spleen, stomach, intestine, and cecum weighed as percentage of pre-slaughter weight. Dressing percentage was calculated by dividing the hot dressed carcass weight by pre-slaughter weight and expressed as a percentage.

Blood biochemical parameters

At the end of experiment, blood samples (5 ml from four groups) were randomly collected during slaughter time. Plasma was separated from blood by centrifugation at 3000 rpm for 15 min and stored at −20°C till assayed. Plasma total protein, albumin, total cholesterol and triglycerides were measured calorimetrically using commercial kits (purchased from Bio-diagnostic, Cairo, Egypt) according to the manufacturers’ instructions. Total protein was determined according to Orsonneau et al. (1989). Albumin was determined according to the method of Doumas et al. (1971). Plasma globulin concentration was calculated by the difference between total protein and albumin. Triglycerides were determined according to Wahlefeld (1974). Blood plasma malondialdehyde (MDA), Total antioxidant capacity (T-AOC) was determined according to Koracevic et al. (2001) and catalase (CAT) activity was measured according to Aebi (1984).

Statistical analysis

Data were analyzed by the least square analysis of variance using the General Linear Model Procedure (SAS, 2004). The design was one way analysis and the model was as follows:

\[
Y_{ij} = \mu + T_i + e_{ij} \]

Where, \( Y_{ij} \) = any observation of \( j^{th} \) animal within \( i^{th} \) treatment. \( \mu \) = overall mean. \( T_i \) = effect of \( i^{th} \) treatment (i: 1-4). \( e_{ij} \) = experimental error.

Duncan Multiple Range Test (Duncan, 1955) was used to test the level of significant among means.
RESULTS AND DISCUSSION

**Chlorophylls and carotenoids**

Growth conditions. This goes back to its growth pattern, where this alga able to complete its life Chlorophyll content of *Amphora* as well as most of its familiar is very sensitive to the ambient cycle within a wide range of nutrients content and salinity margin. It is closely associated with the carotenoids content, where decreasing of chlorophyll content is accompanied with the rise of carotenoids and oils with a marked decrease in protein content (Table 3).

| Pigment               | Concentration (mg.g⁻¹) |
|-----------------------|------------------------|
| Total chlorophyll     | 28.09 ± 0.04           |
| Chlorophyll a         | 19.94 ± 0.06           |
| Chlorophyll b         | 7.71 ± 0.05            |
| Total carotenoids     | 10.94 ± 0.04           |

**Table (3): Chlorophylls and carotenoids content of three batches outdoor grown A. coffeaeformis**

**Total lipid and fatty acids**

On the average, oil content of *Amphora* ranged reached about 7.0% and the differences were found in concern to outdoor growth condition (temperature and harvesting time). In addition, total unsaturated fatty acids reached 75.0 of total fatty acids and its fraction was 21.43 of ω3 and 12.89 of ω6-fatty acids. (Table 4).

**Table (4): Fatty acids methyl ester profile of A. coffeaeformis**

| C No.      | Fatty acid                      | %  |
|------------|---------------------------------|----|
| C14:0      | Myristic acid                   | 3.33|
| C14:1      | Myristoleic acid                | 1.42|
| C16:0      | Palmitic acid                   | 14.27|
| C16:1      | Palmitoleic acid                | 7.88|
| C18:0      | Stearic acid                    | 2.19|
| C18:1      | Oleic acid                      | 19.26|
| C18:2 n6   | Linoleic acid                   | 10.06|
| C18:3 n6   | ω-linolenic acid (GLA)          | 1.97|
| C18:3 n3   | α-linolenic acid                | 7.69|
| C20:0      | Arachidonic acid                | 4.16|
| C20:1      | Eicosenoic acid                 | 3.16|
| C20:2      | Eicosadienoic acid              | 2.78|
| C20:3 n3   | Eicosatrienoic acid             | 3.14|
| C20:4      | Eicosatetraenoic acid           | 6.02|
| C22:1      | Erucic acid                     | 1.06|
| C20:5 n3   | Eicopentanoic acid              | 4.19|
| C22:6 n3   | Docosahexaenoic acid            | 6.41|
| TSFA       |                                 | 24.96|
| TUSFA      |                                 | 75.04|

**Total phenolic**

Phenolic compounds in the acetone extract of *A. Coffeaeformis* are presented in Table (5).
Table (5): HPLC analysis of the phenolic in the acetone extract of *A. Coffeaeformis*

| Compound                | Concentration (μg·g⁻¹) |
|-------------------------|------------------------|
| Gallic acid             | 20.19                  |
| Protocatechuic acid     | 17.63                  |
| p-Hydroxybenzoic acid   | 6.12                   |
| Catechin                | 41.17                  |
| Chlorogenic acid        | 12.56                  |
| Caffeic acid            | 16.35                  |
| p-Coumaric acid         | 36.14                  |
| Cinamic acid            | 14.09                  |

**Antioxidant activity**

Table (6): Total polyphenols and antioxidant efficiency of *A. coffeaeformis* acetone extracts against free radicals

| Polyphenol mg100 g⁻¹ gallic | Reducing power μg.ml⁻¹ | Antioxidant capacity mg. g⁻¹ gallic | Antioxidant Activity % |
|-----------------------------|------------------------|-----------------------------------|------------------------|
| 0.531±0.004                 | 8.01 ± 0.041           | 85.22±0.09                        | 87                     |

**Productive performance**

The results of the effect of ACE in drinking water on V-line rabbits growth performance under heat stress conditions are presented in Table (7). The results showed that there were no significant differences between groups in final body weight or body weight gain at the age of 12 week. However, group T2 recorded significantly (P<0.05) the highest feed intake and the worst feed conversion. While, the best feed conversion recorded by T1(1ml vit.&min. mix/L drinking water) and T3 (1ml ACE/L drinking water) compared to T2 (0.5 ml ACE/L drinking water).

The improvement of feed conversion with supplementation *A. coffeaeformis*, may attribute to the improved intestinal tract conditions and nutrient availability which due to the antimicrobial effects of *Amphora* (Ayoub et al., 2019). In Addition, *A. coffeaeformis* contains several nutrients, especially, unsaturated fatty acids that reached 75.0 of total fatty acids and its fraction was 21.43 of ω3 and 12.89 of ω6-fatty acids. (Table 4) and that may improve growth. Moreover, through attenuation of oxidative stress, enhancement of antioxidant enzymes activities through high contents of phenolic compounds (Table 5). These results are in agreement with the finding of Abdelnour et al. (2019) who supplemented rabbit diets with *Chlorella vulgaris* Microalgae. Also, with Peiretti and Meineri (2008), Kim et al. (2010), Seyidoğlu and Galip (2013) and Khanna et al. (2016) who showed that the final weight, weight gain did not differ significantly as a result of supplemented rabbit diets with *Spirulina platensis* microalgae. Moreover, Ayoub et al. (2019) showed that supplemented Nile tilapia diets with *A coffeaeformis* diatoms algae with three concentration (10, 20 and 30g / kg diet) leads to enhance the growth performance, feed efficiency.

Table (7): Productive performance of growing V-line rabbits drinking water supplemented with ACE.

| Trait                      | Treatment               | SEM  |
|----------------------------|-------------------------|------|
|                            | C          | T1   | T2   | T3   |
| Initial body weight (IBW, g)| 852.5     | 861.9 | 849.2 | 849.6 | 47.5 |
| Final body weight (FBW, g)  | 1740.8    | 1815.7 | 1692.3 | 1777.8 | 51.1 |
| Total body gain (TBG, g)   | 888.3     | 953.8 | 843.1 | 928.2 | 39.8 |
| Total feed intake (TFI, g) | 3615.1ab  | 3475.3b | 3672.9a | 3500.1ab | 60.8 |
| Feed conversion ratio (FCR) | 4.17ab    | 3.69b | 4.55a | 3.81b | 0.21 |

*ab Means bearing different superscripts within the same row are significantly different (P<0.05).
Control = rabbits drank water without additives, T1 = rabbits drank water supplemented with vitamins and minerals, T2 = rabbits drank water supplemented with 0.5ml ACE, T3 = rabbits drank water supplemented with 1ml ACE.
**Carcass traits**

The results of carcass traits that shown in Table (8) shows that supplementation of rabbit diets with ACE at the level of 0.5 or 1 ml/l drinking water had no impact on the percentage of internal organs (kidney, liver, heart, spleen, lung, stomach and ceacum) or ceacum length. However, T1 group recorded significantly (p < 0.05) the highest dressing percentage compared to T2 group. This results are in agreement with Abdelnour et al. (2019) who indicated that supplemented the rabbit diets with *Chlorella vulgaris* microalgae did not induce significant differences (p > 0.05) in carcass traits (dressing percentage, giblets, heart, kidney, lung, and liver) as compared to the control animals. However, Kim et al. (2010) after 8 weeks of treatment rabbits with 1 or 5% *Spirulina platensis* microalgae, organ weights were not significantly different in spleen, kidney, and heart among the groups. Moreover, *Spirulina microalgae* addition did not significantly influence the carcass yield or the proportions of the various carcass parts and organs (Peiretti and Meineri, 2011).

**Table (8): Carcass traits of growing V-line rabbits supplemented with ACE in drinking water.**

| Trait | Treatment | C       | T1      | T2      | T3      | SEM |
|-------|-----------|---------|---------|---------|---------|-----|
| Pre-slaughter weight (g) |          | 1687.5  | 1718.3  | 1759.2  | 1759.2  | 55.33 |
| Dressed weight (g)       |          | 1065.0  | 1100.8  | 1065.0  | 1092.5  | 41.36 |
| Empty carcass (g)        |          | 983.3   | 991.7   | 972.5   | 1016.7  | 42.76 |
| Dressing %               |          | 63.1ab  | 64.1a   | 60.4b   | 62.0ab  | 0.95 |
| Kidney %                 |          | 0.74    | 0.79    | 0.80    | 0.76    | 0.04 |
| Liver %                  |          | 2.8     | 2.8     | 2.9     | 3.0     | 0.18 |
| Heart %                  |          | 0.31    | 0.31    | 0.30    | 0.32    | 0.02 |
| Spleen %                 |          | 0.05    | 0.05    | 0.05    | 0.05    | 0.01 |
| Lung %                   |          | 1.0      | 0.95    | 0.88    | 0.81    | 0.12 |
| Stomach %                |          | 5.7     | 5.5     | 5.7     | 5.1     | 0.29 |
| Ceacum %                 |          | 4.8     | 4.9     | 7.0     | 5.6     | 0.71 |
| Ceacum length (cm)       |          | 45.0    | 44.3    | 45.7    | 45.4    | 1.49 |

*a,b* Means bearing different superscripts within the same row are significantly different (P<0.05).  
Control = rabbits drank water without additives, T1 = rabbits drank water supplemented with vitamins and minerals, T2 = rabbits drank water supplemented with 0.5ml ACE, T3 = rabbits drank water supplemented with 1ml ACE.

**Blood biochemical components**

**Blood hematological**

The results of blood hematology are presented in Table (9). There were no significant different among groups in Hematocrit, Total leucocytes count, Lymphocytes and Neutrophils percentage, while T3 (1 ml ACE/L drinking water) group recorded significantly (p < 0.05) the highest values of Hemoglobin and Red blood cells count (10.73 and 5.45*10⁶ respectively) compared to control (9.07 and 4.33*10⁶ respectively). These results are in contrast with the finding of Abdelnour et al. (2019) who indicated that dietary *Chlorella vulgaris* Microalgae supplementation had a significant effect (p < 0.05) in the all blood hematology traits were detected except for hemoglobin and red blood cells count. El-Raief (2017) indicated that treatment of does with *Spirulina platensis* significantly (P<0.05) increased hemoglobin (Hb) concentration, count of red blood cells (RBCs) and hematocrit value (Ht). In the current study improving of Hb and RBCs (p < 0.05) in A coffeaeformis group T3 might be due to the strong antioxidant effect of on hematopoietic cells, which appears to be particularly vulnerable in the presence of unchecked accumulation of reactive oxygen species, ROS (Kong et al., 2004). Also, high content of USFs and omega3 fatty acid of A. coffeaeformis extract (Table 4). El-Meghazy et al. (2014) showed that feeding diet supplemented with omega-3 were significantly increased the percentages of hemoglobin, platelets and the mean corpuscular hemoglobin.

**Blood metabolites**

Results of Blood metabolites are shown in Table (10). Supplementation of ACE to growing rabbit drinking water insignificantly affected on the total protein, albumin, globulin, total cholesterol, triglycerides, alanine aminotransferase (GPT), uric acid and creatinine. While, T1 (vit. and min. group) recorded significantly (p < 0.05) the highest value of Aspartate aminotransferase (GOT) compared to

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control group. These results are in agreement with Abdelnour et al. (2019) who showed that most of the serum parameters were non-significantly different by Chlorella vulgaris microalgae supplementation in rabbit diets. Also, Seyidoğlu and Galip (2014) and Khanna et al. (2016) who indicated that there were no significant changes in serum biochemical indices as a result of supplemented rabbit diets with Spirulina platensis. While there were significant decreased triglycerides, total cholesterol and activity AST and ALT in blood plasma in Spirulina platensis microalgae groups compared to control (El-Ratel, 2017). Moreover, Ayoub et al. (2019) showed that serum ALT and AST were within normal values in all treatment groups treated with A. coffeaeformis microalgae compared the control group of Nile tilapia fish.

Table (9): Blood hematological of growing V-line rabbits drinking water supplemented with ACE.

| Parameters                  | Treatments | SEM |
|-----------------------------|------------|-----|
|                             | C          | T1  | T2  | T3  |
| Hemoglobin (Hb)             | 9.07b      | 10.4ab | 9.22b | 10.73a | 0.39 |
| Hematocrite (Ht)            | 31.23      | 36.5 | 31.7 | 36.5 | 1.53 |
| Red blood cells (RBCs)*10⁶  | 4.33c      | 5.23ab | 4.62bc | 5.45a | 0.22 |
| Total leucocytes            | 6600       | 7266 | 6725 | 7566 | 980  |
| Lymphocytes                 | 40.0       | 47.0 | 44.5 | 37.33 | 4.45 |
| Neutrophils                 | 50.33      | 40.0 | 45.75 | 50.33 | 5.17 |

Means bearing different superscripts within the same row are significantly different (P<0.05).

Control = rabbits drank water without additives, T1 = rabbits drank water supplemented with vitamins and minerals, T2 = rabbits drank water supplemented with 0.5ml ACE, T3 = rabbits drank water supplemented with 1ml ACE.

Table (10): Blood metabolites of growing V-line rabbits drank water supplemented with ACE.

| Parameter              | Treatment | SEM |
|------------------------|-----------|-----|
|                        | C         | T1  | T2  | T3  |
| Total protein mg/dl    | 6.02      | 6.13 | 6.31 | 6.10 | 0.47 |
| Albumin mg/dl          | 3.58      | 3.68 | 3.67 | 3.54 | 0.25 |
| Globulin mg/dl         | 2.44      | 2.45 | 2.64 | 2.57 | 0.23 |
| Total cholesterol mg/dl| 196.67    | 186.15 | 195.75 | 198.83 | 11.81 |
| Triglycerides mg/dl    | 132.10    | 129.22 | 130.66 | 133.69 | 8.55 |
| ALT U/l                | 62.15     | 68.37 | 63.90 | 68.11 | 3.47 |
| AST U/l                | 64.28b    | 81.98a | 65.37b | 74.79ab | 4.63 |
| Uric acid mg/dl        | 5.55      | 5.46 | 5.46 | 5.60 | 0.38 |
| Creatinine mg/dl       | 0.90      | 0.86 | 0.88 | 0.92 | 0.11 |

Means bearing different superscripts within the same row are significantly different (P<0.05).

Control = rabbits drank water without additives, T1 = rabbits drank water supplemented with vitamins and minerals, T2 = rabbits drank water supplemented with 0.5ml ACE, T3 = rabbits drank water supplemented with 1ml ACE.

Blood antioxidant activity

The effect of ACE on blood antioxidant activity of rabbits under heat stress conditions are summarized in Table (11). All supplemented groups had significantly (p < 0.05) lower values of TAOC compared to control. While A. coffeaeformis extract recorded significantly (p < 0.05) better values of malondialdehyde (MDA) and catalase activity compared to control and T1. In present study decreasing of plasma MDA as a result of supplementation rabbit drinking water with ACE is an index of lipid peroxidation and oxidative stress decreasing (Safari et al., 2018). Also, the higher (P<0.05) activity of catalase enzyme (192.15, 212.80 and 249.30 in T1, T2 and T3 respectively) considered as indication increasing cellular defense against oxygen free radicals and oxidative stress (Bernabucci et al., 2002). It could be concluded that A. coffeaeformis algae extract enhanced the antioxidative status of rabbits by minimizing lipid peroxidation and increase the activity of catalase.

These results are in agreement with Abdelnour et al. (2019) who showed that supplementing growing rabbit diets with Chlorella vulgaris microalgae reduced the serum levels of malondialdehyde (MDA) compared to the control. While, No significant changes were detected in the activities of TAC compared to control. Also, El-Ratel and Gabr (2019) showed that Spirulina platensis supplementation group had significantly (p<0.05) better, antioxidant capacity (total antioxidant capacity, glutathione, malondialdehyde and catalase) in heat stressed rabbits. Also, El-Ratel 2017 indicated that total
antioxidant capacity (TAC) increased significantly (P<0.05) in blood plasma of doe rabbits administrated with Spirulina Alga. In addition, Kim et al. (2010) showed that Oxidative stress biomarkers were significantly improved in the liver and red blood cells of rabbits fed *Spirulina platensis*. Moreover, Mobarez et al. (2018) indicated that supplementing laying hen diets with 3 g SP/kg diet resulted in a significant increase (P≤0.01) in TAOC compared to the control group. In Addition, El-Sayed (2018) indicated that acetone extract of *A. coffeaeformis* alga exhibited the highest scavenging activity against attack of free radicals generated as a result of oxidative stress and induced by paracetamol in liver tissues in rats.

### Table (11): Blood antioxidant activity of growing V-line rabbits drinking water supplemented with ACE

| Parameter       | Treatment | C    | T1   | T2   | T3   | SEM |
|-----------------|-----------|------|------|------|------|-----|
| TAOC (mmol/l)   |           | 1.14\(^a\) | 0.84\(^b\) | 0.77\(^b\) | 0.67\(^b\) | 0.06 |
| Catalase (U/g)  |           | 152.90\(^c\) | 192.15\(^b\) | 212.80\(^b\) | 249.30\(^a\) | 9.84 |
| MDA (mmol/l)    |           | 2.73\(^a\) | 2.32\(^b\) | 1.90\(^c\) | 1.74\(^c\) | 0.07 |

\(^a\), \(^b\), \(^c\) Means bearing different superscripts within the same row are significantly different (P<0.05).

Control = rabbits drank water without additives, T1 = rabbits drank water supplemented with vitamins and minerals, T2 = rabbits drank water supplemented with 0.5ml ACE, T3 = rabbits drank water supplemented with 1ml ACE.

### CONCLUSION

It can be concluded that the present study demonstrated that *A. coffeaeformis* alga extract have the potential to be used as a sources of natural antioxidant and nutrients for growing rabbits without causing any adverse effects on growth or physiological functions, and the best dose in drinking water is 1ml/L. Moreover, there are not enough studies about the effects of diatoms on animals. So, we need more researches about that.

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المغذة على طحلب الامفورا

V-line

الإداء الإنتاجي والفسيولوجي والنشاط المضاد للاكسدة لارانب

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أجري هذا البحث بهدف تقييم استخدام مستخلص طحلب الامفورا كفيوفورمس في مياه الشرب تحت ظروف الإجهاد الحراري

أولا : تم تحضير الطحلب في وحدة بيوتكنولوجيا الطحالب بالمركز القومي للبحوث مع تقدير تركيب الاحماض الدهنية ونشاط المعضد

المضاد للكيسيك

ثانيا : تم استخدام عدد 60 أرنب خط V-line على مدة 5.5 أسابيع. وتم تقسيمهم إلى أربعة مجموعات (15 أرنب لكل مجموعة) وكان

V-line

تقرير النتائج هو التالي:

1. لم يؤدي استخدام طحلب الامفورا لم يؤدي إلى اختلافات معنوية في الوزن النهائي للجسم أو زيادة في الوزن بينما تحسن معنوي استخدام

2. لم تؤثر المعاملات المختلفة على مواصفات الذبيحة ووزان الأعضاء الداخلية ومؤشرات كيميائية البالغة كان هناك زيادة معنوية في نسبة الهيموجلوبين للمجموعة الرابعة, أيضا سجلت معدلات مادة MDA وزيادة نشاط

نستنتج من تلك الدراسة أنه في حالة الإجهاد الحراري أو التأكسدي أو الإجهاد الناجم عن عوامل أخرى يمكن استخدام طحلب الأمفورا كفيوفورمس في مياه الشرب بمعدل 1 مل / لتر ماء شرب. دون تأثير سلبي على الحالة الصحية والفسيولوجية للارانب.