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EGYPTIAN ACADEMIC JOURNAL OF

BIOLOGICAL SCIENCES ZOOLOGY

ISSN
2090-0759

WWW.EAJBS.EG.NET

Vol. 17 No. 1 (2025)

Citation: *Egypt. Acad. J. Biolog. Sci. (B. Zoology) Vol. 17(1) pp:137-165 (2025)*
DOI: 10.21608/EAJBSZ.2025.419744

Egypt. Acad. J. Biolog. Sci., 17(1):137-165(2025)

Egyptian Academic Journal of Biological Sciences
B. Zoology
ISSN: 2090 – 0759
Res. Ethics No.: 03-2024-0009
http://eajbsz.journals.ek b.eg/

Effect of Microalgae *Nannochloropsis oculata* and *Moringa oleifera* Leaves Additives on Growth Performance and Blood Constituents of Hi-Plus Rabbits Under North Sinai Conditions, Egypt

Elham E. Elhady[1]; Hemmat M. Abdelhafez[1]; Morsy, A. S.[2]; Farag, B. M.[2] and Doaa Gewily[1*]

[1]Department of Zoology, Faculty of Science (Girls), Al-Azhar University, Cairo, Egypt.
[2]Department of Animal and Poultry Physiology, Desert Research Center,Egypt.
***E-mail:** doaagewily.el.8.172@azhar.edu.eg

ARTICLE INFO
Article History
Received:24/2/2025
Accepted:28/3/2025
Available:2/4/2025

Keywords:
Blood parameters; Moringa oleifera; Nannochloropsis oculata algae, rabbit; productive performance.

ABSTRACT

This study set out to explore the impact of naturally occurring antioxidants present in microalgae *Nannochloropsis oculata* (NCO) and/or *Moringa oleifera* (MO) leaves on the blood constituents and productive performance of Hi-Plus growing rabbits in North Sinai. Sixty weaned Hi-Plus rabbits were categorized into four equal treatments at random. Every treatment was divided into three equal replicates at random. The first treatment was used as a control and did not include supplements. In addition to the basic diet, the second treatment rabbits were given 20 g of microalgae NCO / kg diet (2.0%). The rabbits in the third treatment were fed the basic diet plus 50 g of MO / kg diet (5.0%). The rabbits in the fourth treatment were fed the basal diet plus 20 g NCO and 50 g MO/ kg diet. The findings demonstrated that the rabbits receiving MO had higher body weights (BW) at 8 and 9 weeks ($P<0.05$) than the rabbits in the control group. When compared to the control group, the rabbits fed (NCO + MO) had the highest ($P<0.05$) BW at 10th to 12th weeks. At 7-8 weeks, the rabbits that received MO gained more body weight ($P<0.05$) than the rabbits in the control group, NCO, and NCO+MO. When compared to the rabbits in the control group, the rabbits fed NCO+MO showed the significantly highest body weight gain at 9–10, 10–11, and 11–12 weeks, in addition to the highest overall weight gain. The NCO+MO group demonstrated improvement ($P<0.05$) in feed conversion at 5–6, 9-10, 10-11, 11-12 and TFC compared to the control group. Plasma total protein and globulin concentrations were increased ($P<0.05$) in the rabbits fed NCO+MO compared to the control group. Cholesterol, low-density lipoprotein (LDL), aspartate aminotransferase (AST) and alanine aminotransferases (ALT), and urea levels were decreased ($P<0.05$) in the rabbits fed NCO, MO, and NCO+MO compared to the control group. Glutathione peroxidase (GSX) and total antioxidant capacity were increased ($P<0.05$) in rabbits fed NCO, MO, and NCO+MO as compared with the control group. However, malondialdehyde (MDA) was decreased ($P<0.05$) in rabbits fed NCO, MO, and NCO+MO as compared to the control group. Insulin, growth, and triiodothyronine hormones were increased ($P<0.05$) in the rabbits fed NCO, MO, and NCO+MO compared to the control group. Examination of the liver tissue sections seven weeks post NCO, MO, and NCO+MO feeding showed well-developed liver architectures with a normal appearance of liver cells and blood sinusoids. In conclusion, marine microalgae *Nannochloropsis oculata* at 2% and *Moringa oleifera* at 5% added to the diets of Hi-Plus growing rabbits in North Sinai settings improved growth performance, certain hematological parameters, hormonal patterns, and oxidative status by shielding the tissues from oxidative damage without negatively affecting the health of the rabbits.

INTRODUCTION

Rabbit meat is renowned for being low in cholesterol, high in proteins, and having a desirable percentage of polyunsaturated fatty acids (PUFA). With protein making up 80% of the energy value, it has reasonably high energy content (603 kJ/100g meat). Because of its favorable qualities, including its fatty acid profile, mineral and vitamin contents, and potential for additional enrichment through feeding, rabbit meat may be perfect for creating specialized functional foods (Hassan *et al*., 2015). Humans may consume excellent meat from rabbits, and they may be a significant way to address certain of the problems connected with the meat crisis (Reda *et al*., 2024).Researchers have shown that adding natural growth promoters to rabbit diets, including prebiotics, probiotics, and phytogenic supplements, can improve immunity responses, efficiency of feed, and growth performance while reducing high rates of morbidity and mortality, particularly during the weaning phase (Abdelnour *et al*., 2018; Abd El-Hack *et al*., 2019). Furthermore, much research has shown that including plants that are rich in polyphenols in a rabbit's diet can improve its growth and pace of digestion (Alagawany *et al*., 2018; Elwan *et al*., 2020).

To overcome the challenges brought on by feed shortages that impede animal production, researchers in Egypt have been looking for safe and efficient natural feed additives to improve productivity, feed efficiency, and economic efficiency without compromising the animals' well-being (Abdou *et al.*, 2024).

Consequently, microalgae supplements have now emerged as a good substitute for fatty fish, especially when it comes to n3 and n6 fatty acids, to boost animal productivity. Additionally, it has been noted that adding microalgae to animal diets enhanced meat quality, immunity, growth performance, and antioxidant indices in ruminants, chickens, rats, rabbits, and pigs (Madeira *et al.*, 2017; Abdelnour *et al.,* 2019; Pavan -Kumar and Sibi, 2020). Algae biomass seems like a highly promising option for animal feed. Research indicates that almost 30% of the world's algal productivity is consumed by animals (Abd El- Hack *et al*., 2024). Microalgae have become important functional feed additives because of their excellent nutritional value and functional chemical content (Mueller *et al*., 2024).

Animals fed *Nannochloropsis oculata* may produce compounds enhanced by this type of n-3 PUFA. However, monogastric mammals like pigs, rabbits, and chickens have difficulty digesting microalgae due to their resistant cell wall (Lum *et al*., 2013). Therefore, it might help to enhance reproductive and productive performance. According to Hibberd (2008), *N. gaditana, N. salina, N. limnetica, N. granulata, N. oceanica, and N. oculata* are the six different species of *Nannochloropsis*. Numerous *in-vitro* and *in-vivo* studies have demonstrated the beneficial effects of *N. oculata* on palatability and lack of toxicity (Kafaie *et al*., 2012), immunity (Derner *et al*., 2006; Colla *et al.*, 2007), anti-inflammatory and anti-cancer (Sanjeewa *et al*., 2016), antioxidant effects (Elsheikh *et al*., 2018) and ease of digesting (Kholif *et al*., 2020).

Alternatively, the little tree known as *Moringa (Moringa oleifera Lam),* which originated in the sub-Himalayan mountains of North West India, is now found natively in many parts of Africa, Arabia, South East Asia, the Pacific and Caribbean Islands, and South America. *Moringa oleifera* commonly referred to as the "tree of life," is an Indian plant with numerous health advantages. It is utilized in the nourishment of humans and animals for industrial purposes, to treat certain illnesses, and to purify water (Foidl *et al.*, 2001).

According to certain research by Sarwatt *et al*. (2002) and Makker and Becher (1997), cattle that were fed *Moringa oleifera* as a rich source of protein grew greater body weight than those not provided with food. Shea (2023) discovered that while there is not much to no change in the quantity of *Moringa* leaves consumed when they are fed fresh or dried, dried leaves retain all of their nutritional content even after being preserved for long periods.

Researchers worldwide have been concentrating on *Moringa oleifera.* Vitamins A, C , and α-Tocopherol, carotenoids, phenols (gallic,ellagic, ferulic acid, and chlorogenic), flavonoids (kaempferol, quercetin, and rutin), and other natural antioxidant molecules are all present in *Moringa oleifera* leaf extract (Safwat *et al.,* 2024). Consequently, this study's goal was to find out the impact of microalgae *Nannochloropsis oculata* and/or *Moringa oleifera* leaves as natural antioxidants on the productive performance and blood constituents in Hi-Plus growing rabbits under North Sinai conditions. Such effect would be revealed by certain biological manifestations that are measurable through some physiological and histological parameters.

MATERIALS AND METHODS

The current investigation was conducted from September 2021 to November 2021 at a private rabbit farm (Latitude 31° 29′ N; Longitude 32° 34′ E), North Sinai Governorate, in collaboration with the Egyptian Center of Excellence for Bio-Saline Agriculture, Systel Telecom Company, the Department of Animal and Poultry Physiology, Desert Research Center, Ministry of Agriculture, Cairo, Egypt. The laboratory analysis was conducted at the Animal and Poultry Physiology Laboratory, which belongs to the Animal and Poultry Production Division, Desert Research Center, Ministry of Agriculture and Reclamation, Cairo, Egypt. Histo-morphological examinations carried out at Al-Azhar University's Faculty of Science (Girls).

Experimental Design:

Sixty weaned Hi-Plus rabbits, weighing 686.77 ± 15.64 g at 5 weeks of age, were divided into four identical treatments (15 rabbits per treatment). Three equal replicates of each treatment were created at random (5 rabbits per replicate). The 1st treatment (C) acted as a control and was given the basal meal devoid of any additives. The 2nd treatment (NCO) rabbits were fed the basic diet + 20 g microalgae *Nannochloropsis oculata* / kg diet (2.0 %) (El-Sayed *et al*., 2022). The 3rd treatment (MO) rabbits were given the basal diet + 50 g *Moringa oleifera* / kg diet (5.0 %) (Abubakar *et al*., 2021). The 4th treatment (NCO+MO) rabbits were fed the basal diet + 20 g microalgae *Nannochloropsis oculata* + 50 g *Moringa oleifera* / kg diet. Animals were slaughtered at the 12th week of age the blood was taken for physiological parameters and the liver for histological studies.

Experimental Animals, Feeding and Maintenance:

Hi-Plus rabbits were kept in the same sanitary and administrative settings. The rabbits were vaccinated, treated with antibiotics, and checked for illnesses. NRC (1994) claims that rabbits were kept in cages made of wire until they were 12 weeks old (the end of the experiment), clean fresh water was given, and they were fed on a commercial concentrate pelleted diet of growing rabbits that contained 17.0% crude protein, 13.0% crude fiber, 2.0% fat, 0.3% minerals combination and 2700 kcal/kg energy that was digested (Table 1). The light period lasted 16 hours, and the average interior temperature and relative humidity were maintained at 26.1 °C and 50.2%, respectively, during the trial period.

As per the guidelines for the use of animals provided by the Institute of Animal Ethics Committee (2010/63/EU of the European Parliament and of the Council of September 22, 2010), experiments were conducted.

Table 1: Composition and chemical analysis of basal diet (% dry matter basis)

Ingredients	**Basal diet**	***Nannochloropsis oculata* (2%)**	***Moringa oleifera* (5%)**
Nannochloropsis oculata	0.0	2.0	0.0
Moringa oleifera	0.0	0.0	5.0
Soybean meal (48% CP)	9.0	6.7	6.2
Barley	11.0	11.0	11.0
Wheat bran	14.0	14.0	14.0
Corn	19.0	19.3	17.0
Clover hay	29.0	29.0	29.0
Fennel hay	13.0	13.0	12.8
Molasses	3.00	3.00	3.00
Di- calcium phosphate	1.00	1.00	1.00
DL-Methionine	0.40	0.40	0.40
Sodium chloride	0.30	0.30	0.30
Vit. Min. premix*	0.30	0.30	0.30
Total	100	100	100
Chemical analysis			
Dry matter (DM)	87.88	87.88	87.88
Organic matter (OM)	90.88	90.88	90.88
Crude protein (CP)	17.56	17.56	17.53
Crude fiber (CF)	13.26	13.26	13.26
Ether extract (EE)	1.980	1.980	1.980
Nitrogen free extract (NFE)	58.08	58.08	58.08
Ash	9.120	9.120	9.120
Digestible energy (Kcal/Kg DM)	2700	2700	2700

*Vitamins and mineral mixture supplied per kg of diet: Vitamin A 10,000 IU, Vitamin D3,1,800 UI; Vitamin E, 15 mg; vitamin K3, 4.5 mg; Vitamin B1, 0.5 mg; Vitamin B2, 4 mg; Vitamin B12, 0.001 mg; Folic acid, 0.1 mg; Pantothenic acid, 7 mg; Nicotinic acid, 20 mg; I, 1 mg; Mn, 60 mg; Cu, 5.5 mg, Zn, 75 mg; Fe, 40 mg; Co, 0.3 mg; Se, 100 mg, Robenidine, 52.8 mg.

The Microalga, *Nannochloropsis oculata* & *Moringa oleifera*:

The National Research Center (NRC), Giza, Egypt's Biotechnology Microalgae Culture Unit produced and helpfully supplied the microalga *Nannochloropsis oculata* utilized in this investigation.After being collected, the microalgae were centrifuged and stored until the completion of the culture time at 4 ^{o}C in the refrigerator. The National Research Center (NRC) in Giza, Egypt, duly prepared and supplied the *Moringa oleifera*. The chemical composition of microalgae *Nannochloropsis oculata* and *Moringa oleifera* extracts were identified using gas chromatography mass at complex laboratories of National Research Centre, Dokki, Giza, Egypt.

The components of microalgae *Nannochloropsis oculata* and *Moringa oleifera* extracts are identified and quantified in Table (2).

Table 2: The Chemical composition, mineral and amino acids profile of *Nannochloropsis oculate* and *Moringa oleifera* constituents by GC mass.

Items	***Nannochloropsis oculate***	***Moringa oleifera***
Chemical composition (g/100g)		
Moisture	7.15	6.20
Crude protein	55.78	27.60
Fat	6.61	17.66
Ash	12.29	11.23
Total carbohydrates	18.17	38.57
Minerals profile (mg/100g)		
Fe	29.35	29.20
Zn	1.02	4.73
Sodium	1862.70	30.17
Calcium	229.00	2210.10
Potassium	798.00	1941.53
Magnesium	173.00	411.00
Amino acids profile (mg/g)		
Methionine	69.52	9.98
Cystine	17.30	20.66
Phenylanlanine	16.24	46.27
Lysine	15.20	33.19
Isoleucine	55.95	44.49
Leucine	65.11	53.88
Aspartic acid	30.16	62.77
Glutamic acid	15.07	151.43
Histidine	13.22	20.59
Tyrosine	87.69	23.87
Threonine	39.21	32.88
Valine	50.36	31.59
Serine	11.64	43.37
Glycine	9.98	50.95
Proline	31.52	24.58
Alanine	20.24	32.93
Arginine	8.56	82.97

Experimental Measurements:

Productive Performance:

Individual body weight was weekly recorded to the closest (0.1 g) from the 5th week (initial body weight) to the 12th week (final body weight) of age. The calculation of body weight gain (BWG) involved deducting the average initial body weight of a certain period from the average final body weight at the same period as follows: Body weight gain = W2 – W1

Where: W1 = body weight at the start of a period.

W2 = body weight at the end of the same period.

The amounts of feed intake were weekly recorded for each treatment by differences between the total amounts of diet offered to rabbits in treatment at the beginning of the week and the remaining feed amounts at the end of the same week. Feed intake per rabbit was calculated throughout the experimental period by dividing the total amount of feed intake by the total number of rabbits in a treatment. A gram of feed was needed to create one gram of body weight growth, which is the feed conversion ratio.

FCR (%) is equal to the average feed intake divided by the body weight growth of each rabbit.

Blood Parameters:

Blood samples were taken biweekly (4 times) throughout the experiment period into tubes that contained EDTA as an anticoagulant (5 rabbits/treatment). Following centrifugation at 5000 g for 15 minutes, plasma was collected and kept at -20°C for further analysis. The bioanalysis of plasma was carried out for quantitative determination of blood parameters by spectrophotometer.

Blood Metabolites:

Using commercial kits (Spectrum business, Egypt), the following parameters were measured in plasma: total proteins (TP), albumin (ALB), glucose (GLU), cholesterol (CHOL), high-density lipoprotein (HDL), triglycerides (TG), creatinine (CR), and urea (UR) according to Finley *et al.* (1978).

The levels of aspartate aminotransferase (AST) and alanine aminotransferases (ALT) were estimated according to Henry (1964). Howe (1921) used the formula Globulin = (total Protein - albumin) to determine the concentration of Globulin (GLO).

According to Koracevic *et al.* (2001), commercial kits (Biodiagnostic Research, Egypt) were used for the colorimetric measurement of glutathione peroxidase (GSX), malondialdehyde (MDA), and total antioxidant capacity (TAC).

Blood Hormones:

Utilizing ELISA kits (Monobind, USA), the hormones triiodothyronine (T3), thyroxine (T4), growth (GH), and insulin were examined in accordance with Wheeler *et al.* (1994).

Statistical Analysis:

The General Linear Model Procedure was applied to analyze the data using the least squares analysis of variance **(SAS, 2004).** The design was one-way analysis and the model was as follows:

$Y_{ij} = \mu + Tr_i + e_{ij}$ Where,

Y_{ij} = any observation of j^{th} rabbit within i^{th} treatment.

μ = overall mean.

Tr_i = effect of i^{th} treatment (i: 1-4).

e_{ij} = experimental error.

Duncan Multiple Range Test **(**Duncan, 1955) was employed to ascertain the degree of significance between means.

Histological Investigations of The Liver:

For light microscopic investigations, after blood collection, the animals were rapidly sacrificed to dissect out liver samples. The liver cut into small pieces was kept in formalin (10%) solution at ph. 7.0 for 5 days, followed by washing, dehydrating in ascending grades of ethyl alcohol, cleared in xylene Then embedded in molten paraffin wax at 58–62 degrees Celsius. The Leica rotatory microtome (RM 20352035; Leica Microsystems, Wetzlar, Germany) was then used to segment the paraffin blocks at a thickness of 5 µm (Suvarna *et al*., 2013). Then the sections were stained with hematoxylin and eosin (H&E) (Cook and Stirlin, 1994). The selected sections were examined with the light microscope and photographed with U3cmos14000Kpa (usb2.0) model of the camera employed to take the photos for histological examination.

RESULTS AND DISCUSSION

Growth Performance:

Live Body Weight:

Results of body weight (BW) at 6, 7, 8, 9, 10, 11, and 12 weeks as impacted by *Nannochloropsis oculata* algae (NCO) and *Moringa oleifera* (MO) supplementation in the rabbit diets were illustrated in (Fig. 1).

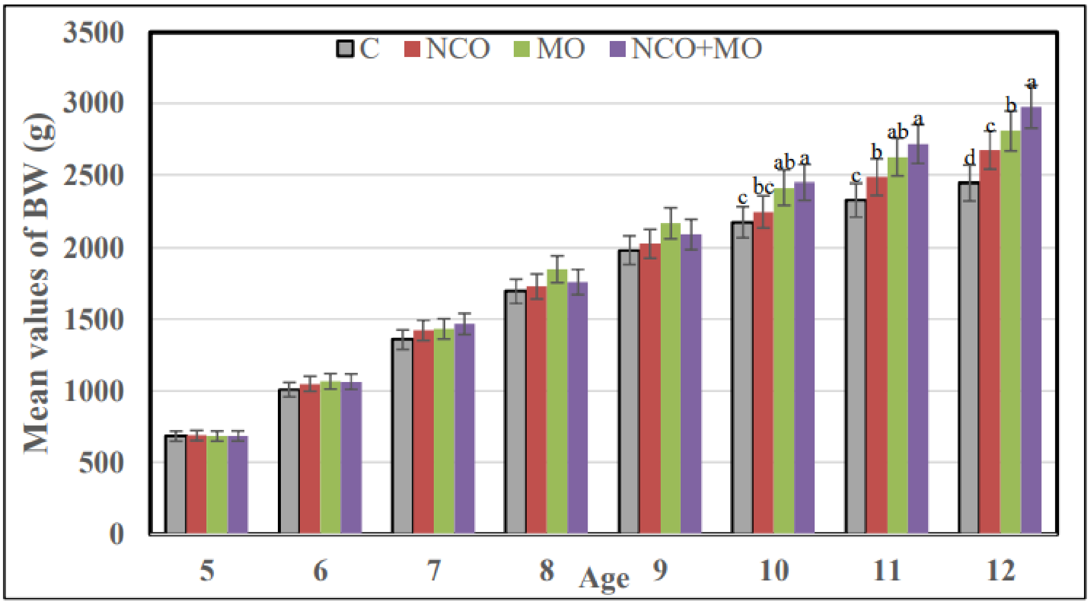

Fig. 1: Effect of *Nannochloropsis oculata* algae and/or *Moringa oleifera* on body weight (BW, g) of Hi-Plus growing rabbits during the period from 5th to 12th weeks of age.
Data represent means± SE (n=15). C= control group, NCO= rabbits received 2 % *Nannochloropsis oculata* algae, MO= rabbits received 5 % *Moringa oleifera*, NCO+MO= rabbits received 2 % *Nannochloropsis oculata* algae+5 % *Moringa oleifera*. a-d Means bearing different superscripts within the same item are significantly different (P<0.05).

At 8th and 9th weeks, the body weight (BW) of rabbits that were given MO was significantly higher (P<0.05) by 9.04 and 9.46%, respectively, in comparison to the control group (C). The findings showed that rabbits receiving a diet of 2% NCO + 5% MO achieved the greatest BW at (from 10th to 12th) weeks, showing significant differences when compared to the control group. This finding is consistent with that of Odetola *et al.* (2012), who discovered that rabbits fed a diet containing *Moringa oleifera* leaf meal had better final body weights; this could be because the feed had more protein. Additionally, Ibrahim *et al.* (2014) discovered that the final live body weight of rabbits fed diets supplemented with 4 g of *Moringa* peregrine seeds (MPS/kg) was 10.1% greater than that of the control group. Dongnon *et al.* (2012) discovered that young rabbits fed 3% Moringa oleifera were more likely to develop, weigh, and survive than those fed the usual supplement without any additions. This may be attributed to its antibacterial properties and excellent protein digestibility. The crude protein content of *Moringa oleifera* leaf powder (PML) is approximately 33.82% higher than that of commercial feed. This was in agreement with Ghebreselassic *et al.* (2011), who demonstrated that mice given the aqueous leaf extract of *M.* stenopetala experienced a rise in body weight. However, New Zealand White (NZW) rabbits fed *Moringa oleifera* leaf meal (MOLM) at a rate of 12.5, 20, 40 and 60 % showed non-significant increases in ultimate body weight, daily weight gain, and relative growth rate (Ahmed, 2017; Awed, 2019).

However, Abd EL-Hack *et al.* (2024) observed that adding microalgae *spirulina* (SP) to quail diets enhanced live weight without having any negative effects. The high levels of protein, alpha-linolenic acid, arachidonic acid, ω3, ω6, ω9, and the sum of ω in microalgae may be the cause of this improvement in BW. Additionally, because SP contains high-quality protein, vital fatty acids, minerals, vitamins, carotenoids, and other components that boost maternal health, Abdou (2024) discovered that feeding SP in doe rabbits may increase body weight (BW).

Body Weight Gain:

The rabbits who received 5% MO had considerably (P<0.05) higher body weight gain (BWG) at 7-8 weeks than the rabbits in the control group, NCO, and NCO+MO, respectively. However, compared to the rabbits in the control group, the rabbits fed NCO+MO showed the significantly highest body weight gain at 9–10, 10–11, and 11–12 weeks in addition to the highest total weight gain (TWG) (Fig. 2).

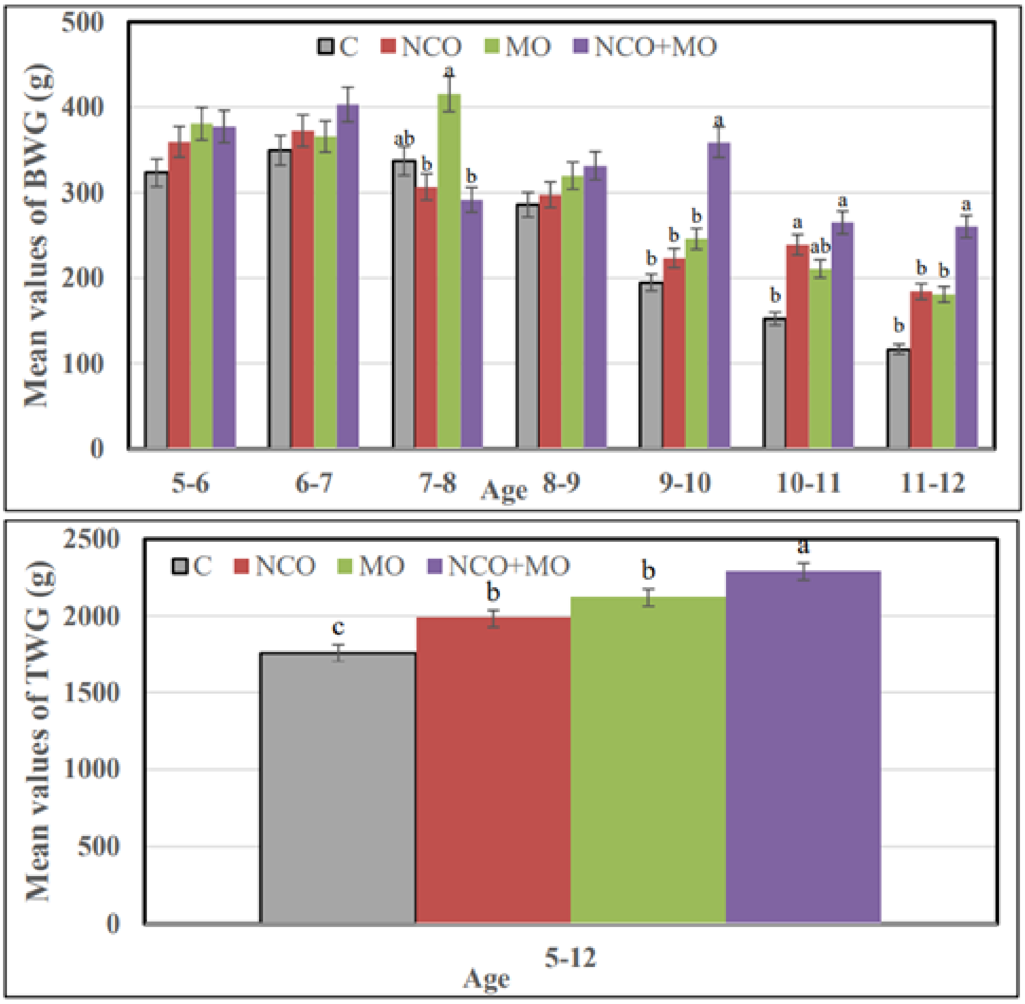

Fig.2: Effect of *Nannochloropsis oculata* algae and /or *Moringa oleifera* on body weight gain (BWG, g) of Hi-Plus growing rabbits during the period from 5th to 12th weeks of age.

Data represent means± SE (n=15). C= control group, NCO= rabbits received 2 % *Nannochloropsis oculata* algae, MO= rabbits received 5 % *Moringa oleifera*, NCO+MO= rabbits received 2 % *Nannochloropsis oculata* algae+5 % *Moringa oleifera*.

BWG=body weight gain, TWG=total weight gain.

$^{a-e}$ Means bearing different superscripts within the same item are significantly different (P<0.05).

These findings correspond with those of Yan *et al.* (2012), who discovered that supplementing growing pigs *Chlorella vulgaris* increased weight gain when comparing the supplemented groups to the control. The growth stimulation may be caused by bioactive substances such as the S-nucleotide adenosyl peptide complex, polysaccharides, and phenolic compounds abundant in the microalga (Kay and Barton, 1991).

According to Sikiru *et al.* (2021) and El Bahr *et a*l. (2020), the increase in feed utilization efficiency and weight gain bolstered earlier findings that suggested the microalgal is a growth promoter. According to El Hawy *et al.* (2022), rabbit bucks supplemented with 0.50% or 1 %microalgae meal illustrated rising in live body weight, total weight gain, and growth rate when compared to the control group. However, as stated by Mankga *et al.* (2022), Imtiaz *et al*. (2023), and Maqsood *et al.* (2024), the highest concentrations of *Moringa Oleifera* leaf meal (MOLM) did not inhibit the rabbits' body weight gain. Numerous studies (Omara *et al.*, 2018; Jiwuba and Ogbuewu, 2019; Selim *et al.*, 2021) reported similar results when using 10, 20, and 30% MOLM of the rabbit diets. This is because MOLM contains a significant amount of bioactive compounds, minerals (calcium, magnesium, zinc, copper, iron, selenium, potassium, and phosphorus), amino acids (lysine and methionine), carotene, and vitamins (A, B, C, and K) that are known to promote animal growth (El Badawi *et al.*, 2016; Sebola *et al.*, 2017; El-Desoky *et al.*, 2018; Mahfuz and Piao., 2019; Rashad, 2020) furthermore, the bioactive compounds found in *Moringa oleifera* may be accountable for the higher live weight gain (Cui *et al.*, 2018; Sun *et al.*, 2018).

Additionally, Nuhu (2010), Ufele *et al.* (2013), and Abubakar *et al.* (2015) observed that rabbits' daily live weight gain increased as the amount of MOLM in their diets increased, and that the rabbits' high weight gain was caused by the high protein content of the MOLM diet, which may be due to the diet's balance of amino acids. Additionally, the rabbits' daily weight gain is increased by using *Moringa oleifera* leaf meal (MOLM), which can replace a specific percentage of soya bean meal in the diet without impairing the rabbits' performance (Alemede *et al.*, 2014).

Feed Intake:

Feed intake (FI) at various weeks (5-6, 6-7, 7-8, 8-9, 9-10, 10-11, 11-12 weeks, and total feed intake) was lower (P<0.05) in the rabbits fed NCO, MO, and NCO+MO than in the rabbits of the control group (Fig. 3). The differences in FI between the experimental groups were significant, and the rabbits fed NCO+MO possessed the lowest TFI when compared to the rabbits of the other groups.

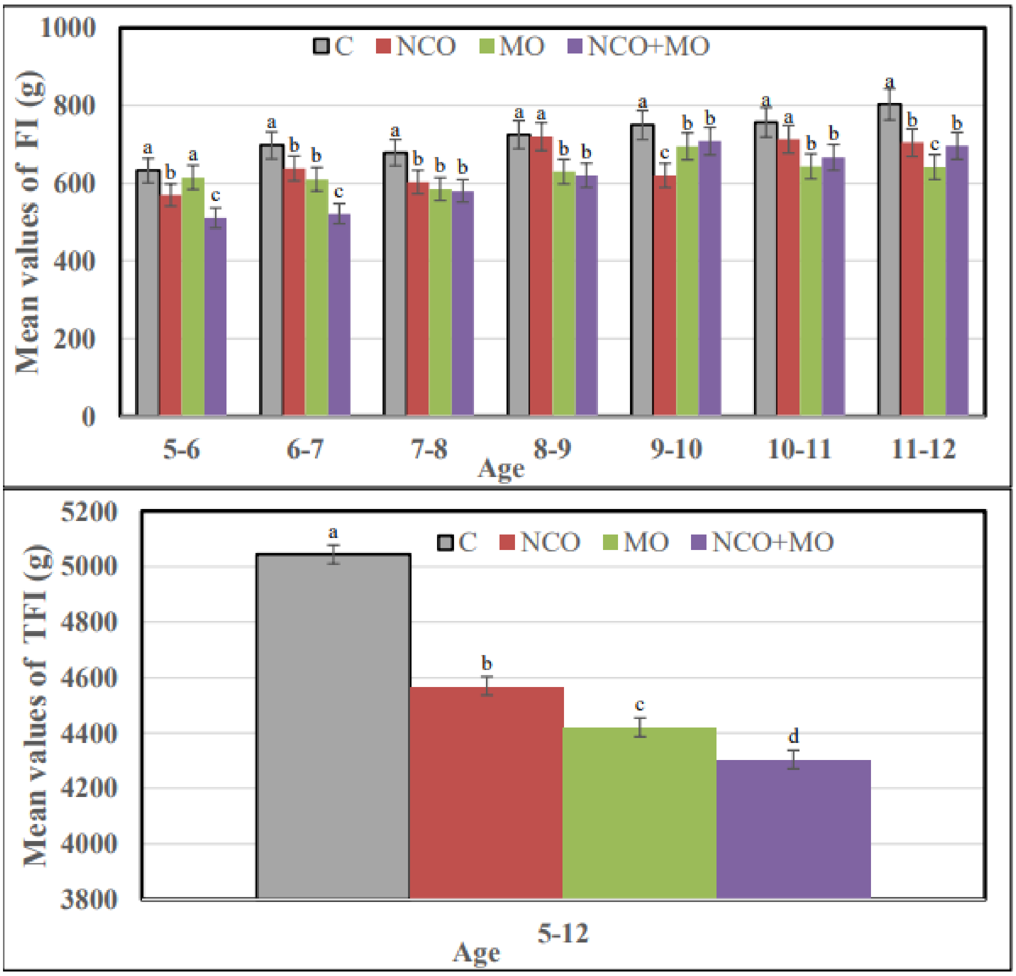

Fig. 3: Effect of *Nannochloropsis oculata* algae and/or *Moringa oleifera* on feed intake (FI, g) of Hi-Plus growing rabbits during the period from 5th to 12th weeks of age.
Data represent means± SE (n=15).
C= control group, NCO= rabbits received 2 % *Nannochloropsis oculata* algae, MO= rabbits received 5 % *Moringa oleifera*, NCO+MO= rabbits received 2 % *Nannochloropsis oculata* algae+5 % *Moringa oleifera*.
$^{a-d}$ Means bearing different superscripts within the same item are significantly different (P<0.05).

Animal physiology is improved by feeding small amounts of microalgae meal, which improves organ histology, feed conversion ratio, weight gain, immunological response, disease resistance, and gut function (Bonos *et al.*, 2016; Mirzaie *et al.*, 2018). According to Kurd and Samavati (2015), microalgae are rich in compounds that have biological activity and act as antioxidants. Important components found in microalgae include a high protein content (60–70% dry matter), minerals (Ca, Cr, K, Mg, Cu, Fe, Na, P, Mn, Zn, and Se), amino acids, vitamins (B12 and β-carotene), and polyunsaturated fatty acids (γ-linolenic acid). (Hoseini *et al.*, 2013).

The findings of this investigation are comparable to those of Ibrahim *et al.* (2014), who found that daily feed consumption increased as Moringa peregrine seed meal increased when compared to participants in the control group.

Conversely, Mankga (2022); Ahmed (2017), and Badawi *et al.* (2017) found that when a certain level of MOLM inclusion rose, feed intake decreased. This may be explained by the tannins and saponins found in *Moringa oleifera* leaf meal (MOLM), which can change feed intake by influencing the taste, palatability, and functional qualities of diets (Stevens *et al.*, 2015; Disetlhe *et al.*, 2018).

1.4. Feed Conversion Ratio:

The NCO+MO group demonstrated a significant improvement in feed conversion (FC) at 5–6, 9-10, 10-11, 11-12, and TFC (12 weeks) contrasted with the control group (Fig. 4). However, rabbits fed NCO showed significantly improved FC at 10-11, 11-12, and 5-12 (TFC) compared to the control group. Meanwhile, rabbits fed MO illustrated improved ($P<0.05$) FC at 10-11, 11-12, and 5-12 compared to the control group.

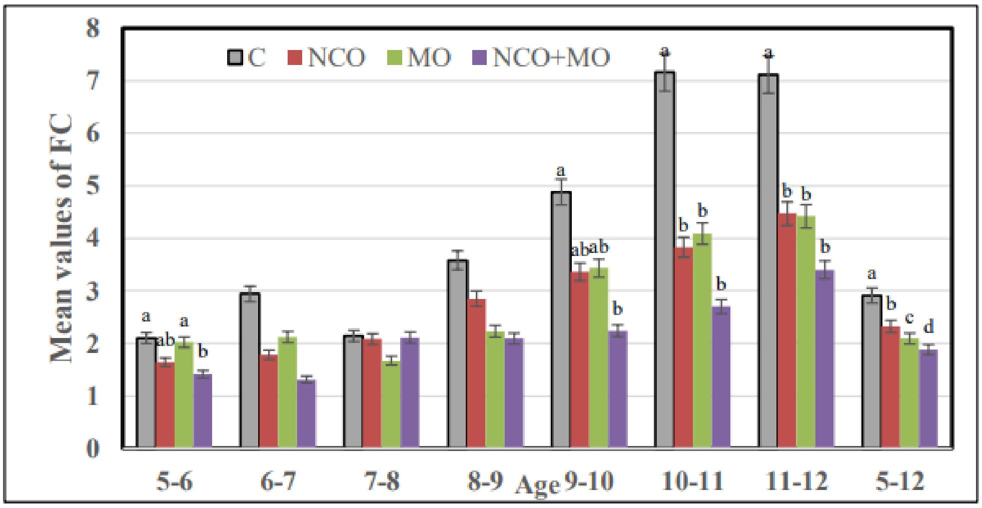

Fig. 4: Effect of *Nannochloropsis oculata* algae and/or *Moringa oleifera* on feed conversion (FC) ratio of Hi-Plus growing rabbits during the period from 5th to 12th weeks of age**.**

Data represent means± SE (n=15).

C= control group, NCO= rabbits received 2 % *Nannochloropsis oculata* algae, MO= rabbits received 5 % *Moringa oleifera*, NCO+MO= rabbits received 2 % *Nannochloropsis oculata* algae+5 % *Moringa oleifera*.

FC=feed conversion, TFC=total feed conversion

a-c Means bearing different superscripts within the same item are significantly different ($P<0.05$).

The outcomes are consistent with those of Badawi *et al.* (2017) and Frederick (2010), who demonstrated that the inclusion of MOLM improves the FCR when compared to the control group. Additionally, Ibrahim *et al.* (2014) and Eldeeb *et al.* (2014) reported that rabbits fed 4 and 8 g of *Moringa* seeds or leaves had an improved feed conversion ratio, which they attributed to the increase in body weight gain, the improvement in nutrient digestibility of diets, and the high protein content of the leaves, which is abundant in every necessary amino acid and can support livestock production (Odetola *et al.*, 2012; Omara *et al.*, 2017; Jiwuba and Ogbuewu, 2019).

However, the microalgae results are consistent with those of Ragab *et al.* (2019) and Abd El-Hamid *et al.* (2022), who credited the rise in FCR to the effectiveness of dietary supplements containing a high and plentiful amount of microalgae protein *Nanochloropsis oculata* (Lum *et al.*, 2013; Cavonius, 2016). However, Hamza (2019) demonstrated the effect of *Spirulina* on feed conversion ratio significantly improved as a result of improved daily body weight gain.

Hematological Parameters:

Total protein and Its Fractions:

The amounts of globulin and total protein in plasma were significantly ($P<0.05$)

higher in the NCO+MO-fed rabbits than in the control group. However, there was a non-significant difference in albumin concentration (Fig. 5).

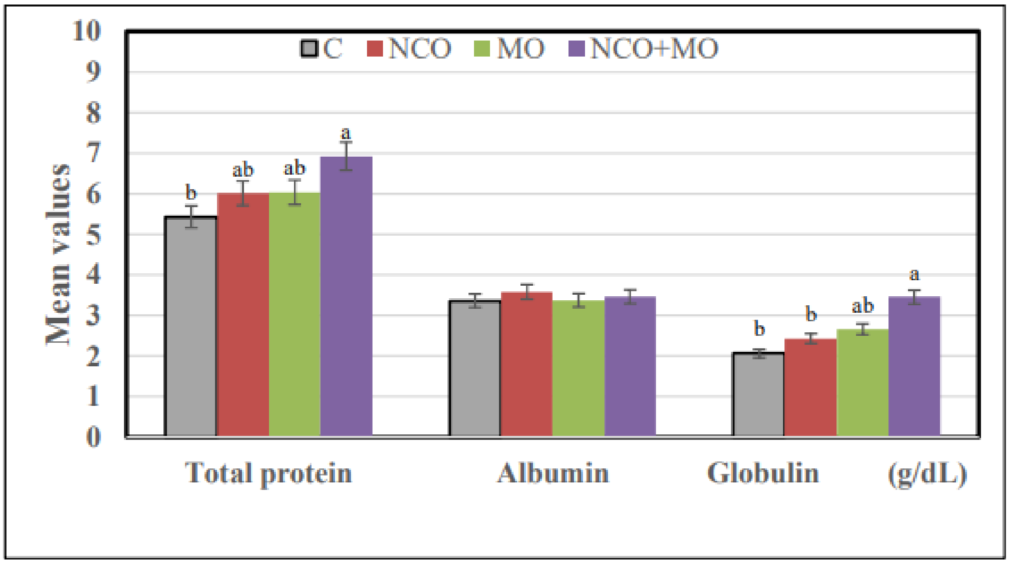

Fig. 5: Effect of *Nannochloropsis oculata* algae and/or *Moringa oleifera* on blood total protein and its fractions of Hi-Plus growing rabbits.
Data represent means± SE (n=15).
C= control group, NCO= rabbits received 2 % *Nannochloropsis oculata* algae, MO= rabbits received 5 % *Moringa oleifera*, NCO+MO= rabbits received 2 % *Nannochloropsis oculata* algae+5 % *Moringa oleifera*.
[a-b] Means bearing different superscripts within the same item are significantly different ($P<0.05$).

Better immune and liver functioning were shown by higher serum levels of globulin and total protein and reduced ALT and AST levels in the groups fed MOL (Igbinaduwa *et al.*, 2016 and Selim *et al.*, 2021). Because *Moringa oleifera* extract contains bioactive flavonoids like quercetin, it has been shown to have hepatoprotective properties (Misra *et al.*, 2014). According to studies by Sun *et al.* (2018); Salem *et al.* (2020) and El-Kashef (2022) adding MOL to the diet improved the blood parameters of developing rabbits. Due to its antioxidant components, including vitamin C, *Moringa oleifera* is recognized as a powerful antioxidant feed ingredient that may be essential for improving the health of developing rabbits (Mahfuz *et al.*, 2019; Su *et al.*, 2020).

The higher values of serum total protein and globulin concentrations in the rabbits fed microalgae might be because microalgae *N. oculate* contains high-quality and abundant protein and amino acids. (Abd Elhamid *et al.*, 2022). Additionally, Emam *et al.* (2024) revealed that the rise in total protein levels in hens fed *N. oculata* was accompanied by an increase in serum globulin concentration; this could be associated with the immune system's development through the production of immune globulin, which makes it a reliable indication of immunity response.

Blood Metabolites:

The results illustrated that cholesterol and LDL levels were significantly reduced ($P<0.05$) in the rabbits fed NCO, MO, and NCO+MO compared to the control group (Fig. 6).

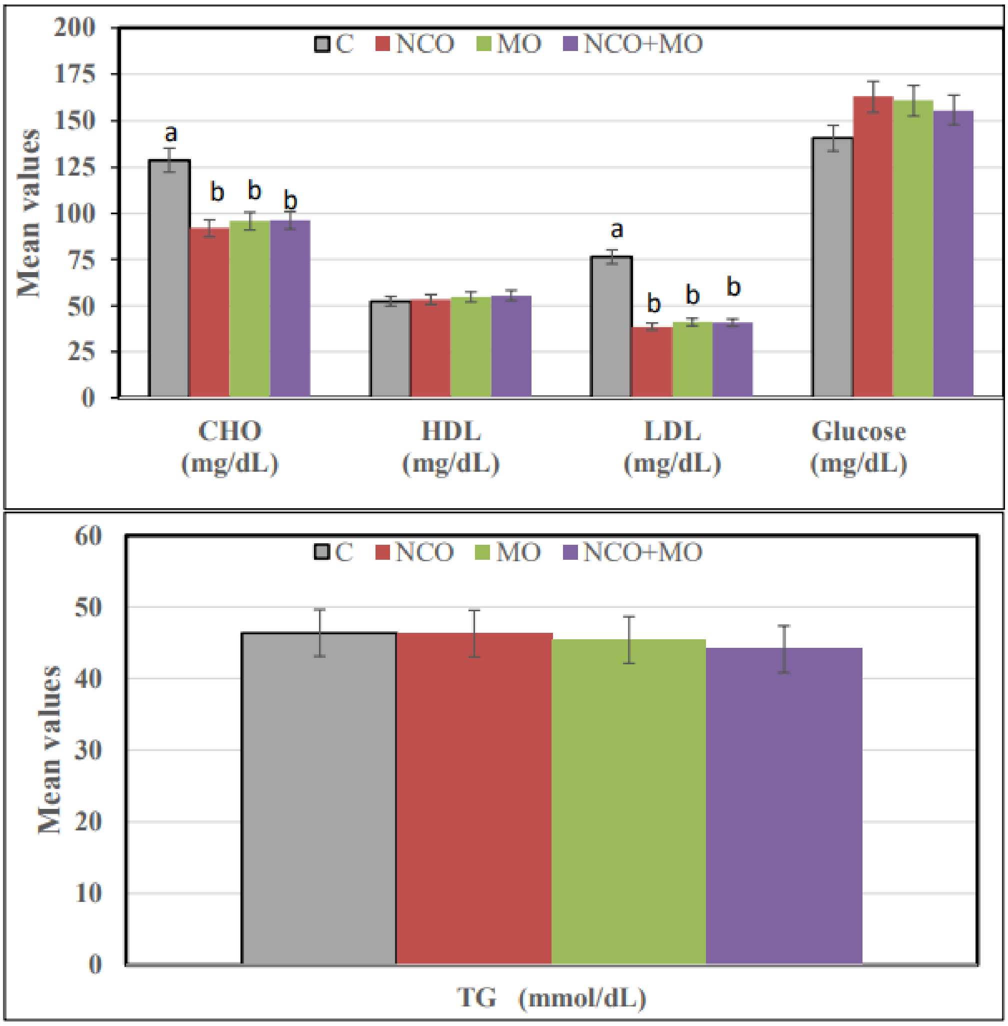

Fig. 6: Effect of *Nannochloropsis oculata* algae and/or *Moringa oleifera* on blood metabolites parameters of Hi-Plus growing rabbits.
Data represent means± SE (n=15).
C= control group, NCO= rabbits received 2 % *Nannochloropsis oculata* algae, MO= rabbits received 5 % *Moringa oleifera*, NCO+MO= rabbits received 2 % *Nannochloropsis oculata* algae+5 % *Moringa oleifera*.
HDL=high density lipoprotein, LDL=low density lipoprotein, TG=tri-glycerides.
[a-b] Means bearing different superscripts within the same item are significantly different (P<0.05).

No significant differences were noted in HDL, TG, and glucose levels among treatments. Moreover, glucose concentration was insignificantly increased in the rabbits fed NCO, MO, and NCO+MO compared to rabbits in the control group.

The increased serum glucose and decreased cholesterol levels and triglycerides by increasing MOLM intake means MOLM may have hypolipidemic effects (Ahemen *et al.*, 2013). These findings increased as the intention to minimize triglyceride and increased good cholesterol levels demonstrated that the liver was functioning effectively. They discovered that the extract from *moringa* leaves might contain hypolipidemic substances. As a result, there was less lipid mobilization and a lower cholesterol level. A key enzyme in the manufacture of cholesterol, hydroxymethyl glutaryl CoA reductase, may have inhibitory impacts on the liver's cholesterol levels. Also, Jain *et al.* (2010); Ibrahim *et al.* (2014), and Salem *et al.* (2020) revealed that the administration of *Moringa peregrine* seeds (MPS) reduced the concentrations of glucose, cholesterol, LDL, HDL, and triglycerides. This implies that MPS could influence peripheral tissues in a manner akin to that of insulin, either by encouraging glucose uptake and metabolism or by inhibiting gluconeogenesis. MPS may have the effect of increasing tissue consumption of glucose (Jabeen et al., 2008 and Luqman et al., 2012) by suppressing hepatic gluconeogenesis or absorption of glucose into the muscles and adipose tissues (Kamanyi *et al.*, 1994; Desta *et al.*, 2011). According to reports, B-sitosterol, the bioactive phytoconstituent extracted from *Moringa oleifera*, is thought to reduce plasma concentrations of LDL, which is the method by which cholesterol is reduced (Saluja *et al*., 1978; Kane and Malloy, 1982; Ghasi *et al*., 2000).

Furthermore, *M. oleifera* leaves contain polyphenols that inhibit pancreatic lipase activity, which prevents the absorption of triacylglycerol, according to Akip *et al*. (2024). They also inhibit the generation of intestinal lipoproteins, cholesterol esterification, and β-apoprotein synthesis in the intestinal mucosa. Furthermore, sitosterol, which is found in *M. oleifera* leaves, may lower cholesterol levels by encouraging fecal excretion (in the form of steroids) and lowering the absorption of endogenous cholesterol (Bidura *et al.,* 2020). *M. oleifera* may also activate lipoprotein lipase, which increases lecithin cholesterol acyltransferase activity in HDL and encourages cholesterol absorption from blood vessels. This could lead to lower cholesterol levels through better scavenging and excretion (Gasmalbari *et al.*, 2020).

Pankaj and Varma (2013) connected the hypolipidemic effect of *S. platensis* to the presence of g-linoleic acid, which contributes to the breakdown of TG and the catabolism of cholesterol (Froyen, 2022). However, comparable findings demonstrated that the meal supplemented with *N. oculata* in rabbits had no impact on serum glucose levels (Barbara Howe, 2012). Additionally, goats' lipid metabolites were unaffected by supplementing their diet with varying amounts of microalgae *N. oculata* (5 or 10 g/h/d) (Kholif *et al.*, 2020).

According to El-Hawy *et al.* (2022), buck rabbits fed 0.5 and 1.0% microalgae meal *(Nannochloropsis oculata*) had noticeably higher serum glucose levels than bucks in the control group. In the meantime, the total cholesterol concentration significantly dropped when compared to the control group's dollars. The elevated glucose concentrations observed could be attributed to the microalgae meal's high protein, essential amino acid, vitamin, mineral, phospholipid, and antioxidant contents (Jung *et al.*, 2019).

Liver and Kidney Functions:

Alanine transferase (ALT), aspartic transaminase (AST), and urea (UR) concentrations were reduced (P<0.05) in the rabbits fed NCO, MO, and NCO+MO compared to rabbits in the control group (Fig. 7). However, the results showed that creatinine concentration was insignificantly decreased in the rabbits fed NCO, MO, and NCO+MO compared to rabbits in the control group.

Serum ALT and AST activity decreased, suggesting that supplements containing the microalgae *N. oculata* may protect the liver (Sakr *et al.,* 2019). It's possible that microalgae could prevent liver dysfunction. Possibly as a result of its high concentration of many essential nutrients that offer a variety of health benefits (Abdou *et al.*, 2024).

Nevertheless, Hassanein *et al.* (2014) discovered that adding *Spirulina platensis* and *Chlorella vulgaris* microalgae to rabbits' diets increased their serum creatinine levels as well as the activity of the AST and ALT enzymes. Additionally, Salim *et al.* (2019) showed that adding microalgae extract from *Amphora coffeaeformis* to rabbit drinking water increased the levels of creatinine, ALT, and AST numerically. Furthermore, the rabbit that consumed *Spirulina platensis* microalgae showed a notable shift in the serum concentration activities of AST and ALT, according to El-Ratel (2017). As mentioned by Azab *et al.* (2013), AST and ALT activity serve as markers of hepatotoxicity.

According to Emam *et al.* (2024), the type of algae, the quantity administered, and other factors may all have an impact on how the algae affects liver enzymes. Nonetheless, the inclusion of *N. oculata* algae might be thought to play a useful part in maintaining the hepatocellular membrane's structural integrity. Because of its antioxidant qualities in algae, *N. oculata* supplements may have therapeutic value in protecting the liver from oxidative damage under various stress conditions. When hepatocyte membranes are damaged, several enzymes normally found in the cytosol are released into the bloodstream.

However, the plasma AST of Japanese quail chicks displayed no significant differences among MOLM (0.0, 0.2, 0.4, and 0.6%) treatment diets, according to Kout Elkloub *et al.* (2015). This could indicate that the birds fed diets containing MOLM had normal liver function. However, the reduction in ALT activity shown in birds fed 0.4% and 0.6% MOLM may indicate that MOLM possesses characteristics that potentially impact liver health. According to Olatunji *et al.* (2015), the rabbits were able to tolerate the anti-nutrient in the leaf meal since AST and ALT were not significantly impacted by the varying inclusion of MOLM.

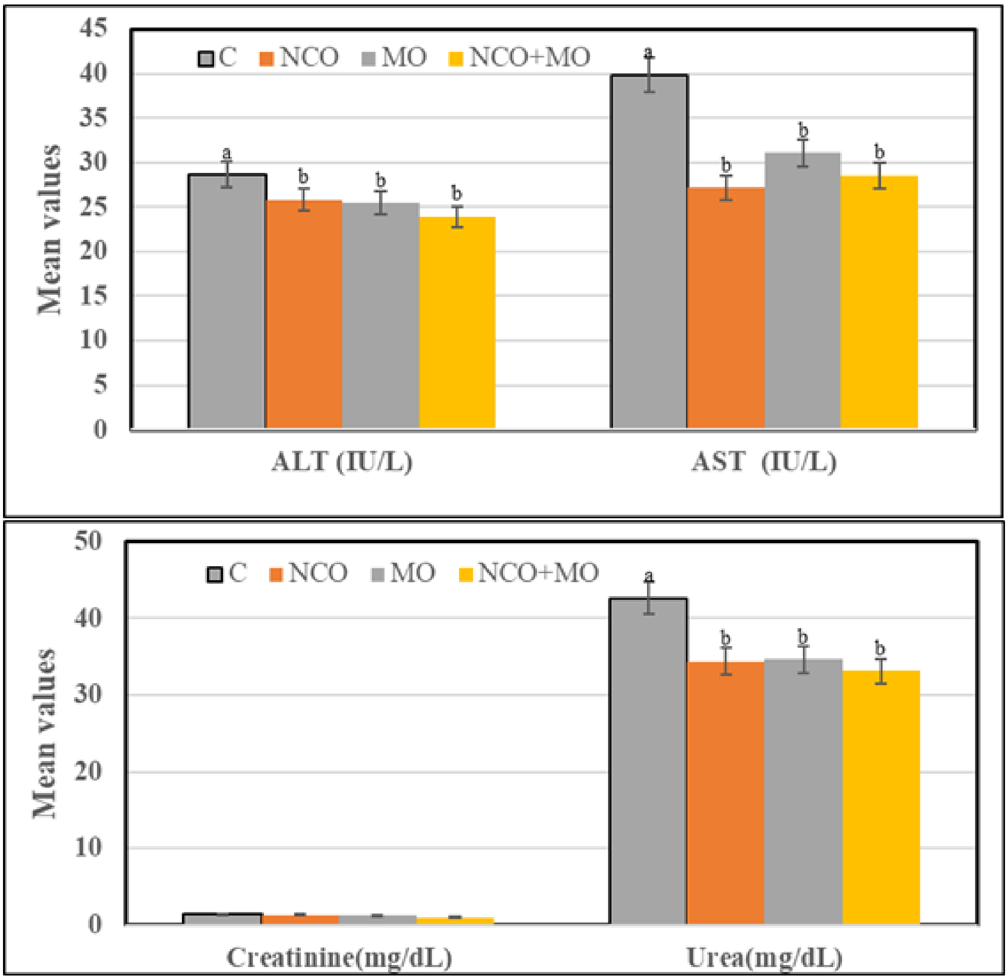

Fig. 7: Effect of *Nannochloropsis oculata* algae and/or *Moringa oleifera* on liver and kidney parameters of Hi-Plus growing rabbits.
Data represent means± SE (n=15).
C= control group, NCO= rabbits received 2 % *Nannochloropsis oculata* algae, MO= rabbits received 5 % *Moringa oleifera*, NCO+MO= rabbits received 2 % *Nannochloropsis oculata* algae+5 % *Moringa oleifera*.
ALT=alanine transaminase, AST=aspartic transaminase.
[a-b] Means bearing different superscripts within the same item are significantly different (P<0.05).

Antioxidative Parameters:

Glutathione peroxidase (GSX) and total antioxidant capacity (TAC) were increased(P<0.05) in rabbits fed NCO, MO, and NCO+MO as compared with the control group. However, malondialdehyde (MDA) was decreased (P<0.05) in rabbits fed NCO, MO, and NCO+MO compared with the control group (Fig. 8).

Nannochloropsis oculata's antioxidant components, including carotenoids, fucoxanthin, astaxanthin, and vitamins, showed the capacity to scavenge free radicals and reduce lipid peroxidation in rabbits (Turrens, 2003).

According to El-Hawy *et al.* (2022), The total antioxidant capacity of *Nannochloropsis oculata* was considerably raised by dietary microalgae at both treatment levels (0.50% and 1.0%). This conclusion is in line with recent research indicating that the richness of natural biological compounds in treated microalgae meals might cause a rise in serum TAC levels. These substances may help reduce oxidative stress by boosting antioxidant enzymes and non-enzymes (Abdelnour *et al.*, 2020a; Abdelnour *et al.*, 2020b and Abd El-Hamid *et al.*, 2022).

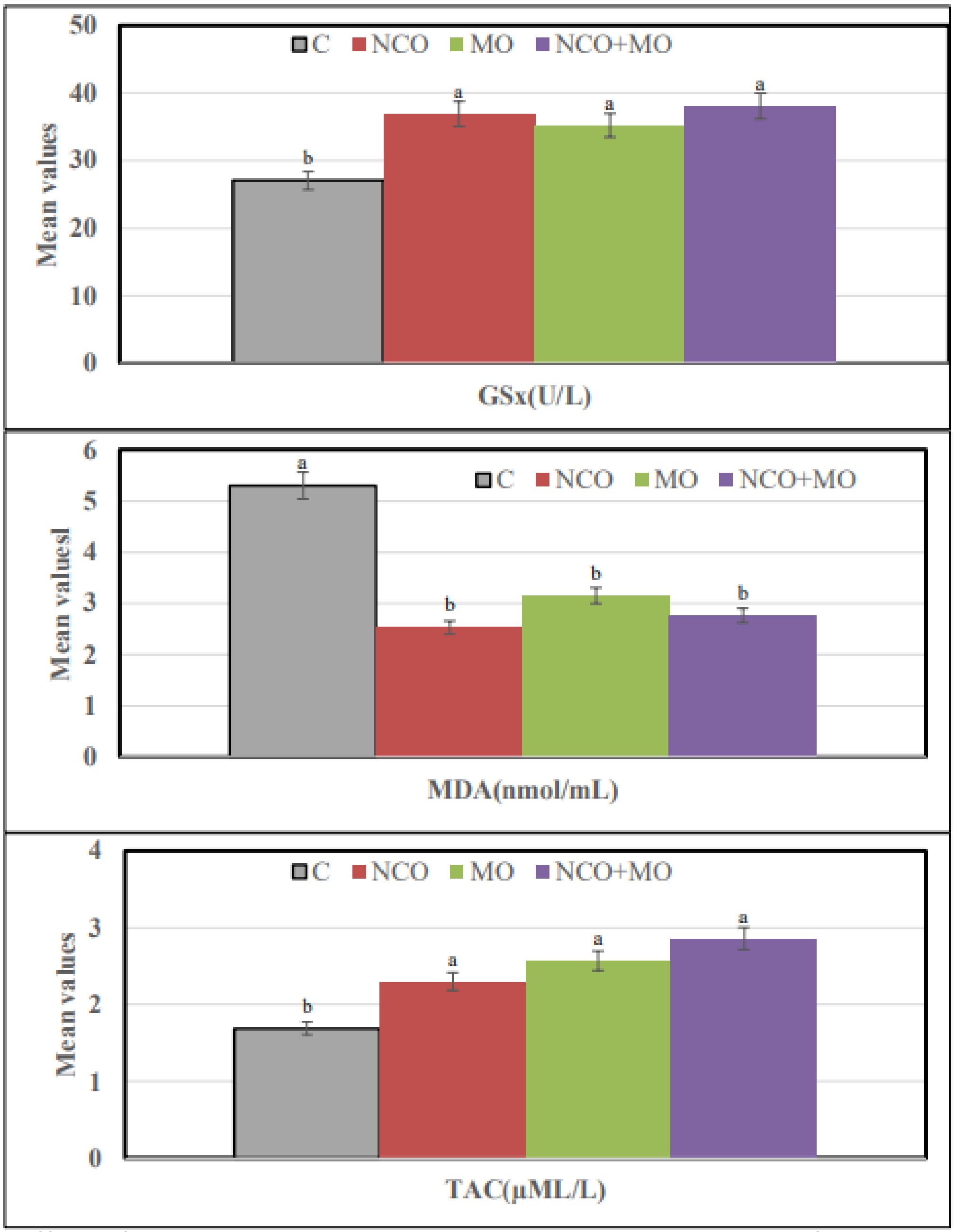

Fig. 8: Effect of *Nannochloropsis oculata* algae and/or *Moringa oleifera* on antioxidant parameters of Hi-Plus growing rabbits.
Data represent means± SE (n=15).
C= control group, NCO= rabbits received 2 % *Nannochloropsis oculata* algae, MO= rabbits received 5 % *Moringa oleifera*, NCO+MO= rabbits received 2 % *Nannochloropsis oculata* algae+5 % *Moringa oleifera*.
GSX=Glutathione peroxidase, MDA= Malondialdehyde, TAC=total antioxidant capacity
[a-b] Means bearing different superscripts within the same item are significantly different (P<0.05).

The body's metabolism normally produces oxygen free radicals in a dynamic balance governed by the antioxidant system. However, an increase in oxygen free radicals or a breakdown of the antioxidant system can upset this equilibrium, leading to lipid peroxidation and oxidative cell injury (Xu and Pan, 2013). The state of the body's antioxidant mechanism, which represents the body's ability to metabolize oxygen free radicals and shield animal tissues from oxidative stress, was therefore made clear by the antioxidant enzymes. Sugar complexes, including glucose, various galactose, mannoses, rhamnose, N-acetylglucosamine, N-acetylgalactosamine, and arabinose residues, are linked to certain biological processes. These are characterized by immunological activity (1, 3-glucan) in all microalga species.

According to Sikiru *et al.* (2021), giving New Zealand White rabbits a supplement of

Chlorella vulgaris dramatically reduced the production of malondialdehyde (MDA), suggesting that the supplement stopped lipid peroxidation. Better feed conversion ratios and weight increase were observed in the treatment groups when compared to the control group because prevention of lipid peroxidation led to improved feed consumption. Apart from the direct effects of these chemicals on growth promotion, the observed growth promotion may also be due to enhanced antioxidant protection, which is a determinant of immunological modulation. Previous research on microalgae indicates that supplementing with it raises the concentration of probiotics in animals' digestive tracts, which improves immunity and maximizes the use of ingested nutrients (Janczyk *et al.*, 2009).

Additionally, Omar *et al.* (2024) observed that phycocyanins and phycobilins, two antioxidant chemicals found in *spirulina*, may make catalytic enzymes more active such as GPX, CAT, and SOD or inhibit the activity of catalytic enzymes like lipoxygenase and cyclooxygenase. According to Coelho *et al.* (2021) and Zanaty *et al.* (2024), adding microalgae (*chlorella and spirulina*) to broiler chicken diets significantly increased glutathione activity (GPx) and decreased malondialdehyde activity (MDA).

Nevertheless, Reda *et al.* (2024) discovered that growing rabbits given *Moringa oleifera* leaf extract (MOLE) revealed advancements in antioxidant indicators. Interestingly, MOLE showed the capacity to decrease MDA and boost the intracellular antioxidant defense system (CAT, GSH, and SOD) in various experimental models (Mthiyane *et al.*, 2022). Additionally, Hashem *et al.* (2019) found that while MOLE administration increased antioxidant enzyme activity, it decreased the growing rabbits' MDA content. Increased MOLE concentration in rabbit diets was consistently associated with an increase in total antioxidant capacity, improving the oxidative status of the animals (Ojo *et al.*, 2017). MO-derived polysaccharides decreased ROS and MDA levels while increasing GSH-PX, CAT, and SOD activities (Gu *et al.*, 2022).

The MOLE antioxidant activity may be induced by the existence of high concentrations of the most potent antioxidant, vitamin C, as well as other active substances with antioxidant potential, such as phytol and isobenzofuran-1-one-3-acetic acid (Ezhilan and Neelamegam, 2012 and Kadhim *et al.*, 2016). Many antioxidants, including glucosinolate and flavonoids like rutin, quercetin, and kaempferol, as well as phenolic acids (chlorogenic, ellagic, caffeic, ferulic acid, and gallic) are present in significant concentrations in *Moringa oleifera* (Mbikay, 2012). According to Sreelatha and Padma (2009), these antioxidants can stop oxidative damage by increasing antioxidant enzymes, which reduce the production of free radicals and lipid peroxidation. Additionally, MOLE may have an antioxidant effect since it contains polyphenols, anthocyanins, glycosides, and thiocarbamates that eliminate free radicals (Luqman *et al.*, 2012; Kassem *et al.*, 2022 and Maher *et al.* (2023). In line with Selim *et al.* (2021), who discovered that MOL reduced MDA at levels of 2.5, 5.0, and 7.5% of the growing rabbits' diet. *Moringa oleifera* leaf (MOL) has strong antioxidant properties, which may be the cause of this case (Yang *et al.*, 2006). Furthermore, MOL powder and extract had a direct antioxidant action, which decreased MDA levels and lipid peroxidation caused by hydroxyl in rats (Serafini *et al.*, 2011).

The addition of *Moringa* leaf flavonoids also decreased the activity of MDA and raised the activity of T-SOD and T-AOC in plasma, according to Liu *et al.* (2023). These results are compatible with reports that flavonoids cause the liver and kidney of rats to produce more antioxidant enzymes. Because flavonoids increase SOD and GSH activity while lowering MDA concentrations, they can improve antioxidant capacity, boost non-specific immunity, and lessen oxidative stress. To neutralize oxygen-free radicals and eliminate hydrogen peroxide and superoxide ions, flavonoids function as reducing agents and hydrogen donors (Kahkonen, 1999).

Hormonal Parameters:

Insulin, growth (GH), and triiodothyronine (T_3) hormones were increased ($P<0.05$) in the rabbits fed NCO, MO, and NCO+MO compared with the control group. However, thyroxine (T_4) hormone was insignificant increased in the rabbits fed NCO, MO and NCO+MO compared with the control group (Fig. 9).

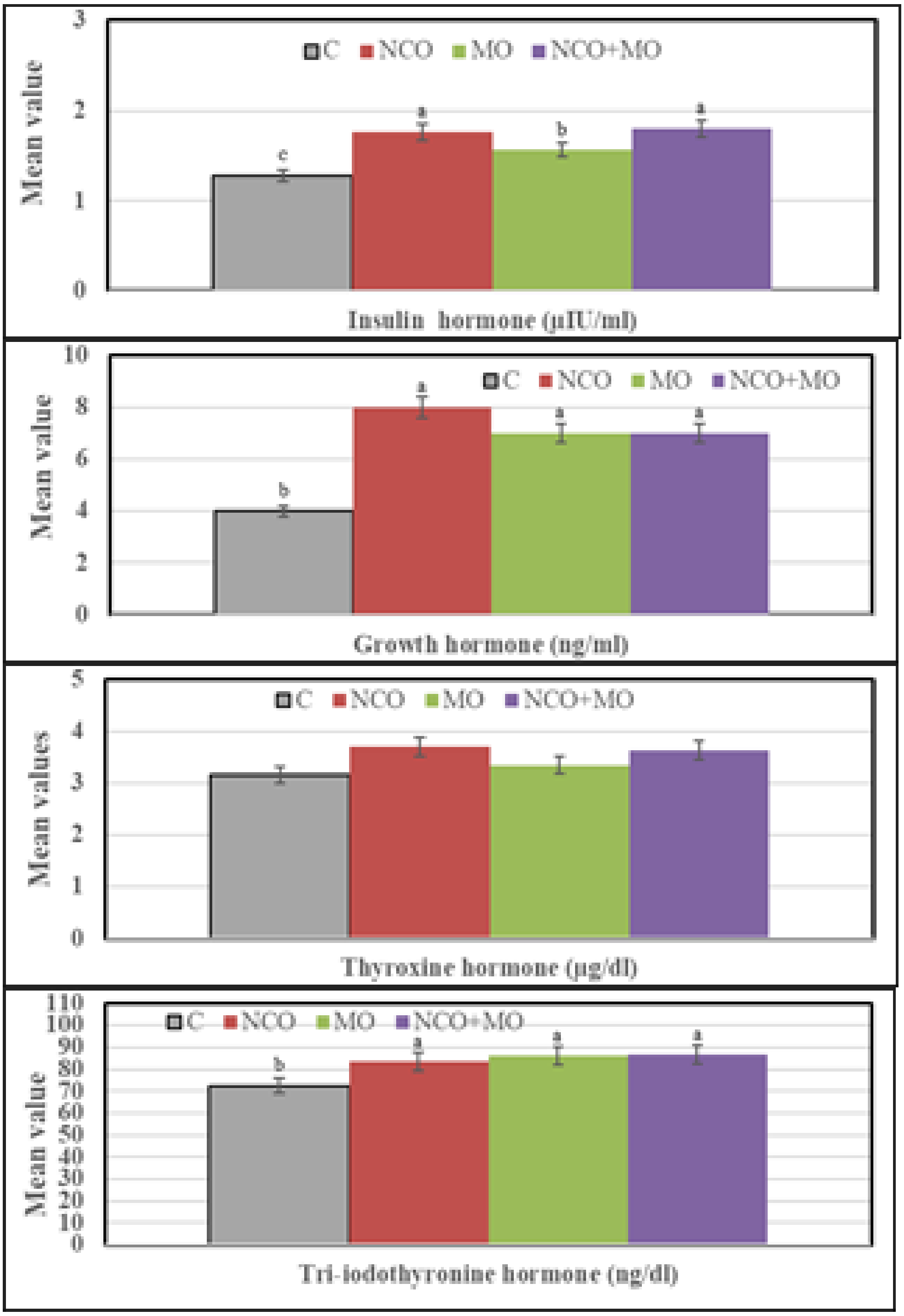

Fig. 9: Effect of *Nannochloropsis oculata* algae and / or *Moringa oleifera* on hormonal parameters of Hi-Plus growing rabbits.
Data represent means± SE (n=15).
C= control group, NCO= rabbits received 2 % *Nannochloropsis oculata* algae, MO= rabbits received 5 % *Moringa oleifera*, NCO+MO= rabbits received 2 % *Nannochloropsis oculata* algae+5 % *Moringa oleifera*.
GH=growth hormone, T_3= tri-iodothyronine, T_4=thyroxine
$^{a-b}$ Means bearing different superscripts within the same item are significantly different (P<0.05).

Although some studies examined how microalgae affected hormonal profiles, numerous studies found that microalgae were rich in various forms of long- and short-chain unsaturated fatty acids (Susana *et al.*, 2018; Salim *et al.*, 2019; and Kholif *et al.*, 2020). The hormone triiodothyronine is crucial for controlling metabolism (Tao *et al.*, 2006). Heat stress conditions and feed intake have a negative relationship with T3 hormone concentration;

therefore, rabbits exposed to heat stress conditions have lower T3 levels (Uni *et al.*, 2001; Attia *et al.*, 2016). Biologically active substances present in microalgae, like carotenoids, astaxanthin, and fucoxanthin have been shown to improve oxidative stress by inhibiting lipid peroxidation (Turrens, 2003). These compounds may also help lessen the negative impacts of stress (Sakr *et al.*, 2019). Consequently, more iodine is incorporated into thyroglobulin, which is used to produce T4 and T3. El-Hawy *et al.* (2022) showed, however, that serum T3 considerably rose in meals supplemented with microalgae in contrast to the control group. Similarly, Abd El-Hamid *et al.* (2022) discovered that when the dietary supplementation ratio of microalgae increased, doe rabbits fed 0.5% or 1.0% microalgae meal throughout the summer had noticeably greater blood levels of T3 and thyroxine (T4) hormones. Conversely, however, Morsy *et al.* (2017) discovered that the buck California rabbits given 3% *Moringa peregrine* leaves (MPL) had considerably greater levels of insulin hormone than the bucks in the control groups. This was because MPL can increase insulin sensitivity. Additionally, El-Kashef (2022) observed that goat children given *Moringa oleifera* leaf had higher T3 concentrations. This could be because MOL has a lot of antioxidants, which can improve physiological responses and lessen the adverse consequences of heat stress.

The Histological Observations of the Liver Tissues:

A-The Control Group:

Figures 10 (A - C) showed normal structure in sections of the liver tissue of the untreated control rabbits. Typical hepatic lobules are composed of the central vein which is found centrally from which irregular cords or plates of epithelial polyhedral cells with acidophilic cytoplasm and prominent rounded nuclei of hepatocytes are radiating. Sinusoid capillaries flew into the central vein of each lobule and separated hepatocyte plates. These capillaries carry blood from the central vein to the portal tra3id. Additionally, endothelial cells and phagocytic kupffer cells line the sinusoids.

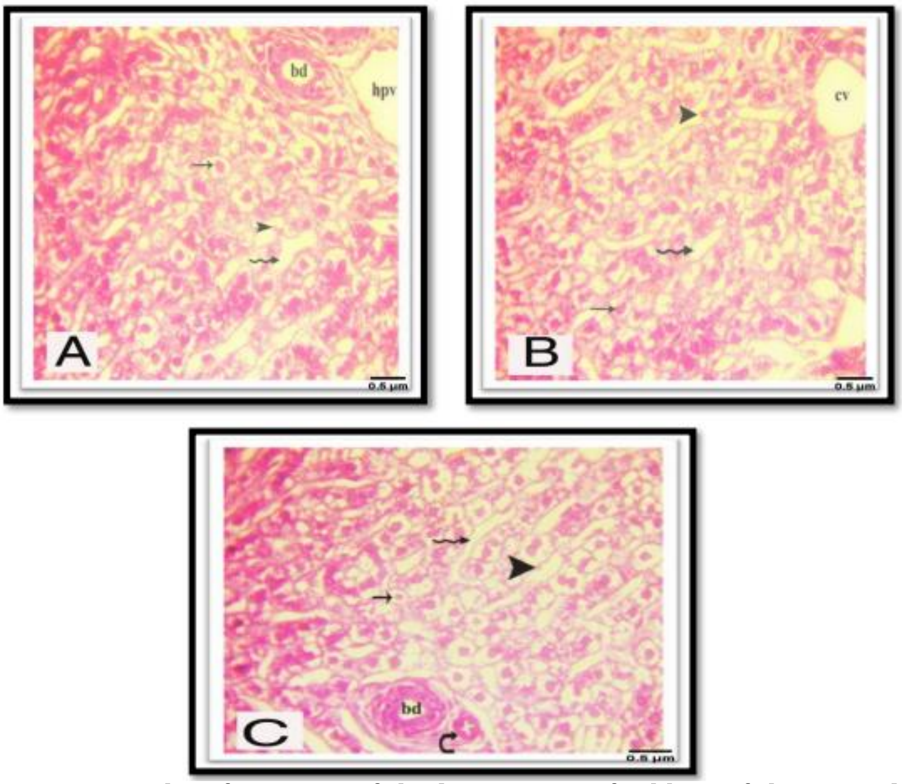

Fig. 10. Photomicrographs of sections of the liver tissue of rabbits of the control group after seven weeks showing:

A-C: normal hepatocytes (**arrow**) and well-arranged hepatic cords with the normal hepatic portal vein (**HPV**), a branch of the hepatic artery (**curved arrow**), and bile duct (**bd**). The central vein (**CV**), cords of hepatocytes that radiate from it and are separated from each other by blood sinusoids (**corrugated arrow**) which are lined with endothelial cells, and phagocytic kupffer cells (head **arrow**) are also observed.

B-The Experimental Groups:

1- ***Nannochloropsis oculata algae treated group (NCO):***

Examination of slices of the liver tissue seven weeks post NCO extract feeding showed well-developed liver architectures with normal appearance of liver cells and blood sinusoids **(**Figs. 11 A-B).

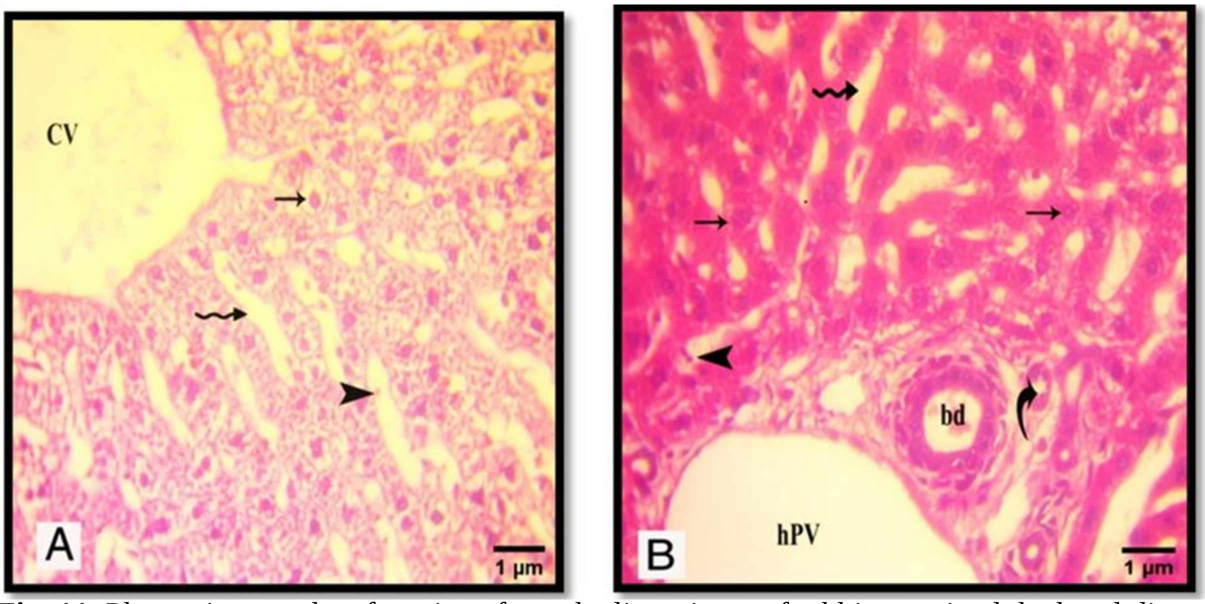

Fig. 11. Photomicrographs of sections from the liver tissue of rabbits received the basal diet + 2% of *Nannochloropsis oculata* for seven weeks showing:

A&B: normal central **(CV)** and hepatic portal **(HPV)** veins. Normal hepatocytes **(arrow)** which split by blood sinusoids **(corrugated arrow).** Well-appeared kupffer cells **(head arrow).** Portal areas contain branches of the hepatic portal vein **(HPV),** branches of the hepatic artery **(curved arrow),** and bile ducts **(bd).**

2-Moringa oleifera **Treated Group (MO):**

The *Moringa oleifera* group after seven weeks of feeding revealed clearly characterized hepatic sinusoids that included kupffer cells inside the well-defined hepatic parenchyma **(**Figs. 12 A&B). No histopathological changes could be seen.

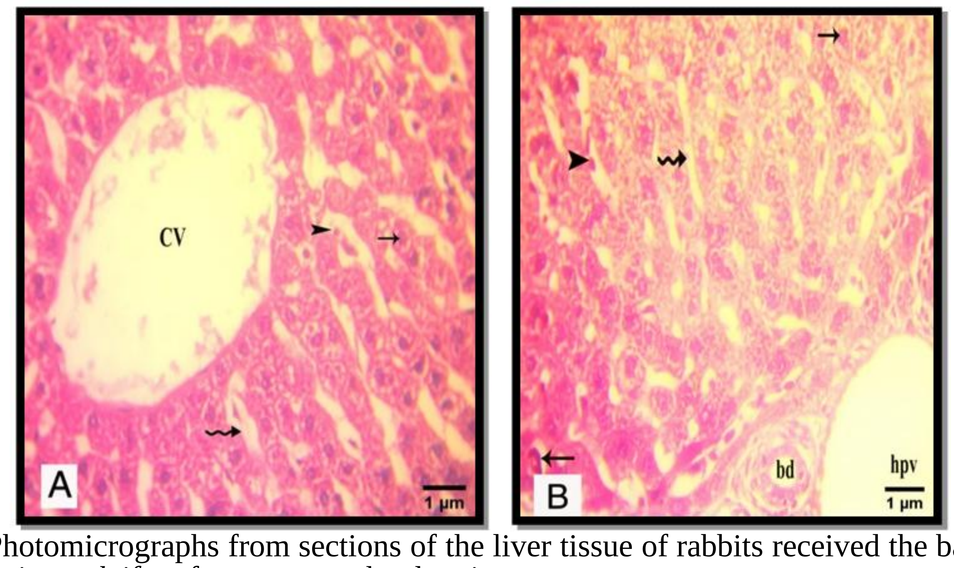

Fig. 12. Photomicrographs from sections of the liver tissue of rabbits received the basal diet + 5% of *Moringa oleifera* for seven weeks showing:

A&B: well, developed central and portal areas include branches of the hepatic portal vein **(HPV),** branches of the hepatic artery **(curved arrow)** with normal hepatocytes **(arrow),** blood sinusoids **(corrugated arrow),** kupffer cells **(head arrow)** and bile ducts **(bd).**

3-*Nannochloropsis oculata* and *Moringa oleifera* Treated Group (NCO+MO):

The **NCO+MO** group illustrated more noticeable and improved hepatic parenchyma with kupffer cells and distinct binucleated hepatocytes **(**Figs.13 A&B**)**.

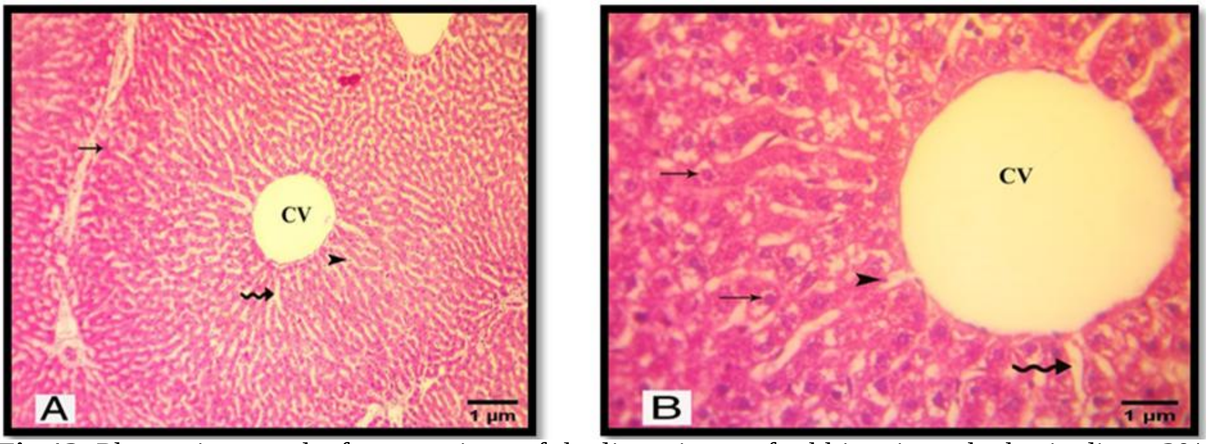

Fig.13. Photomicrographs from sections of the liver tissue of rabbits given the basic diet + 2% *Nannochloropsis oculata* & 5% *Moringa oleifera* for seven weeks showing:
A&B: improved and more noticeable hepatic parenchyma with kupffer cells and clear binucleated hepatocytes **(head arrow).** Normal hepatic portal vein **(HPV),** bile duct **(bd),** and thin sinusoids **(corrugated arrow)** are also noted

In the current investigation, examination of sections of the liver tissue seven weeks post NCO, MO, and NCO+MO feeding showed well-developed liver architectures with a normal appearance of liver cells and blood sinusoids. Serum ALT and AST activity were reduced in this investigation, suggesting that MO supplementation and the microalgae *N. oculata* may protect liver tissues.

According to Mekonnen *et al.* (2005) *in vitro* cytotoxicity investigation, hepatocyte cell viability was unaffected by an aqueous extract of *M. stenopetala* leaves. This is in line with the results of Ghebreselassie *et al.* (2011), who noted that mice given aqueous leaf extract from *M. stenopetala* at all dosages did not exhibit any morphological abnormalities in their liver cells. The active ingredients in *M. stenopetala* and similar species like *M. oleifera* (Behen) leaves are glucosinolates, such as 4-(alpha-L-rhamnosyloxy) benzyl glucosinolate, which is broken down by myrosinase to produce 4-(alpha-L-rhamnosyloxy) benzyl isocyanate. In addition to mustard oil, phenol carboxylic acids, and fatty acids include oleic acid (60–70%), palmitic acid (3–12%), stearic acid (3–12%), eicosanoic acid, and lignoceric acid.

As noted by Ogunnaike (2014), hepatocytes are more noticeable and seem more distinct. These findings showed no harmful impacts on the liver tissues from the administered *Moringa* leaf extract. Additionally, it might suggest that it could promote improved hepatocyte health and function, which enhances hepatocyte functions. The well-established assertion that *Moringa oleifera* is nontoxic and has hepatoprotective properties is confirmed by Dwomoh *et al.* (2024). This finding is consistent with research reported that daily use of *Moringa oleifera* as a spice and herb should be promoted because it has no negative effects on histology or liver function (Aborhyem *et al.*, 2016).

Furthermore, Abdulazeez *et al.* (2020a) noted that although *Moringa Oleifera* might improve liver function and histology, they warned that large dosages (1000 and 2000 mg/kg) of *Moringa* extract had negative implications on Wistar rats' liver, kidney, and brain. Consequently, moderate to low oral dosages of *Moringa oleifera* might be safe and not have cytotoxic implications on the kidneys, liver, or brain. However, in doe rabbits treated with *Spirulina platensis* (SP), El-Ratel *et al.* (2017) discovered that the liver displayed normal and intact architecture. Normal hepatic lobules and a central hepatic vein make up the liver. Because of the neuroprotective properties of *Spirulina platensis* (Aziz *et al.*, 2014), anticancer properties (Konickova *et al.*, 2014), immunomodulatory (Sahan *et al.*, 2015) and anti-inflammatory (Abdel-Daim *et al.*, 2015) properties.

General Conclusion:

In the current research, growing Hi-plus rabbits were chosen as experimental animals considered producing the greatest meat out of all domesticated animals. Because they require less land for production, demand less work, and produce less greenhouse emissions, rabbits are appealing. Research indicates rabbits are becoming more popular since they can produce muscles from 20% of the protein in their diet compared to those with rapid growth and short gestation times (FAO, 2012).

In addition to the high price of rabbit feeding, this study used *Moringa Oleifera* (MOL) and *Nannochloropsis oculata* as alternative sources. Due to MOL's many applications as an inexpensive animal feed, nutritionists have been investigating its effects on livestock performance. Particular emphasis should be paid to the important nutrient levels of MOL, the tree's preferred leaf meal for animals. Tiny amounts of anti-nutritional substances such as tannins, saponins, trypsin inhibitors, and lectins are found in the leaves. The leaves are abundant in every necessary amino acid and high protein, which can help with livestock production. They are also a great source of calcium, iron, copper, sulfur, and important vitamins A, C, and B. The plant's emergence as one of the main local feed ingredients for cattle, pigs, and poultry could be justified by its ability to deliver such potent feed ingredients in addition to its capacity to absorb and neutralize hazardous substances in feed (El Deeb *et al.*, 2015).

However, microalgae are intriguing feedstuffs, especially when considering sustainability. Their inclusion in farm animal diets may improve meat's quality and nutritional content. Microalgae contain significant concentrations of both macro and micro-elements and because of their protein and polysaccharide qualities, they can improve the health and productivity of animals while also improving their growth performance and feed efficiency. Additionally, the high digestibility of amino acids and bioactive substances in algae supplements may have contributed to this increase (Emam *et al.*, 2024). The microalga *Nannochloropsis oculata* has a high protein and eicosapentaenoic acid (EPA) content. Its resistant cell wall reduces the digestion of nutrients (Ribeiro *et al.*, 2020) Accordingly, *N. Oculata* supports productive performance by improving protein absorption, oxidation status, and blood lipid profile (Abdel-Wareth *et al.*, 2024).

In conclusion, In the circumstances of North Sinai, addition of marine microalgae *Nannochloropsis oculata* at a level of 2 % and *Moringa Oleifera* at a level of 5 % (NCO+MO) to the Hi-Plus growing rabbit's diets improved growth performance (body weight gain and feed conversion), improved some hematological parameters, hormonal patterns and oxidative status via prevents oxidative damage to the tissues without any detrimental effect on rabbit health status. It is conceivable that adding *Moringa oleifera* and *Nannochloropsis oculata* as natural feed supplements to growing rabbit diets could improve the health of the animals. Therefore, it is a way to give customers wholesome food and to stop utilizing medications or antibiotics to stimulate development during the fattening phase. Future studies on the impact of microalgae and *Moringa* on the histo-morphological parameters are needed.

Declarations:

Ethical Approval: This study does not contain any studies with human participants or animals performed by any of the authors.

Competing interests: The authors declare that there is no conflict of interest.

Author's Contributions: Elham E. Elhady carried out field execution to all experiment stages, collect blood samples and field data and contributed in wrote this article. Hemmat M. Abdelhafez wrote this article and contributed in drafting the manuscript and revision and help in histo-morphological parameters. Morsy, A.S. performed the statistical analysis of the results, contributed in drafting the manuscript and revision. Farag, B. M. helped in blood plasma biochemical analysis. Doaa Gewily wrote this article and contributed in drafting the manuscript and revision and help in histo-morphological parameters. All authors planned the research and approved the final manuscript.

Funding: The present document was achieved in the frame of project "Utilization of Marine Algae for Salt Fodders, Milk, Meat and Fish production under saline conditions" which was funded by SYSTEL Telecom Company, Egypt. Also, the authors gratefully acknowledge their appreciation to Dr. Ismail Mashhour, Chairman of SYSTEL Telecom, for his support.

Availability of Data and Materials: All data sets are available in the manuscript and supplementary file.

Acknowledgments: In particular, the writers are grateful to Dr. Hassan El-Shaer for making this research possible through the SYSTEL Telecom firm in Egypt's "Utilization of Marine Algae for Salt Fodders, Milk, Meat, and Fish production under saline conditions" project. Additionally, Dr. Ismail Mashhour, Chairman of SYSTEL Telecom, is acknowledged by the writers for his assistance.

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