

## **EFFECT OF USING *SPIRULINA PLATENSIS* AND *CHLORELLA VULGARIS* AS FEED ADDITIVES ON GROWING RABBIT PERFORMANCE**

**Hanan A.M. Hassanein<sup>1</sup>; Marvat M. Arafa<sup>1</sup>, M.A. Abo Warda<sup>1</sup> and Azza A. Abd-Elall<sup>2</sup>**

<sup>1</sup> Animal production Research, Institute Agriculture Research center, Giza, Egypt.

<sup>2</sup> Soils, Water and Environ. Research, Institute Agriculture Research center, Giza, Egypt

### **ABSTRACT**

*The aim of this study was to evaluate the effect of dietary inclusion of microalga Spirulina platensis (SP) and Chlorella vulgaris (CV) as feed additives in balanced diets formulated for growing rabbits on digestibility, growth performance, carcass traits, some blood parameters of rabbits. Sixty New Zealand White (NZW) rabbits (five weeks old with an average initial live body weight (626.85 g) were divided into five experimental groups (12 in each) for the feeding trail where rabbits in control group was fed 100% complete feed diet without algae supplements (T1), while both kinds of algae were added at 0.75 or 1.5g/Kg diets as T2 and T3 for spirulina diets and at the same two levels T4 and T5 for chlorella supplement, respectively. Rabbits fed the experimental diets till twelve weeks old and then digestibility trail was carried out using three animals for each group. Slaughtered test for carcass characteristics evaluation was done in addition some blood parameters were determined as well.*

*Results indicated that the final LBW was significantly higher with both levels of SP supplement (T2&T3) than that of control (T1), but the other two levels of CV supplement (T4 &T5) were insignificantly higher than of control, with the highest value being occurred with T3. Similarly, total body weight gain was significantly higher with both levels of SP during the whole experimental period than that of control group, while the value of the two levels of CV were insignificant higher than that of control one. The highest value was associated with T3 followed in descending order by T2, T4, T5 and lastly the lowest value with control (T1). Only feed intake was significant affected by the addition of high level of SP, while the other tested dietary treatments did not significantly influenced, over the whole experimental period, compared with control. Feed conversion was improved significantly due to all algae treatments except that of the high level of CV supplement (T5) which nearly equal to that of control group. Non significant differences among treatments were found in respect of digestion coefficients of DM, OM, CP, CF, EE and NFE nutrients. Concerning the results of slaughter test, slaughter weight for the two levels of SP- diets had significant higher values than those of CV-diets and control one.*

Similar trend was observed respecting hot carcass weight among experimental treatments. However, both weights and percentages of liver, kidney, heart and total gilet, didn't significant affected by dietary treatments. Also no significant differences among treatments respecting the dressing percentage was found. The lowest significant ( $P \leq 0.05$ ) cholesterol value (37.64 mg/dl) was obtained by rabbits fed diet with high level of SP compared with the control (52.74 mg/dl). Rabbit fed diets supplemented with both levels of SP and also the low CV- diet had performed more economically than the other treatments.

**Conclusively**, results indicated that both levels of *Spirulina* – diets markedly improved growth performance of growing rabbits and in addition reduced liver enzyme activities, cholesterol and total lipids concentration in blood compared with control and the *Chlorella* algae supplement

**Key words:** *Spirulina*, *Chlorella*, rabbits, growth performance, carcass traits.

## INTRODUCTION

*Spirulina platensis* is a microalga with appropriate composition to be used as a food supplement. Which commonly used by humans and animals as protein source. Several studies respecting palatability, lack of toxicity and easy digestion, antioxidant actions, hypocholesterolemic, anticancer, immunostimulant, anti-inflammatory, antiviral, have been conducted to verify the possible benefits of spirulina and some properties have been verified (Rodriguez-Hernández *et al.*, 2001; Derner *et al.*, 2006; Colla *et al.*, 2007). For animal feed, according to Ross and Dominy (1990), the addition of 1.5 to 12% of spirulina into the diets for broilers can replace other protein sources, especially soybean meal, with satisfactory growth rates and feed efficiency. However, the optimal levels for using this alga as a substitute source of the conventional protein on a diet are still controversial (Lacaz-Ruiz, 2003 and Becker, 2007).

Février and Sève (1976) incorporated dehydrated *Spirulina* (*Spirulina maxima*) in the diets of 12-day-old weaned pigs. From 12 to 21 days of age, *Spirulina* was incorporated at a level of 12 % (25 % of total crude protein) in the ration, replacing dried skim milk. From 21 to 42 days of age, *Spirulina* was fed at a level of 8% of the diet, replacing soybean meal. Although there was some reduction in digestibility of the diet when *Spirulina* was incorporated, growth was satisfactory and equivalent in all groups. The authors concluded that the metabolic utilization of the fraction of absorbed feed was better for the *Spirulina* group than that of control group where this response could be noted during the period between 12 and 21 days, although the supply of lysine in the *Spirulina* group was 12 % lower in comparison with control one.

Yap *et al.* (1982) replaced one-half of soybean meal (33 % of total dietary protein) in a corn-soybean meal/dried skim milk starter diet with algal proteins (*Spirulina maxima*, *Spirulina platensis*, and *Chlorella* sp.). The trial was performed with Yorkshire pigs weaned to a dry diet at 4 to 8 days of age. There was no significant difference between control and algal diets during the 15- and 26-day trial periods in growth, diarrhea, loss of appetite, or toxicity. The researchers concluded that at least one-half of the protein supplied by soybean meal (one-third of the dietary protein) could be replaced by algal protein without adverse effects.

Hugh *et al.* (1985) included up to 9 % dehydrated *Spirulina* (*Spirulina platensis*) as protein replacement in swine starter diets, which comprised up to 32 % of the total dietary crude protein. Satisfactory live animal performance was obtained with the 3- to 4-week-old crossbred weanling pigs. No apparent toxicity problems arose with any *Spirulina* level used.

Also, Grinstead *et al.* (2000) performed feeding experiments with dehydrated *Spirulina platensis* and weanling pigs those initially weighted  $3.7 \pm 0.85$  kg and aged 11 - 12 days). From days 0 to 14 after weaning, pigs were fed a control diet or pelleted diets containing 0.2, 0.5, or 2 % *Spirulina platensis* replacing soybean meal on an equal lysine basis. With 2 % *Spirulina platensis*, only 3.2 to 3.4 % of total dietary lysine were replaced. All diets contained a medication (carbodox) and zinc oxide for growth promotion. No differences in pig performance, measured as daily feed intake and gain, were observed during this interval. In contrast to pelleted diets, meal diets resulted in inconsistent responses to *Spirulina platensis*.

Therefore, the aim of this study was to investigate the effects of dietary supplementation of *Spirulina platensis* and *Chlorella vulgaris* on performance, digestibility and serum biochemical parameters of rabbit.

## MATERIAL AND METHODS

The present study was carried out at El- Gemeza Research Station, El-Gharbia government, Animal Production Research Institute, Agriculture Research Center, Ministry of agriculture, Egypt. The experiment aimed to evaluate some algae as additives in growing rabbit diet.

### *Preparation of algae*

In this experiment, the effects of adding *Spirulina* (SP) and *Chlorella* (CV) algae on rabbit performance, nutrient digestibility and some blood metabolite concentrations were investigated. The experimental algae were planted at Agricultural Microbial Department, Soils, water and Environment Research Institute (SWERI), Agricultural Research Center (ARC) Giza, Egypt, using *S. platensis* strain LEB-18. The pilot plant for production of biomass was located near the shore of Mangueira Lagoon (33° 30' 13" S; 53° 08' 59" W) and consisted of raceway tanks of different

dimensions and volumes depending on their purpose. The tanks are protected from UV radiation by the use of a transparent film. When the concentration reached 0.50 g L<sup>-1</sup> (maximum growth), the biomass was separated by filtration, washed and dried at 50°C for 5 h and stored under refrigeration. Spirulina biomass value was calculated through Optical Density (OD) measurements using a spectrophotometer (UV/VIS to 670 nm) and a calibration curve of OD against dry weight (g L<sup>-1</sup>) of Spirulina biomass. After harvesting, the biomass was crushed into a ball mill (88 µm) for analyses.

#### ***Animal and Management***

A total number of 60 New Zealand White (NZW) weaned rabbits at 5 weeks of age and weighed 626.58± 5.12gm were assigned into 5 treatments of 12 rabbits each. Rabbits were housed in galvanized metal rabbit battery cages (3 rabbits /cage) supplied with separated feeders. Diets were offered in pellets form ad libitum and fresh water was available all times. All animals were kept under the same management and hygienic conditions. Both feed intake and live body weight were recorded weekly and feed conversion ratio was calculated

#### ***Experimental diets***

The experimental algae were analyzed to evaluate its chemical composition to formulate the experimental diets as shown in Table 1. Five diets were formulated for the feeding trial, where rabbits in the control group was fed 100% complete feed diet without algae (T<sub>1</sub>), while both kinds of algae were added at 0.75 g Sp/ kg diet and 1.5 g Sp/ kg diet as T<sub>2</sub>, T<sub>3</sub> for spirulina diets and at the same two levels T<sub>4</sub>, T<sub>5</sub> for chlorella ones respectively. All diets were designed to be in Iso-protein and Iso-caloric state and to satisfy the nutrient requirements according to NRC (1977) and the experimental period lasted for twelve weeks of rabbit's age .

#### ***Digestibility trials***

At the last week of the feeding trial period, five digestibility trials were carried out using fifteen NZW rabbits (3 rabbits for each group) where they were individually kept in metabolic cages that allow to collect feces and urine separately. The trials lasted for 7 days, 3 as a primary period followed by 4 days for measurements of actual consumed feed and feces output. Samples of daily feces (25%) of each rabbit were collected every day, dried at 60-70°C for 48h, bulked, mixed, finally ground and kept for chemical analysis, to determine the apparent digestion coefficients of diet nutrients.

**Slaughter trials**

At the end of the growth experimental period, Fifteen randomly chosen rabbits (12 weeks of age), three representing each dietary group were kept off feed 15 hours for slaughtering and collecting blood samples . Giblets weights (Liver, kidneys and heart) and carcass measurements were obtained as percentage of the live body weight according the technique of Blasco *et al.* (1992) and dressing percentage (DP% )was calculated as the follows:

$$DP\% = \text{Carcass wt \%} + \text{Giblets wt \%}, \text{ based on fasting live body weight.}$$

**Table (1).** Composition and calculated analysis of the basal diets.

<b>Ingredients</b>	<b>%</b>
Clover hay	30.0
Barley grains	32.0
Soybean meal ,44%CP	22.9
Wheat bran	09.2
Molasses	03.0
Limestone	01.0
Di- calcium phosphate	00.9
DI-Methionine	00.2
Common salt	00.3
Premix ( 1)	00.3
Anti coccidiosis	00.1
Antifungal	00.1
<b>Calculated chemical analysis<sup>2</sup></b>	<b>(%)</b>
Dry matter	87.33
Ether extract	5.04
Crude protein	16.28
Crude fiber	13.38
Ash	5.49
Ca	1.11
P	0.50
Neutral detergent fiber	30.1
Acid deter	16.63

( 1 ) : Each3 kilogram of Vit+ Min mixture provides: Vitamin A,1200IU; Vitamin E, 20IU ; menadione, 1.3 mg; Vit. D3 2500ICU; Riboflavin, 5.5 mg; Ca. Pantothenate,12 mg ;nicotinic acid ,50 mg ; Choline chloride ,600 mg ; Vit . B12, 10 µg; Vit B6, 3mg ; Thiamine , 3 mg ; Folic acid , 1.0 mg , d-biotin , 50 µg. Trace mineral (milligrams per kg of diet ):Mn.,80; Zn,60 ; Fe,35 CU,8; Se,0.60 .

(2) =Calculated values according to Central Laboratory for Foods & Feeds (CLFF, 2001).

**Analytical methods:**

Chemical composition of the experimental diets and feces were analyzed according to A.O.A.C.(2000). Serum total protein and albumin were determined according to Armstrong and Carr (1964) and Doumas et al.(1971). Serum globulin was calculated by the difference between serum total protein and albumin. Serum transaminases (AST and ALT) and creatinine were measured in serum by specific diagnostic kits according to the recommendations mentioned by Bogin and Keller (1987).

***Economic efficiency***

The data was calculated based on the Egyptian market prices of diets and the live body weight at 2014 as follows:

Net revenue = Total feed cost - Price of total weight gain.

Economic efficiency = Net revenue / Total feed cost.

***Statistical analysis***

Statistical analysis was computed using the general linear model (GLM) procedure (SAS ,1990) and the significant differences among treatments means were separate by Duncan's multiple range test (Duncan,1955).

**RESULTS AND DISCUSSION*****Chemical composition:***

The chemical analysis of cultivated algae which presented in Table (2) indicated that the *Spirulina* and *Chlorella* had highly protein percentage and this result is in agreement with that recorded by (Kay, 1991 and Belay, 2002) who found that protein percentage was ranged from 55- 67. Also (Karkos *et al.*, 2008) revealed that *Spirulina* has become popular and successfully used as a food supplement or used in animal feeding as earlier reported by (Qureshi *et al.*, 1995 and Grinstead *et al.*, 2000).

**Table 2.** Chemical composition (%) of the *Spirulina platensis* (SP) and *Chlorella vulgaris*(CV).

Component	<i>Spirulina platensis</i>	<i>Chlorella vulgaris</i>
<b>Protein</b>	55	60.5
<b>Carbohydrate</b>	25	3.7
<b>Fat</b>	6	12.8
<b>Fiber</b>	6	13.0
<b>Ash</b>	9	4.5
<b>Moisture</b>	4.5	5.4

***Digestion coefficients***

Results presented in Table (3) showed non significant differences among treatments in respect of digestion coefficients of DM, OM, CP, CF, EE and NFE and hence the high level of SP had the best values. These results are disagreed with the findings of Heidarpour *et al.*, 2011 and Peiretti and Meineri 2008, who reported that the high level of spirulina caused decreases in digestibility coefficients of

Holstein calves and rabbits respectively. Also peiretti and Meineri (2008) showed that, with the exception of ether extract, the nutrient digestibilities of rabbits diets with an inclusion of 12 % of a green algae (*S. acutus*) were lower than those of rabbit diets with the same inclusion level of soybean meal.

Whatever Spirulina or other green algae, the digestibility with rabbits was higher than with other monogastric species which may be due to their caecal microbial activity and the nutritional contribution of caecotrophy, that is similar to that of ruminants which have given the best performance and the thermal shock of the pelting process would improve the digestibility of microalgae in rabbit diets (Battaglini *et al.*, 1979).

**Table 3.** Digestion coefficients (%) of the experimental diets fed to growing rabbits.

Items	Treatments				
	T1	T2	T3	T4	T5
<b>DM</b>	69.19	69.75	70.01	69.41	69.33
<b>OM</b>	70.74	70.07	70.39	69.85	69.14
<b>CP</b>	72.26	73.49	73.55	72.20	71.55
<b>EE</b>	85.57	87.99	86.18	85.30	83.76
<b>CF</b>	41.48	41.21	42.67	40.92	40.59
<b>NFE</b>	72.75	70.87	73.34	70.86	70.57

***Growth performance***

Live body weight (LBW), body weight gain (BWG) and feed conversion are shown in Table 4. The differences among treatments of initial body weights at 5 weeks of age were statistically insignificant and ranged between 623.3 and 631.7g. Final LBW was significantly higher with both levels of SP supplement (T2&T3) than that of control (T1), but the other two levels of CV supplement (T4 & T5) were insignificantly higher than that of control, with the highest value being occurred with T3. Similarly the total body weight gain (BWG) was significantly (P<0.05) higher with both levels of Sp supplementation during the whole experimental period than that of control group, while the values of the two levels of CV supplement were insignificant higher than that of control one . The highest value was occurred with T3 followed in descending order by T2, T4, T5 and lastly the lowest value with control (T1). The possible reason for this important might be due to improving the these efficiency of feed utilization. The results are in agreement with those achieved by (Mariey *et al.*, 2012) who reported that body weight was higher significantly when supplemented algae Spirulina to the poultry than that of control diets. Also, Ross and Dominy (1990) found that including spirulina at 50 to 100g/kg diets resulted in normal growth of poultry and found that growth was depressed only when spirulina rate reached 200g/kg diets. While, Grinstead *et al.* (2000) showed inconsistent and minimal improvement in growth performance when pigs were fed diets containing 0.2, 0.5 and 2% SP. In rabbit

diets using 5,10 and 15% of spirulina showed no significant differences on final weight and daily weight gain as reported by (Peiretti and Meineri, 2008). Also, Holman *et al.* (2013) reported that spirulina level of 20% in sheep rations did not significantly improve live weight compared to control group. In the same trend Raach- Moujahed *et al.* (2011) found that increasing Spirulina rate in diets did not significantly affect body weight, daily weight gain or total gain.

Only feed intake was significantly affected by the addition of high level of SP, while the other tested dietary treatments did not significantly influenced over. The whole experimental period compared with control. These results were different than that found with Raach-Moujahed *et al.*(2011) who mention that total intake was not affected by dietary spirulina, supplement.

Feed conversion ratio was improved significantly ( $P<0.05$ ) due to all algae treatments except that of high level of CV supplement (T5) which approximately equal to that of control (T1).The highest feed consumption in rabbit fed diet containing 1.5 g SP compared to lower levels of Sp and other algae could be explained due to the fact that Sp is a rich source of vitamins especially B-complex, oligomonosaccharides and other nutrients. Those might have contributed to increase the palatability of diet. Comparable results were obtained by Zeinhom (2004) who found that inclusion of algae in fish diets insignificantly ( $P<0.05$ ) improved the feed conversion ratio (2.33) and Protein efficiency ratio (1.34), whereas feed intake was significantly increased and therefore improved fish performance.

The beneficial effect of algae is attributed to the fact that it is naturally rich source of several nutrients, especially vitamins, minerals, essential fatty acids, amino acids and other nutrients that may promote faster growth (Gershwin and Belay, 2008). Also Ross *et al.*, (1994) and Nikodèmusz *et al.*, (2010) reported that Spirulina containing diet attained the best feed conversion of laying hens, and Heidarpour *et al.*, (2011) reported that feed conversion was better for Holstein calves fed Spirulina platensis diets than control group. The improvement of the feed conversion ratio, in particular at the rate of 2.5% of diet indicates a better efficiency of metabolic processes of the nutrients. For this reason, Blum and Calet (1975), Toyomizu *et al.* (2001) and Razafindrajaona *et al.* (2008), recommend the use of low dose (lower than 5% spirulina).

Regarding some available contradictory results in the literatures, Dalle Zotte *et al.* (2013) indicated that female rabbits were reached a higher body weight due to high feed intake with *S. platensis* addition. The efficiency of *S. platensis* for rabbit growing has been tested by Peiretti and Meineri (2008a). They showed that the final weight, weight gain and feed efficiency did not differ significantly among the dietary treatments, but *S. platensis* inclusion at a level of 10% gave the highest feed intake. Similarly, Gerencsér *et al.*(2012) had found no statistical differences for

**Table 4.** Growth performance of New Zealand White rabbits as affected by addition algae species and levels.

Items	Treatments					±SE
	T1	T2	T3	T4	T5	
<i>Live body weight(g) at:</i>						
5 wks	623.3	627.9	625.8	631.7	624.2	4.79
12 wks	1815.8 <sup>c</sup>	1944.6 <sup>ab</sup>	1986.3 <sup>a</sup>	1898.3 <sup>abc</sup>	1862.9 <sup>bc</sup>	39.6
Total body weight gain(g)	1192.5 <sup>c</sup>	1316.7 <sup>ab</sup>	1360.4 <sup>a</sup>	1266.7 <sup>abc</sup>	1238.8 <sup>bc</sup>	37.5
Feed consumption(g)	4554.2 <sup>b</sup>	4572.5 <sup>b</sup>	4827.4 <sup>a</sup>	4528.3 <sup>b</sup>	4571.6 <sup>b</sup>	77.8
Feed conversion	3.84 <sup>a</sup>	3.49 <sup>b</sup>	3.57 <sup>b</sup>	3.60 <sup>b</sup>	3.72 <sup>ab</sup>	0.076

<sup>a, b, c</sup> Means within the same row with different litters differ significantly at P<0.05.

final weight and weight gain due to the addition of spirulina supplementation to rabbit diets.

#### ***Carcass traits***

Data of rabbit carcass traits are presented in Table 5. Regarding the fasting slaughter weight, the two levels of SP- diets (T2 & T3) had significant higher values than those of CV- diets (T4&T5) and control one (T1). Similar trend was observed in respect of hot carcass weight among experimental treatments, in which also the SP groups and CV- low level group had higher values than those of CV high level group and control one.

However, both weights and percentages of liver, kidney, heart and total giblet, didn't significant affected by dietary treatments (T2, T3, T4 and T5) in comparison with control (T1). Also, no significant differences among treatments respecting dressing percentage were found. In matching with the present results Raach-Moujahed *et al.* (2011) mentioned that Spirulina algae supplement improved the carcass yield of broiler at a rate of 2.5% of incorporation (75.4%). A similar result were observed by Razafindrajaona *et al.*(2008) who found that Spirulina incorporated in the diets of broiler (100 mg/kg body weight) had improved the carcass yield, with an additional profit from +7% when it is used only during the starter phase of growth up to +11.4% when added during the entire period of rearing.

According to the same authors, the improvement of carcass yield indicate a better development of the "noble" pieces, in particular the thighs and the breast, indicating that Spirulina improved not only the nutritional value of the diet but also its biological efficiency inducing a rise in the noble pieces of broiler.



### **Blood parameters**

Blood parameters data are presented in Table 6. There were no significant differences among treatments regarding the concentrations of total protein, albumin and globulin. The lack of significant effects of algae addition to the diets of rabbits might be partially due to the inclusion of premix component in the diets where it was contained an important packages of vitamins and trace minerals those rendered the blood metabolites to be in the *S. platensis* and *Chlorella vulgaris* are used as a supplement because of their protein, vitamin and mineral contents. However, addition of SP may inhibit the growth of harmful bacteria in intestine (Bhowmik 2009) because harmful enteric bacteria secretes inflammatory agents and lead to increase in globulin synthesis of liver or of other tissues such as lymphatic tissue or plasma cells. On the other hand, Heidarpour *et al.*(2011) studied on albumin, globulin and their assigned ratio in calves fed diets with *S. platensis* in levels of 0, 2, 6 and 25 g/ day, and found no significant effect on serum albumin and globulin levels among treatment groups. Also, Moreira *et al.* (2011) found no significant effect of *S. platensis* on serum albumin and protein levels. However, Mariey *et al.*(2012) stated that SP level at 0.2% had a significant increase in plasma total protein, albumin and globulin in laying hens. Bezerra *et al.*(2009) determined the high serum protein value in lambs fed 0, 5 and 10 g SP. These researchers suggested that the high value of serum protein, globulin and albumin may be due to protein quality and quantity of *S. platensis*. That being is rich in polyunsaturated fatty acids and phycocyanin.

Concerning the blood plasma concentrations of total lipid and triglycerides the differences among treatments did not significant, while cholesterol concentration for T3 was represented the lowest ( $P<0.05$ ) value and T1 and T5 had the highest ( $P<0.05$ ) ones. In line with the present results, Torres *et al.* (1998) and Fong *et al.* (2000) reported a significant reduction of triglycerides and cholesterol concentrations for rats or mice fed *Spirulina* diets. There were significant ( $P<0.05$ ) reductions in cholesterol. The reduction rate of cholesterol relative to that of the control group exhibited by rabbits fed the 1.5 and 0.75 Sp/kg was 37.64 and 45.88 %, respectively.

The current results are in harmony with those reported by Sakaida Takashi (2003) who found that hens fed tested diets supplemented with varying levels of *Spirulina* tended to decrease the cholesterol content in the egg yolk of hens. The mechanism in which SP plays a significant role in lipid metabolism led to significant decrease in total cholesterol, total lipids, LDL-cholesterol and VDL-cholesterol and blood glucose (Kamalpreet *et al.*, 2008). Also, decreases in total cholesterol and LDL were reported when SP was added into human diet (Lee *et al.*, 2008).

This cholesterol serum reduction has been stated as the effect of SP on lipoproteins metabolism and the increase of the lipoprotein enzyme activity levels (Karkos *et al.*, 2008). The presence of antioxidant compounds like phycocyanin and  $\beta$ -carotene, linolenic acid and sulfated polysaccharide in *S. platensis* can be

developing the properties which being decrease the plasma lipid levels. (Nagaoka *et al.*, 2005).

Definitely, Seo *et al.* (2004) reported that  $\beta$ - carotene reduced the elevation of cholesterol and triglycerides of diabetic rats. Presumably, one of the essential metabolic actions of spirulina algae is the improvement of several biochemical parameters and in turn these favorable changes can be reduction of histopathological changes of liver. The hepatoprotective of *S. platensis* may be due to its active principles those being representing in antioxidant activities. These results are, in consistent with the earlier findings with rats (Kato *et al.*, 1984) rabbits (Cheong *et al.*, 2010) and human (Ruitang and Chow, 2010).

Although the mechanism by which the SP reduces cholesterol has not been fully examined, the hypocholesterolemic actions of SP involve reducing plasma and liver cholesterol levels due to the increase in lipoprotein lipase and hepatic triglyceride lipase activity (Karkos *et al.*, 2008), inhibition of both jejunal cholesterol absorption and ileal bile acid resorption (Nagaoka *et al.*, 2005), in addition to modifying lipoproteins metabolism (decrease of low density lipoprotein and increase of high density lipoprotein (Torres-duran *et al.*, 2007). Alternatively, the hypocholesterolemic activity of Spirulina is related to the large amount of cystine found in the C-phycoyanin protein of Spirulina (Nagaoka *et al.*, 2005). Spirulina has been reported to have a hypolipidemic effect due to the C-phycoyanin protein which inhibits the pancreatic lipase activity in a dose-dependent manner (Torres-Duran *et al.*, 2007).

The liver function enzyme alanine aminotransferase (ALT) was not significantly affected by SP or CV levels, while enzyme aspartate aminotransferase (AST) had significantly affected by high SP and low levels, CV where they were recorded the lowest ( $P \leq 0.05$ ) compared with the control.

The activities of AST and ALT are working as an indicators of hepatotoxicity condition (Azab *et al.*, 2013). This results indicating that algae supplements may play a protective role against liver dysfunctions (Bhattacharyya and Mehta, 2012). Concerning blood creatinine concentrations, results proved that there were non significant difference almostly among treatments respecting this item.

#### ***Economic evaluation***

The price of diets was based on the price of ingredients in the Egyptian market during (2014) and the data of economic efficiency are presented in Table 7. It was showed that rabbits fed diets supplemented with both levels of SP and the low level of CV had economically performed better than the other treatments. These results are agreed with those obtained Mariey *et al.* (2012) who used spirulina at 0.1, 1.5, 2.0% in laying hens rations.

**Table 6.** Serum constituents of NZW rabbits fed the experimental diet that including two types of algae.

Items	Treatments					±SE
	T1	T2	T3	T4	T5	
Total protein (g/dl )	6.60	6.53	6.76	6.65	6.94	0.29
Albumin (g/dl)	3.46	3.57	3.39	3.87	3.71	0.18
Globulin (g/dl)	3.14	2.96	3.37	2.77	3.23	0.36
Total lipid (mg/dl)	375.72	364.68	335.13	351.35	342.34	11.86
Triglycerides (mg/dl)	117.45	113.09	112.25	115.9	114.20	1.80
Cholesterol (mg/dl)	52.74 <sup>a</sup>	45.88 <sup>b</sup>	37.64 <sup>c</sup>	44.60 <sup>b</sup>	50.78 <sup>a</sup>	1.49
Creatinine(mg/dl)	1.44 <sup>a</sup>	0.95 <sup>b</sup>	1.20 <sup>ab</sup>	1.31 <sup>a</sup>	1.33 <sup>a</sup>	0.08
AST (U/L)	53.33 <sup>a</sup>	51.67 <sup>ab</sup>	47.00 <sup>c</sup>	47.66 <sup>bc</sup>	53.00 <sup>a</sup>	1.32
ALT (U/L)	73.33	72.33	70.33	71.33	72.00	3.10

AST = Aspartate amino transferase. ALT = Alanine amino transferase.  
 Mean values in the same row with different superscripts differ (P< 0.05).

**Table (7).** Economic efficiency of the experimental diets.

Items	Treatments				
	T1	T2	T3	T4	T5
Price of diets (L.E/kg)	2.700	2.719	2.738	2.719	2.738
Total Feed consumption (kg)	4.554	4.827	4.572	4.528	4.571
Total Feed cost (L.E)(a)	12.29	13.24	12.71	12.31	12.52
Total weight gain (Kg)	1.192	1.360	1.316	1.266	1.238
Price of LBW (LE/Kg)	28	28	28	28	28
Total revenue (LE)(B)	33.38	38.08	36.85	35.45	34.66
Net revenue (LE)	21.08	24.84	24.14	23.14	22.14
Economic efficiency	1.71	1.88	1.89	1.87	1.76
Relative economic efficiency	100	109.94	110.53	109.36	102.92

Economic efficiency% = (Total revenue – total feed cost)/total feed cost X 100 = Net revenue/total feed cost X 100.

**Conclusively**, it could be concluded that both levels of Spirulina supplement into the diet of rabbits markedly improved the growth performance of growing rabbits and in addition reduced liver enzymes activities , cholesterol and total lipid concentrations in blood, compared with control and the chlorella algae supplements.

## REFERENCES

- AOAC. (2000). *Official Methods of Analysis*, 17th ed. Association of Official Analytical Chemists, Arlington, VA, USA.
- Armstrong,W.D. and Carr, C.W. (1964). *Physiological Chemistry: Laboratory Directions. 3rd Edition, Bunge Publishing Co., Minneopolis Minnesota, U.S.A.*

- Azab, S.; Abdel-Daim, M., and Eldahshan, O. (2013)** Phytochemical, cytotoxic, hepatoprotective and antioxidant properties of *Delonix regia* leaves extract. *Med. Chem. Res.*, **22**: 4269-4277.
- Battaglini, M.,(1979).** Algae and yeasts for rabbits. *Coniglicolt.*, **16**: 39–40.
- Becker, B.W.(2007).** Micro-algae as a source of protein. *Biotechnology Advances*, Vol.**25** (2), p.207-210.
- Belay, A. (2002).** The Potential Application of *Spirulina* (*Arthrospira*) as a Nutritional and Therapeutic Supplement in Health Management, *J. Am. Nutraceut. Assoc.*, **5**: 27-48.
- Bezerra, L.R.; Azevedo Silva, A.M.; Azevedo, S.A.; Rodrigues, O.G.; Azevedo, P.S.; and Sousa Mendes, R.(2009).** Serum concentrations of proteins and minerals in lambs artificially fed with *Spirulina platensis*-enriched milk. *Acta Vet Brasil*, **3** (3): 132-137.
- Bhattacharyya, S., & Mehta, P. (2012).** The hepatoprotective potential of *Spirulina* and vitamin C supplementation in cisplatin toxicity. *Food Funct*, **3**: 164-169.
- Bhowmik, D.; Dubey, J. and Mehra, S. (2009).** Probiotic efficiency of *Spirulina platensis* - Stimulating growth of lactic acid bacteria. *World J Dairy & Food Sci*, **4** (2): 160-163, 2009
- Blasco A., Ouhayoun J., Masoero G. (1992).** Status of rabbit meat and carcass: Criteria and terminology. *Options Méditerranéennes, Série Séminaires*, No **17**, 105-120.
- Blum, J.C. and Calet, C. (1975).** Food value of spirulina algae for growth of the broiler-type chicken. *Annales de la Nutrition et de l'Alimentation*, **29**: 651-674.
- Bogin, E. and Keller, P. (1987).** Application of clinical biochemistry to medically relevant animal models and standardization and quality control in animal biochemistry. *J. Clin. Biochem*, **25**:873-878.
- Cheong, S. H., Kim, M. Y., Sok, D. E., Hwang, S. Y., Kim, J. H., Kim, H. R., ... Kim, M. R. (2010).** *Spirulina* prevents atherosclerosis by reducing hypercholesterolemia in rabbits fed a high-cholesterol diet. *J. Nutr. Sci. Vitaminol.*, **56**: 34-40.
- Colla, L.M.; Furlong, E.B. and Costa, J.A.V. (2007).**Antioxidant properties of *Spirulina* (*Arthrospira platensis*) cultivated under different temperatures and nitrogen regimes. *Brazilian Archives of Biology and Technology*, Vol.**50**, n.1, p.161-167,
- DalleZotte, A.; Sartori, A.; Bohatir, P.; Re´mignon, H. and Ricci, R. (2013).** Effect of dietary supplementation of *Spirulina* (*Arthrospira platensis*) and Thyme (*Thymus vulgaris*) on growth performance, apparent digestibility and health status of companion dwarf rabbits. *Livestock Sci.*, **152**: 182-191.
- Derner, R.B.; Ohse, S.; Villela, M. ; Carvalho, S. M. and Fett , R. (2006).** Microalgas, produtos e aplicações. *Ciência Rural*, Vol.**36**, n.6, p.1959-1967.

- Doumas, B.T; Watson, W.A. and Biggs, H.G. (1971).** Albumin standards and the measurement of serum albumin with bromocresol green. *Clin. Chem. Acta.* **31**: 87- 96.
- Duncan, D.B. (1955).** Multiple range and multiple F- test. *Biometrics*, **11**:1-42.
- Février, C., Sève, B. (1976).** Essais d'incorporation de spiruline (*Spirulina maxima*) dans le aliments des porcins. *Ann. Nur. Aliment.* **29**: 625-630.
- Fong, B.; Cheung, M. and Lee, M. (2000)** Effect of dietary Spirulina on plasma cholesterol and triglyceride levels in mice. *In: Abstracts. 4th Asia-Pacific Conference on Algal Biotechnology*,pp.150.
- Gerencsér, Z.; Szendrő, Z.; Matics, Z.; Radnai, I.; Kovács, M. ;Nagy, I.; Dal Bosco, A. and Dalle Zotte, A.(2012).** Dietary supplementation of Spirulina (*Arthrospira platensis*) and Thyme (*Thymus vulgaris* L.). Part 1: Effect on productive performance of growing rabbits. *World Rabbit Science Association Proceedings 10th World Rabbit Congress*, September 3-6, Sharm El- Sheikh -Egypt, 657-661.
- Gershwin, M.E. and Belay, A.(2008).** Spirulina and its therapeutic implications as a food product.In *Spirulina in Human Nutrition and Health. Boca Raton: CRC Press*; 2008:51-70.
- Grinstead, G.S.; Tokach, M.D.; Dritz, S.S.; Goodband, R.D. and Nelssen, J.L.(2000).** Effects of *Spirulina platensis* on growth performance of weanling pigs. *Anim. Feed Sci. Technol.*, **83**: 237-247.
- Heidarpour, A.; Fourouzande, H.; Shahraki, A.D. and Eghbaisaie, S. (2011)** Effects of *Spirulina platensis* on performance, digestibility and serum biochemical parameters of Holstein calves. *African Journal of Agricultural Research*, Vol. **6**(22), pp. 5061-5065.
- Holman, B. W. and Malau-Aduli, A. E. (2013).** Spirulina as a livestock supplement and animal feed. *J. Anim. Physiol. Anim. Nutr.*, **97**(4), 615-623.
- Hugh, W.I.; Dominy, W. and Duerr, E. (1985).** Evaluation of dehydrate spirulina (*Spirulina platensis*) as a protein replacement in swine starter diets. *University of Hawaii Research and Extension Series*, 056, Honolulu.
- Kamalpreet, K.; Rajbir, S. and Kiran, G. (2008).** Effect of supplementation of spirulina on blood glucose and lipid profile of the non-insulin dependent diabetic male subjects. *J. Dairying, Foods Home Sci.*, **27**(3).
- Karkos, P.D.; Leong, S.C.; Karkos, C.D.; Siraji, N. and Assimkapoulos, D.A. (2008).** Review of spirulina in clinical practice: Evidence-Based human applications. *e CAM Adv. Access*, **14**: 1-4.
- Kato, T.; Takemoto, K.; Katayama, H. and Kuwabara, Y.(1984).** Effects of Spirulina (*Spirulina platensis*) on dietary hypercholesterolemia in rats. *J. Jap. Soc. Nutr. Food Sci.*, **37**: 323-332.
- Kay, R.A. (1991).** Microalgae as food and supplement. *Food Sci. Nutr.*,**30**: 555-573.
- Lacaz-Ruiz, R. (2003).**Espirulina: estudos e trabalhos. São Paulo: Roca, 296p.
- Lee, E.H.; Park, J.; Choi, Y.; Huh, K. and Kim, W. (2008).** A randomized study to establish the effects of spirulina in type 2 diabetes mellitus patients. *Nutr. Res. Pract.*, **2**(4): 295-300.

- Mariey, Y.A.; Samak, H.R. and Ibrahim, M.A.(2012).** Effect of using *Spirulina platensis* algae as a feed additive for poultry diets: 1- Productive and reproductive performances of local laying hens., *Egypt. Poult. Sci.*, **32** (1): 201-215.
- Moreira, L.M.; Rocha, A.S.R.; Ribeiro, C.L.G.; Rodrigues, R.S. and Soares, L.S. (2011).** Nutritional evaluation of single-cell protein produced by *Spirulina platensis*. *Afr. J. Food Sci.*, **5** (15): 799-805.
- Nagaoka, S.; Shimizu, K.; Kaneko, H.; Shibayama, F.; Morikawa, K.; Kanamaru, Y.; Otsuka, A.; Hirahashi, T. and Kato, T.(2005).** A novel protein cphycocyanin plays a crucial role in the hypocholesterolemic action of *Spirulina platensis* concentrate in rats. *J. Nutr.*, **135**: 2425-2430.
- Nikodémusz, E.; Páskai, P.; Tóth, L. and Kozák, J., (2010).** Effect of dietary *Spirulina* supplementation on the reproductive performance of farmed pheasants. *Technical Articles -Poultry Industry*, pp. 1-2.
- NRC (1977).** National Research Council. Nutrient requirements of rabbits. Nat. Acad. Sci., Washington, DC., USA.
- Peiretti, P.G. and Meineri, G. (2008).** Effects of diets with increasing levels of *Spirulina platensis* on the performance and apparent digestibility in growing rabbits. *Livest. Sci.*, **118**: 173-177.
- Peiretti, P.G., Meineri, G., (2008a).** Effects on growth performance, carcass characteristics, and the fat and meat fatty acid profile of rabbits fed diets with chia (*Salvia hispanica* L.) seed supplements. *Meat Sci.*, **80**: 1116–1121.
- Qureshi, M.A.;Kidd, M.T. and Ali, R.A. (1995).** *Spirulina platensis* extract enhances chicken macrophage functions after in vitro exposure. *Journal of Nutritional Immunology*, **3**(4): 35-45.
- Raach-Moujahed , A.; Hassani , S.; Zairi , S.; Bouallegue, M.; Darej , C.; Haddad, B. and Damergi, C. (2011).** Effect of dehydrated *Spirulina platensis* on performances and meat quality of broilers. *Roavs*, 2011, **1**(8), 505-509
- Razafindrajaona, J.M., Rakotozandriny, J.N.,Rakotozandrindrainy, R., Tsivingaina, A., Ramapiherika, K.D. and Randria, J.N. (2008).** Influence de l'incorporation dans les provendes de la spiruline de Madagascar (*Spirulina platensis* var. *toliaensis*) sur la croissance des poulets de chair. International Symposium on *Spirulina*–Toliara Sud-Ouest de MADAGASCAR. April 2008.
- Rodriguez-Hernández, A.; Blé-Castillo, J. L.; Juárez- Oropeza, M.A. (2001)** *Spirulina Maxima* Prevents Fatty Liver Formation In Cd-1 male and female mice with experimental diabetes. *Life Sciences*, Vol.**69**, n.9, p.1029-37.
- Ross, E. and Dominy, W.(1990).** The nutritional value of dehydrated, blue-green algae (*Spirulina platensis*) for poultry. *Poultry Science*, **69**: 794-800.
- Ross, E.; Puapong, D.P.; Cepeda, F.P. and Patterson, P.H.,(1994).** Comparison of freeze-dried and extruded *Spirulina platensis* as yolk pigmenting agents. *Poultry Science*, **73**:1282-1289.

- Ruitang, D., & Chow, T. J. (2010).** Hypolipidemic, antioxidant, and anti-inflammatory activities of microalgae *Spirulina*. *Cardiovasc. Therapeutics*, **8**: 33-45
- Sakaida Takashi, (2003).** Effect of administration of *Spirulina* on egg quality and egg components. *Animal Husbandry*, **57**: (1):191-195.
- SAS (1990).** SAS User's Guide, SAS. Institute Inc, Cary, NC.USA
- Seo, J.S.; Lee, K.S.;Jang, J.H.; Quan, Z.; Yang, K.M. and Burri, B.J. (2004).**The effect of dietary supplementation of  $\beta$ -carotene on lipid metabolism in streptozotocin-induced diabetic rats. *Nutr. Res.*, **24**: 1011-1021.
- Torres-Duran PV, Miranda-Zamora R, Paredes-Carbajal MC, Mascher D, Diaz-Zagoya JC, Juarez-Oropeza M. (1998).** *Spirulina maxima* prevents induction of fatty liver by carbon tetrachloride in the rat. *Biochem. Mol. Biol. Int.*, **44**:787-793.
- Toyomizu, M., Sato, K., Taroda, H., Kato, T. and Akiba, Y. (2001).** Effects of dietary *Spirulina* on meat colour in muscle of broiler chickens. *British Poultry Science*, **42**: 197–202
- Yap, T.N.; Wu, J.F.; Pond, W.G. and Krook, L. (1982).** Feasibility of feeding *Spirulina maxima*, or *Chlorella* sp. to pigs weaned to a dry diet at 4 to 8 days of age. *Nutr. Rep. Int.*, **25**: 543-552.
- Zeinhom, M. M. (2004).** Nutritional and physiological studies on fish. Ph. D. thesis. Faculty of Agriculture, Zagazig University. Egypt.

## تأثير استخدام طحلي الاسبيرولينا والكلوريل كإضافات غذائية على أداء الأرانب النامية

حنان احمد محمود<sup>١</sup> - ميرفت محمود عرفه<sup>٢</sup> - محمد انور ابو وردة<sup>١</sup> - عزة أحمد محمد عبد العال<sup>٢</sup>  
 ١ - معهد بحوث الإنتاج الحيواني - مركز البحوث الزراعية ، الجيزة ، مصر .  
 ٢ - معهد بحوث الاراضى والمياة والبيئة (قسم بحوث الميكربولوجيا الزراعيه)- مركز  
 البحوث الزراعية ، الجيزة ، مصر .

تهدف الدراسة الى تقييم تأثير دمج كلا من طحلي الاسبيرولينا والكلوريل كإضافات غذائية في علائق متزنة للارانب النامية على كل من المهضوم ، الاداء الانتاجي ، صفات الذبيحه وبعض قياسات الدم للارانب. قسمت ٦٠ ارنب نيوزيلندي ابيض ( عمر ٥ اسابيع بمتوسط وزن ٦٢٦,٨٥ جم) الى ٥ مجاميع تجريبية (١٢ ارنب لكل مجموعه) لتجربة التغذية وتغذت المجموعه الكونترول على عليقة كاملة ١٠٠% بدون اضافة طحلب ، بينما اضيف نوعي الطحلب بنسبة ٠,٧٥ او ١,٥ جم/كجم عليقة لعليقة المعامله الثانيه والثالثه من الاسبيرولينا وبنفس المستويات لعلائق المعامله الثالثه والرابعه لإضافة الكلوريل على التوالي. تغذت لارانب على العلائق التجريبية حتى عمر ١٢ اسبوع من العمر ثم اجريت تجربة هضم استخدم فيها ٣ حيوانات من كل مجموعه. قيمت صفات الذبيحه من خلال تجربة ذبح بالاضافة الى تقدير بعض قياسات الدم.

اتضح من النتائج ان الوزن النهائي كان مرتفع مع كلا المستويان من الاسبيرولينا ( المعامله الثانيه و الثالثه) اكثر من الكونترول ولكن المستويان الاخران من اضافة الكلوريل ( المعامله الرابعه والخامسه) كانت الارتفاع غير معنوي عن الكونترول . لوحظ ان اعلى قيمه كانت مع المعامله الثالثه. وبالمثل فان اجمالي الزيادة الوزنيه كانت مرتفعه معنويا مع مستويات الاسبيرولينا خلال اجمالي الفتره التجريبية عن المجموعه الكونترول بينما قيمه ارتفاعها كان غير معنوي مع مستويات الكلوريل عند المقارنه بالمجموعه الكونترول. كانت اعلى قيمه مع المعامله الثالثه وتلتها تنازليا على الترتيب المجموع الثانيه ، الرابعه، الخامسه وكانت اخرها واقلها مع المجموعه الكونترول . تآثر فقط الماكول معنويا مع اضافة مستويات عاليه من الاسبيرولينا بينما لم يحدث تأثير معنوي لغيرها من العلائق التجريبية خلال اجمالي الفتره مقارنه بالكونترول. تحسن التحويل الغذائي معنويا مع معاملات الطحالب فيما عدى المستوى العالي من الكلوريل ( المعامله الخامسه) والتي كانت تقريبا مساويه للمجموعه الكونترول . لم تظهر اي اختلافات معنويه بين المعاملات مع معاملات هضم المركبات الغذائيه لكل من الماده الجافه ، الماده العضويه، والبروتين الخام،

الالياف الخام، مستخلص الايثير و مستخلص خالي النيتروجين . ونتيجة اختبار الذبح اظهرت ارتفاع معنوي في قيمة وزن الذبيحه مع علائق مستويان ال اسبيرولينا مقارنة مع علائق الكلوريل و مجموعة الكونترول. وفي نفس الاتجاه لوحظ نتائج وزن الذبيحه ساخنه بين المجاميع التجريبيه بينما الزن والنسبه المئوية لكل من الكبد والكليتين والقلب ومجموع الجزاء الماكوله لم يتاثر معنويا مع العلائق التجريبيه. كذلك لم يظهر تاثير معنويا بين المعاملات في نسبة التصافي. كانت اقل قيمه معنويه للكوليتسول (٣٧,٦٤ مجم/دسل) مع الارانب المغذاه على مستويان لاسبيرولينا مقارنة بالكونترول (٥٢,٧٤ مجم/دسل). وسجلت الارانب المغذاه على كلا المستويان من الاسبيرولينا وكذلك المستوى المنخفض من الكلوريل اداء انتاجي اكثر اقتصاديا من المغذاه على باقي المعاملات.

**التوصية:** اظهرت النتائج ان كلا المستويان من الاسبيرولينا حسنت بشكل ملحوظ الاداء الانتاجي للارانب الناميه بالاضافه الى انخفاض انزيمات نشاط الكبد والكوليتسول وتركيز اجنالي الدهون في الدم مقارنة بالكونترول وازضافة طحلب الكلوريل.

**الكلمات الداله:** الاسبيرولينا، الكلوريل، الارانب، الاداء الانتاجي، صفات الذبيحه.