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EFFECT OF STOCKING DENSITY AND *SPIRULINA PLATENSIS* ALGAE SUPPLEMENTATION AS A FEED ADDITIVE ON PERFORMANCE AND PHYSIOLOGICAL STATUS OF BROILER CHICKS

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SUMMARY

A trail was conducted to study the beneficial effects of *Spirulina platensis* algae as a feed additive under different stocking density on broiler performance, physiological status and economic efficiency at 6 week of age. A total of 540 unsexed day- old Ross × Ross broiler chicks were distributed into nine treatments resulted from 3×3 factorial arrangement (60 birds each). Birds were allotted to three groups according to stocking density (10, 12 and 14 birds/m^2). Each group was divided into three sub-groups according to adding basal diet along with *Spirulina platensis* supplementation levels (0, 0.15 and 0.25 g/kg diet), Body weight, weight gain, feed consumption, feed conversion ratio, mortality rate, carcass trait, some blood metabolites and economic efficiency were studied. The results showed that, all of studied parameters were significantly ($P \leq 0.05$) improved at stocking density 10 birds/m^2 compared to other treatments, the same trend was observed for group fed diet containing spirulina at level of 0.25 g/kg diet. Additionally, an extra improvement in all performance, physiological parameters and economic aspects was showed as a result to interaction between stocking density of 10 birds/m^2 and supplementation of spirulina at 0.25g/kg diet. Regarding to the effect of spirulina supplementation level and stocking density on broiler performance, physiological status and economical aspect, *Spirulina platensis* algae could be safely used in broiler feeding, at level of 0.25 g /kg diet and 10 birds/m^2 stocking density with superior effects on their productive performance. Also, the current study revealed the ability to produce a safety broiler meat for consumers by using natural substances away from antibiotics or other chemicals.

Keywords: *Stocking density, Spirulina platensis, broiler, performance and physiological status.*

INTRODUCTION

Stocking density of broiler flocks (the number of birds reared in a given housing area), is essential sheltering conditions for broiler flocks. Many studies around the world (Özbey and Esen 2007; Estevez *et al*., 1997 and Azzam and El-Gogary 2015) have been conducted to increase stocking density to maximize profitability. Rearing excessive number of birds per unit area increases stress and disease outbreaks, which lead to growth retardation and degradation in end products quality. On the other hand, placing fewer animals than the optimum number causes economic losses (Koçak, 1985). For this reason, optimum stocking density (moderate number) that will not cause low efficiency and will provide economic benefit should be determined (Erişir and Erişir 2002).

The use of traditional protein sources in poultry diet is becoming very expensive, particularly in countries that import most of their raw materials (El-Adawy *et al*., 2013). Therefore, recent pattern in the feed manufacturing is currently oriented toward the use of natural compounds as different options to synthetic nutrients, antibiotics and other chemicals (Kaoud, 2012).

Blue green algae or cyanobacteria are one of the brilliant common feed additives that can be utilized as a part of poultry diets to reduce the limiting of feedstuffs and their expensive cost specially the genus of Spirulina (Mariey et al., 2014). *Spirulina platensis* as one of the blue green algae; is conceivable wellspring of high protein has been utilized as a part of poultry feeding methodologies. It is represents a potential ingredient for animal feed, due to its high protein content (55-65%) on dry matter basis; What's more, it has a decent amount of all essential amino acids, fatty acids, vitamins, phytochemicals and minerals (Shanmugapriya *et al*., 2015; Abd El-Baky *et al*., 2003; Khan *et al*., 2005; Anusuya *et al*.,1981; Ross and Dominy 1985). In addition, it has Available energy (about 2.50 – 3.29 kcal / gram) and 41% non-phytate phosphorus (Blum *et al.*, 1976; Yoshida and Hoshii, 1980).

The recent investigations of Khan *et al.*, (2005); Islam *et al.*, (2009);Nikodémusz *et al*., (2010); Kaoud, (2012); Kharde *et al.*, (2012); Mariey *et al.*, (2012) and (2014) cited that, the supplementation of dehydrated *Spirulina platensis* at different levels from 0.05 to 1% in poultry diets induced the immune-response by increasing microbial killing, antigen processing and greater T-cell activity; additionally, improved productive performance, nutrient digestibility coefficient and physiological status.

Therefore, the present investigation was conducted to study the effect of stocking density and *Spirulina platensis* supplementation on broiler performance, carcass traits, blood metabolites and economic efficiency.

MATERIALS AND METHODS

A total number of 540 unsexed one-day-old Ross broiler chicks were obtained from commercial hatchery; individually weighed and randomly allotted into nine equal groups, each of which included three replicates. A 3 × 3 experiment with factorial arrangement of treatments, three stocking density (10, 12 and 14 birds/m^2) and three level of *Spirulina platensis* supplementation (0.0, 0.15 and 0.25 g/kg diet). The chicks were grown in a separate floor pens with wood shavings and exposed to 24 h constant light. Temperature was maintained at 34°C for the first 3 days and then gradually reduced according to normal management practices. All chicks were kept under the same managerial, hygienic and environmental conditions.

Feed and water were supplied *ad-libitum*. A basal diet was prepared to contain 3000 kcal ME/kg and 23% crude protein during the starter period till 21 day followed by 3130 kcal ME/kg and 19.31 % crude protein during the finisher period till 42 day. The composition and calculated analysis of the basal diet are presented in Table (1) .

Table (1): Composition and calculated analyses of the experimental diets

Ingredient	Starter	Finisher
Yellow corn	53.30	64.0
Soybean meal (48%)	34.00	25.5
Corn gluten meal (60%)	4.00	3.25
Vig. Oil	4.70	3.75
Ground limestone	1.20	1.20
DI-Calcium Phosphate	2.00	1.50
Salt	0.30	0.30
Permix*	0.20	0.20
DL-methionine	0.10	0.10
L-Iysine	0.10	0.10
Coccidiostate	0.10	0.10
Total	100	100
Calculated analysis**		
Crud protein, %	23.0	19.31
Metabolizable energy (K cal/kg)	3000.0	3130.08
Ether extract (EE%).	2.83	2.89
Crude fiber (CF%)	4.15	3.3
Calcium	1.03	0.93
Available phosphorus, %	0.51	0.42
Lysine, %	1.31	0.97
Methionine, %	0.65	0.63
Meth. + Cyc., %	0.92	0.86

** Each 3 kg of premix contained: vit. A 12000 IU, vit. D 2200IU, vit. E 10 mg, vit. K3 2000 mg, vit. B1 1000 mg, vit. B2 5000 mg, vit. B6 1500 mg, vit. B12 10 mg, pantothenic acid 10 mg, niacin 30 mg, folic acid 1000 mg, biotin 50 mg, choline chloride 300 mg, manganese 60 mg, zinc 50 mg, copper 10 mg, Iron 30 mg, Iodine 1000 mg, selenium 100 mg, cobalt 100 mg and CaCo3 to 3 g.*

*** According to NRC. 1994.*

The response of the chicks was assessed in terms of weekly body weight, feed intake and feed conversion ratio. At 42 days of age, nine birds from each treatment were sacrificed, scalded, de-feathered and carcasses were eviscerated. Three bird's carcasses from each group were used for sensory tests. At the same time, blood samples were collected from sacrificed birds in each group. Plasma was separated by centrifugation at 3000 rpm for 15 min. Plasma samples were analyzed for the concentration of total lipids (Frings and Dunn, 1970), cholesterol (Allain *et al.*, 1974) and triglyceride (Fassati and Prencipe, 1982) by using commercial kits. Concentration of plasma total protein and albumin were colorimetrically estimated (Doumas *et al.*, 1971). Plasma globulin concentration was obtained by subtracting the concentration of albumin from total protein.

Statistical analyses were conducted using analysis of variance on the SAS procedure (2003). Significance of differences between groups was determined using the Duncan multiple range test (Duncan, 1955) test for post-hoc comparisons.

RESULTS AND DISCUSSION

Growth Performance:

Live body weight and weight gain

The effects of stocking density, *Spirulina platensis* supplementation levels and there interaction on performance traits of broiler chicks are represented in Tables (2 and 3). Data revealed that, live body weight at 21 days of age was significantly ($P \leq 0.05$) decreased by 5.9 and 8.5% by increasing stocking density from 10 to12 and 14 bird/m^2, respectively. The same trend was observed at 42 days of age, whereas live body weight was significantly ($P \leq 0.05$) decreased by 5.8 and 15.2% by increasing stocking density from 10 to12 and 14 bird/m^2. Also, the results showed that, at the entire experimental period, live body weight gain was significantly ($P \leq 0.05$) decreased by increasing stocking density from 10 to 12 and 14 birds/m^2.

These findings may be due to that the increasing of stocking density reduce the chance of birds to get their nutritional requirements which lead to a decreasing in body weight and weight gain . These results are compatible with those reported by Feddes *et al*., (2002) and Dozier *et al.*, (2006), who found that final body weight and weight gain decreased by increasing stocking density. Also, Shanawany (1988) reported that stocking density exceeding 30 birds/m^2 led to a greater decrease in final body weight and weight gain as compared with stocking densities of 10 and 20 birds/m^2.

At the end of starter period, body weight of birds fed 0.25 and 0.15 g Spirulina/kg diet was significantly ($P \leq 0.05$) heavier by 6.16 and 2.40% respectively, compared to the control birds. Similar trend was observed at the end of the experimental period, whereas body weight of birds fed 0.25 g Spirulina/kg diet was significantly ($P \leq 0.05$) increased by 9.19% than the control, followed by those fed 0.15 g Spirulina/kg diet (2.25%), respectively .

Body weight gain of broiler chicks fed different levels of dietary Spirulina significantly ($P \leq 0.5$) increased as compared to the control group during starter, finisher and the entire experimental periods, where birds fed 0.25g Spirulina/kg diet significantly ($P \leq 0.05$) showed the highest weight gain, followed by those fed 0.15g Spirulina/kg diet compared to the control group. Weight gain of birds fed 0.25 and 0.15g Spirulina/kg diet represented 6.49 and 2.57% when compared with the control birds during starter period. The corresponding values were 12.02 and 2.18% during the finisher period; 10.12 and 2.31% at marketing age compared to the control group respectively.

These results are in agreement with those obtained by Nikodĕmusz *et al*. (2010) who reported that birds fed dietary Spirulina had benefit effects on productive performance. Also, Kaoud, (2012) found that body weight was significantly ($P \leq 0.05$) increased by the dietary supplementation of *Spirulina platensis*. The significant effect of dietary treatments on body weight of broiler fed spirulina diet may be brought through improving the efficiency of feed utilization. In this regard, Raju *et al.* (2005) concluded that dietary inclusion of Spirulina at a level of 0.05% can partially offset the adverse effects of aflatoxin on growth rate of broiler chickens. Also, Mariey *et al*. (2014) found that body weight of birds fed 0.3 and 0.2 g was significantly ($P<0.05$) heavier by 17.6 and 17.9%, followed by group fed 0.1g Spirulina/kg diet (5.7%), compared to the control birds at 21 weeks of ages. They found also that, similar trend was observed in body weight of birds at day 42, whereas body weight of birds fed 0.3 g Spirulina/kg diet was significantly ($P \leq 0.05$) heavier by 13.5% than the control. In addition, Shanmugapriya *et al.*, (2015) cited that, the addition of *Spirulina platensis* at level of 1% improved body weight and weight gain of broiler chicks compared to control group or 1.5% supplemented level.

The obtained results of interaction between stocking density and Spirulina supplementation on live body weight and weight gain of broiler at 21 or 42 days of age; revealed that, there was significant (P≤0.05) increasing in live body weight and weight gain of broiler chicks housed at 10 birds/m2 of stocking density with adding Spirulina at 0.25g/kg diet compared to other interaction groups.

Table (2): Average body weight of broiler chicks as affected by stocking density and *Spirulina Platensis* supplementation levels.

Treatment	Initial BW (g)	BW at 21 day (g)	Final BW at 42 day (g)
Effect of Stocking Density			
Control (C) 10 bird/m^2	41.2	710.5[a]	2060.7[a]
12 bird/m^2	41.2	668.6[b]	1940.5[b]
14 bird/m^2	41.2	649.9[c]	1747.0[c]
SEM	±0.9	±5.2	±10.2
Effect of *Spirulina platensis* Supplementation Levels			
Control (C) 0.00 g/Kg	41.3	657.6[c]	1845.6[c]
0.15 g/ Kg	41.1	673.4[b]	1887.2[b]
0.25 g/Kg	41.4	698.1[a]	2015.3[a]
SEM	±0.9	±5.2	±9.7
Effect of Interaction between Stocking Density and *Spirulina platensis* Supplementation			
0.00 g/Kg	41.2	691.3	1966.8
10 birds * 0.15 g/Kg	41.0	705.0	1985.4
0.25 g/Kg	41.4	735.3	2230.0
0.00 g/Kg	41.2	647.5	1870.1
12 birds * 0.15 g/Kg	41.0	665.1	1901.3
0.25 g/Kg	41.0	693.2	2050.0
0.00 g/Kg	41.0	634.1	1700.0
14 birds * 0.15 g/Kg	41.2	650.2	1775.0
0.25 g/Kg	41.5	665.4	1766.0
SEM	±0.09	±5.1	±10.1
Significant	NS	*	*

-Means of each column followed by the same letter are not significantly different at the 5% level according to Duncan ,s Multiple Range Test. -SEM indicate standard error of means -NS indicate not significant. - indicate P<0.05*

Feed consumption and feed conversion ratio

The effect of stocking density, Spirulina supplementation level and there interaction on feed consumption (FC) and feed conversion ratio (FCR) of broiler chicks are listed in Table (3) . Broiler chicks reared under stocking density 10 birds/m2 significantly had (P≤0.5) the better feed consumption and feed conversion ratio compared to the other groups during starter, finisher and the entire experimental periods. At the end of starter, finisher and entire experimental periods; feed consumption and feed conversion ratio were significantly (P≤0.05) increased by increasing stocking density from 10, 12 and 14 birds/m^2.

The present results disagreed with findings obtained by Proudfoot and Hulan (1985) and Dozier *et al*., (2006) who reported that feed consumed was not significantly different when broilers were reared under many stocking densities. While feed conversion ratio was numerically higher for the group reared under the highest stocking density in broiler chicks. Also, results disagreed with the findings obtained by Thompson, (1972) who found a significant decrease in feed intake per bird as the space per bird decreased .

Feed consumption and feed conversion ratio of broiler chicks fed different levels of dietary Spirulina were significantly (P≤0.5) decreased as compared to control group during starter, finisher and the entire length of the experimental periods, where birds fed 0.25g Spirulina/kg diet significantly (P≤0.05) showed the lowest feed consumption and had the best FCR. Feed consumption of birds fed 0.25 and 0.15g Spirulina/kg diet represented 1034.37and 1064.24 gm. when compared with the control birds (1102.74gm.) during starter period. The corresponding values were 2400.78 and 2496.04 gm. during the finisher period and 3435.15 and 3560.27 gm. at entire period compared of the control group respectively. Additionally, FCR of birds fed 0.25 and 0.15g Spirulina/kg diet represented 1.58 and 1.69 when compared with the control birds 1.79 during starter period. The corresponding values were 1.83 and 2.10 during the finisher period and 1.73 and 1.93 at entire period compared to the control group respectively.

A significant improvement in feed conversion ratio was achieved by birds fed Spirulina diets may due in part to, an improvement in live body weight or the improvement of viability percentage. The present results agree with findings obtained by Kaoud, (2012) , Kharde *et al*., (2012) and Mariey *et al*., (2014) who reported that feed consumption and feed conversion ratio significantly (P<0.05) improved by dietary inclusion of *Spirulina platensis* as compared the control broilers.

The obtained results due to interaction between stocking density and Spirulina supplementation levels revealed that FC and FCR were improved significant (P≤0.05) when broiler chicks housed under stocking density of 10 birds/m^2 with adding Spirulina at 0.25g/kg diet feed At all experimental period and the entire experimental period .

Table (3): Average body weight gains (BG), feed consumption (FC), feed conversion ratio (FCR) of broiler chicks as affected by stocking density and *Spirulina platensis* supplementation levels and their interaction.

Treatment	Starter Period From 0 – 21 days of age			Finisher Period From 21– 42 days of age			Total Period From 0 – 42 days of age		
	WG	FC	FCR	WG	FC	FCR	WG	FC	FCR
Effect of Stocking Density									
Control (C) 10 bird/m^2	669.37^a	1022.89 ^{a}	1.53 ^{a}	1350.2 ^{a}	2413.78 ^{a}	1.80 ^{a}	2019.5 ^{a}	3436.67 ^{a}	1.70 ^{a}
12 bird/m^2	627.40^b	1075.39 ^{b}	1.72 ^{b}	1271.9 ^{b}	2531.36 ^{c}	1.99 ^{b}	1899.3 ^{b}	3606.75 ^{b}	1.89 ^{b}
14 bird/m^2	608.67^c	1103.07 ^{c}	1.81 ^{c}	1110.5 ^{c}	2520.18 ^{b}	2.27 ^{c}	1719.2 ^{c}	3623.25 ^{c}	2.11 ^{C}
SEM	±5.3	±8.1	±0.06	±6.8	±9.9	±0.05	±9.2	±11.9	±0.05
Effect of *Spirulina platensis* Supplementation Levels									
Control (C) 0.00 g/Kg	616.53 ^{a}	1102. 74 ^{a}	1.79 ^{c}	1187.9 ^{a}	2568.51 ^{a}	2.20 ^{c}	1804.5 ^{a}	3671.25 ^{a}	2.03 ^{c}
0.15 g/ Kg	632.37 ^{b}	1064.24 ^{b}	1.69 ^{b}	1213.8 ^{b}	2496.04 ^{b}	2.10 ^{b}	1846.2 ^{b}	3560.27 ^{b}	1.93 ^{b}
0.25 g/Kg	656.53 ^{c}	1034.37 ^{c}	1.58 ^{a}	1330.7 ^{c}	2400.78 ^{c}	1.83 ^{a}	1987.2 ^{c}	3435.15 ^{c}	1.73 ^{a}
SEM	±5.2	±8.6	±0.05	±7.3	±9.8	±0.04	±9.3	±11.8	±0.06
Effect of Interaction between Stocking Density and *Spirulina platensis* Supplementation									
0.00 g/Kg	650.2	1057.55	1.63	1275.4	2580.22	2.02	1925.6	3637.77	1.89
10 birds *	664.0	1020.11	1.54	1280.4	2430.15	1.89	1944.4	3450.26	1.77
0.15 g/Kg 0.25 g/Kg	693.9	991.00	1.43	1494.7	2230.98	1.49	2188.6	3221.98	1.47
0.00 g/Kg	606.3	1110.73	1.83	1222.6	2560.13	2.09	1828.9	3670.86	2.01
12 birds *	624.1	1070.88	1.72	1236.2	2552.71	2.06	1860.3	3623.59	1.95
0.15 g/Kg 0.25 g/Kg	652.2	1044.57	1.60	1356.8	2481.23	1.83	2008.6	3525.80	1.76
0.00 g/Kg	593.1	1139.94	1.92	1065.9	2565.18	2.41	1659.0	3705.12	2.23
14 birds *	609.0	1101.72	1.81	1124.8	2505.25	2.23	1733.8	3606.97	2.08
0.15 g/Kg 0.25 g/Kg	623.9	1067.54	1.71	1140.7	2490.12	2.18``	1764.6	3557.66	2.02
SEM	±5.0	±9.1	±8.9	±0.04	±9.5	±7.05	±0.04	±9.1	±5.0
Significant	*	*	*	*	*	*	*	*	*

-Means of each column followed by the same letter are not significantly different at the 5% level according to Duncan ,s Multiple Range Test. -SEM indicate standard error of means - indicate P<0.05*

Mortality rate

The effect of stocking density and Spirulina supplementation levels and there interaction on mortality rate percentages of broiler are represented in Table (4). The results showed that at all experimental periods, mortality rate percentages was significantly (P≤0.05) increased by increasing stocking density from 10, 12 and 14 birds/m^2. This result disagreed with the findings obtained by Thomas *et al*., (2004); Dozier *et al*., (2006) and Meluzzi *et al*., (2008) who reported that there was no relationship between stocking density and lesion incidence or mortality rate .

Mortality rate percentages of broiler chicks fed different levels of dietary Spirulina significantly (P≤0.5) decreased as compared to that fed control diet during starter, finisher and the entire length of the experimental periods, where mortality rate percentages of birds fed 0.25 and 0.15g Spirulina/kg diet represented 1.11 and 1.67 % when compared with the control birds during starter period. The corresponding values were 0.56 and 1.11% kg during the finisher period and 1.67 and 2.78% at marketing age compared of the control group respectively . In this regard Gružauskas, *et al.*, (2004) reported that Spirulina improve absorption of minerals, protect from diarrhea, and optimize nutrient digestion processes. Feeding Spirulina containing diets may increase the lactobacillus population and enhance the absorbability of dietary vitamins (Mariey *et al*., 2014). Also, the present results are supported by Belay, (1993) and Baojiang, (1994) who reported that Spirulina is good for the beneficial intestinal flora. Availability of free amino acids often limits the growth of *Lactobacillus* and *Bifidus*. Stimulating these good bacteria reduces problems with *E. coli* and *Candida albicans* .

The results due to interaction between stocking density and Spirulina supplementation on mortality rate percentages of broiler revealed that, mortality rate percentages were showed the highest significantly ($P \leq 0.05$) value when housed broiler by 10 birds/m^2 with adding Spirulina at 0.25g/kg diet feed during all experimental period and the entire experimental period.

Table (4): Effect of stocking density and *Spirulina platensis* supplementation levels on mortality rate.

Treatment	Starter Period From 0 – 21 days of age		Finisher Period From 21– 42 days of age		Total Period From 0 – 42 days of age	
	N	Mo%	N	Mo%	N	Mo%
Effect of Stocking Density						
Control (C) 10 bird/m^2	2	1.11 ^{a}	0	0.00 ^{a}	2	1.11 ^{a}
12 bird/m^2	3	1.67 ^{b}	2	1.11 ^{b}	5	2.78 ^{b}
14 bird/m^2	4	2.22 ^{c}	4	2.22 ^{c}	8	4.44 ^{c}
Effect of *Spirulina platensis* Supplementation Levels						
Control (C) 0.00 g/Kg	4	2.22 ^{c}	3	1.67^c	7	3.89^c
0.15 g/ Kg	3	1.67 ^{b}	2	1.11^b	5	2.78^b
0.25 g/Kg	2	1.11 ^{a}	1	0.56^a	3	1.67^a
Effect of Interaction between Stocking Density and *Spirulina platensis* Supplementation						
0.00 g/Kg	1	1.67	0	0.00	1	1.67
10 birds * 0.15 g/Kg	1	1.67	0	0.00	1	1.67
0.25 g/Kg	0	0.00	0	0.00	0	0.00
0.00 g/Kg	1	1.67	1	1.67	2	3.33
12 birds * 0.15 g/Kg	1	1.67	1	1.67	2	3.33
0.25 g/Kg	1	1.67	0	0.00	1	1.67
0.00 g/Kg	3	5.00	1	1.67	4	6.67
14 birds * 0.15 g/Kg	1	1.67	1	1.67	2	3.33
0.25 g/Kg	1	1.67	1	1.67	2	3.33
Significant		*		*		*

-Means of each column followed by the same letter are not significantly different at the 5% level according to Duncan ,s Multiple Range Test. *-* indicate P<0.05*

Carcass traits

The effect of stocking density and Spirulina supplementation levels and there interaction on carcass traits and lymphoid organs weight of broiler are represented in Tables (5 and 6). The results showed that, all relative carcass traits except abdominal fat% were insignificantly decreased by increasing stocking density from 10, 12 and 14 birds/m^2. These results fairly agree with finding by Cravener *et al*., (1992), Feddes *et al.*, (2002), Dozier *et al*., (2005) who reported that carcass weight of chicks decreased as the level of stocking density increased, but did not influence on other parameters .

The results showed that, dietary Spirulina insignificantly increased absolute weight of carcass, giblets and total edible part compared with the control group. In line with the present results, several authors reported that body weights, weight gain, carcass yield percentage, and feed conversion ratio were increased by dietary inclusion of *Spirulina platensis* as compared to the control diet (Ross and Dominy, 1990; Nikodémusz *et al*., 2010; Kaoud, 2012; Kharde *et al*., 2012 and Mariey *et al*., 2014). It is of interest to note that relative abdominal fat weight (Table 5) was reduced by increasing dietary treatments level. This reduction may be attributed to the reduction in plasma total lipids and cholesterol. In this respect, khan et al., (2005), reported that Spirulina have shown regulatory role on lipid and carbohydrate metabolism by exhibiting glucose and lipid profile correcting activity in animals.

The results due to interaction between stocking density and Spirulina supplementation on carcass traits presented of broiler revealed carcass traits presented were showed the highest value when housed broiler by 10 birds/m^2 with adding Spirulina at 0.25g/kg diet feed at all experimental period and the entire experimental period.

The effect of stocking density and Spirulina supplementation and their interaction on lymphoid organs weight of broiler chicks are presented in Table (6). The results showed that at the end of experimental period, both of absolute and relative lymphoid organs weight of broiler chicks were significantly (P≤0.05) decreased by increasing stocking density from 10, 12 and 14 birds/m^2.

Results showed that absolute and relative weights of bursa and thymus significantly (P≤0.05) increased for all treatment groups compared with the control one. While, weight of spleen had a significant increase only in absolute weight. In accordance with the present results, Kaoud, (2012) reported that the absolute and relative weights of bursa and thymus was increased for all groups fed dietary Spirulina compared with the control group .

These results may be used as an indicator of good health status of chicks fed dietary Spirulina. In this respect Bennett and Stephens (2006) reported that the bursa functions are half of the bird's immune system and the size of the bursa reflects the bird's overall health status. Sick or stressed birds have small bursa while, healthy, productive birds have large bursa. Bursa size is a biological measure of how well flocks are managed and protected from disease. Also, liver weight (Table 5) may indicate that anti-nutritional factors presented in *Spirulina platensis* were not effective. These results were supported by the findings of several scientists (Belay, 1993 and Baojiang, 1994), who reported that Spirulina-polysaccharide acts similarly to phycocayanin. It improves the immune system's ability to detect and destroy foreign microbes or eliminate toxins. It also enhances T-cells and improves thymus gland function. In agreement with the present results, Qureshi *et al*., (1994) reported that dietary Spirulina could increase size of thymus glands for greater T-cell production of broiler chocks. Also, Kaoud, (2012) reported that, the absolute and relative weights of bursa and thymus was increased for all groups fed dietary Spirulina compared with the control group.

Table (5): Effect of stocking density and *Spirulina platensis* supplementation levels on some carcass traits of broiler chicks.

Treatment	% Carcass	% Liver	%Heart	%Gizzard	%Giblets	%Abd. Fat
Effect of Stocking Density						
Control (C) 10 bird/m^2	63.81	2.478	0.68	2.74	5.901	2.71
12 bird/m2	62.64	2.479	0.65	2.73	5.855	2.88
14 bird/m^2	62.03	2.486	0.64	2.72	5.845	3.11
SEM	±1.37	±0.007	±0.009	±0.069	±0.055	±0.04
Effect of *Spirulina platensis* Supplementation Levels						
Control (C) 0.00 g/Kg	62.30	2.481	0.64	2.74	5.864	3.17
0.15 g/ Kg	62.81	2.483	0.65	2.72	5.857	2.88
0.25 g/Kg	63.37	2.481	0.68	2.72	5.881	2.64
SEM	±1.37	±0.004	±0.009	±0.025	±0.062	±0.04
Effect of Interaction between Stocking Density and *Spirulina platensis* Supplementation						
0.00 g/Kg	63.12	2.480	0.65	2.75	5.880	2.99
10 birds * 0.15 g/Kg	63.61	2.477	0.67	2.74	5.887	2.81
0.25 g/Kg	64.70	2.476	0.72	2.74	5.936	2.34
0.00 g/Kg	62.13	2.480	0.64	2.75	5.870	3.10
12 birds * 0.15 g/Kg	62.65	2.476	0.65	2.72	5.846	2.87
0.25 g/Kg	63.15	2.480	0.66	2.71	5.850	2.66
0.00 g/Kg	61.66	2.483	0.63	2.73	5.843	3.41
14 birds * 0.15 g/Kg	62.16	2.487	0.64	2.71	5.837	2.95
0.25 g/Kg	62.27	2.486	0.65	2.72	5.856	2.93
SEM	±1.98	±0.062	±0.099	±0.097	±0.92	±0.22
Significant	NS	NS	NS	NS	NS	NS

-Means of each column followed by the same letter are not significantly different at the 5% level according to Duncan ,s Multiple Range Test. -SEM indicate standard error of means -NS indicate not significant.

Significant (P≤0.05) level of stocking density (10 birds/m^2) and Spirulina supplementation (0.25g/kg diet) interaction was observed for body weight and percentages at all experimental period.

Table (6): Effect of stocking density and *Spirulina platensis* supplementation levels on lymphoid organs weight of broiler chicks.

Treatment	Bursa		Thymus		Spleen	
	W	%	W	%	W	%
Effect of Stocking Density						
Control (C) 10 bird/m^2	4.816 ^{a}	0.233 ^{a}	11.156 ^{a}	0.540 ^{a}	3.138 ^{a}	0.152
12 bird/m^2	4.444 ^{b}	0.228 ^{b}	10.399 ^{b}	0.535 ^{a}	2.867 ^{b}	0.148
14 bird/m^2	3.756 ^{c}	0.225 ^{c}	08.844 ^{c}	0.531 ^{a}	2.462 ^{c}	0.148
SEM	±0.102	±0.005	±0.388	±0.087	±0.061	±0.011
Effect of *Spirulina platensis* Supplementation Levels						
Control (C) 0.00 g/Kg	3.533 ^{c}	0.195 ^{a}	9.279 ^{c}	0.512 ^{a}	2.636 ^{b}	0.145
0.15 g/ Kg	4.489 ^{b}	0.242 ^{b}	9.892 ^{b}	0.534 ^{b}	2.769 ^{b}	0.149
0.25 g/Kg	4.995 ^{a}	0.250 ^{c}	11.23 ^{a}	0.561 ^{c}	3.063 ^{a}	0.153
SEM	±0.102	±0.003	±0.387	±0.087	±0.066	±0.011
Effect of Interaction between Stocking Density and *Spirulina platensis* Supplementation						
0.00 g/Kg	3.877	0.197	10.095	0.513	2.873	0.146
10 birds * 0.15 g/Kg	4.884	0.246	10.642	0.536	3.018	0.152
0.25 g/Kg	5.686	0.255	12.732	0.571	3.523	0.158
0.00 g/Kg	3.650	0.195	9.566	0.511	2.714	0.145
12 birds * 0.15 g/Kg	4.599	0.242	10.131	0.533	2.813	0.148
0.25 g/Kg	5.084	0.248	11.501	0.561	3.075	0.150
0.00 g/Kg	3.072	0.192	8.176	0.511	2.320	0.145
14 birds * 0.15 g/Kg	3.983	0.238	8.903	0.532	2.477	0.148
0.25 g/Kg	4.214	0.246	9.452	0.551	2.590	0.151
SEM	±0.133	±0.043	±0.402	±0.099	±0.122	±0.018
Significant	*	*	*	*	*	NS

-Means of each column followed by the same letter are not significantly different at the 5% level according to Duncan ,s Multiple Range Test. -SEM indicate standard error of means -NS indicate not significant.
- indicate P<0.05*

Blood parameters:

Results presented in Table (7) showed the concentrations of some blood metabolites such: total lipids, triglycerides, cholesterol, total protein, albumin and globulin. All of determined parameters were significantly (P≤ 0.05) increased by increasing stocking density from 10, 12 and 14 birds/m^2 respectively. On the other hand, concentrations of total lipids, triglycerides and cholesterol in blood plasma significantly (P≤ 0.05) decreased by inclusion Spirulina in diets of birds, in particular those contained 0.15 or 0.25 g/kg diet. While, the concentrations of total protein and their fraction were significantly (P≤ 0.05) increased by Spirulina supplementation level increased from 0 to 0.25 g/kg diet.

In accordance with the present results, Torres-Duran *et al.*, (1998) and Fong *et al*., (2000) reported significant reduction of cholesterol and triglyceride concentration for rats or mice fed Spirulina diets. The significant reduction in plasma cholesterol of broiler chickens fed dietary Spirulina could be attributed to reducing the absorption and/or synthesis of cholesterol in the gastro-intestinal tract by Spirulina supplementation that increase *Lactobacillus* population (Tsuchihashi *et al*., 1987 and Mariey, *et al*., 2012). *Lactobacillus* has found to have a high bile salt hydrolytic activity and to reduce the cholesterol in the blood by deconjugating bile salts in the intestine (Surono, 2003), thereby preventing them from acting as precursors in cholesterol synthesis (Abdulrahim *et al*., 1996). Also, Mariey *et al*., (2014) reported that dietary Spirulina supplementation at levels of 0.2 and 0.3 g /kg diet significantly (P<0.05) increased concentration of total proteins, albumin and globulin in blood plasma of birds compared with the control diet. The observed increase in concentration of plasma total protein, albumin and globulin may be related to the high level and good quality of protein contents in Spirulina (55 - 65%). These results are in line with the findings of Tewe, (1985) and Eggum, (1989), who stated that total serum protein, globulin and albumin were directly responsive to protein quantity and quality.

Effect of interaction between Spirulina supplementation and stocking density was listed in Table (7). The best values for blood metabolites parameters were observed in birds fed diet containing 0.25g/kg diet at 10 birds/m^2 stocking density. Whereas, the concentrations of total lipids, triglycerides, cholesterol, total protein, albumin and globulin were improved significantly (P<0.05) when housed broiler at 10 birds/m^2 with adding Spirulina at 0.25g/kg diet compared to other groups..

Table (7): Effect of stocking density and *Spirulina platensis* supplementation levels on some blood constituents of broiler chicks.

Treatment	Total lipids mg/dl	Triglyceride mg/dl	Cholesterol mg/dl	Total protein mg/dl	Albumin mg/dl	Globulin mg/dl
Effect of Stocking Density						
Control (C) 10 bird/m^2	306.7[a]	65.4[b]	123.55[b]	5.22[a]	3.73[a]	1.49
12 bird/m^2	353.7[b]	79.1[a]	140.76[a]	4.55[b]	3.25[b]	1.30
14 bird/m^2	406.6[c]	80.8[a]	142.66[a]	3.86[c]	2.58[c]	1.28
SEM	±3.21	±2.45	±2.44	0.066±	±0.07	0.075
Effect of *Spirulina platensis* Supplementation Levels						
Control (C) 0.00 g/Kg	434.8[a]	87.0[a]	148.56[a]	3.87[c]	2.69[c]	1.17[b]
0.15 g/ Kg	347.9[b]	72.4[b]	135.15[b]	4.64[b]	3.27[b]	1.37[ab]
0.25 g/Kg	284.4[c]	65.9[b]	123.28[c]	5.13[a]	3.60[a]	1.53[a]
SEM	±3.22	±2.49	±2.44	±0.066	±0.09	0.075
Effect of Interaction between Stocking Density and *Spirulina platensis* Supplementation						
0.00 g/Kg	408.2	80.1	141.34	4.16	3.02	1.14
10 birds * 0.15 g/Kg	290.4	61.6	126.31	5.10	3.57	1.53
0.25 g/Kg	221.5	54.5	103.01	6.41	4.60	1.81
0.00 g/Kg	427.3	88.0	151.33	3.92	2.73	1.19
12 birds * 0.15 g/Kg	352.1	75.4	140.80	4.61	3.31	1.30
0.25 g/Kg	281.7	74.0	130.16	5.13	3.72	1.41
0.00 g/Kg	468.8	93.0	153.00	3.52	2.33	1.19
14 birds * 0.15 g/Kg	401.1	80.2	138.33	4.21	2.93	1.28
0.25 g/Kg	350.0	69.1	136.66	3.86	2.48	1.38
SEM	±4.02	±3.51	3.05±	±0.12	±0.77	0.13
Significant	*	*	*	*	*	*

-Means of each column followed by the same letter are not significantly different at the 5% level according to Duncan ,s Multiple Range Test. -SEM indicate standard error of means - indicate P<0.05*

Economical efficiency:

The effects of stocking density and Spirulina supplementation and their interaction on economic efficiency of broiler chicks at 42 days of age are presented in Table (8). The results revealed that decreasing stocking density to 10 birds/m^2 showed the highest value of economic efficiency and relative economic efficiency. Spirulina supplementation at level 0.25 g/kg and 0.15 g/kg diet fed showed the highest value of economic efficiency and relative economic efficiency (116.10 and 101.46) respectively. The highest value of economic efficiency and relative economic efficiency was observed for interaction between stocking density at level 10 birds/m^2 and Spirulina supplementation at 0.25 and 0.15 g/kg diet.

CONCLUSION

Regarding to the effect of stocking density and spirulina supplementation level on broiler performance, physiological status and economical aspect, 10 birds/m2 stocking density and *Spirulina platensis* algae could be safely used in broiler feeding, at level of 0.25 g /kg diet with superior effects on their productive performance. Additionally, the current study revealed the ability to produce a safety broiler meat for consumer by using natural substances away from antibiotics or other chemicals

Table (8): Effect of stocking density and *Spirulina platensis* supplementation levels on economic efficiency.

Treatment	Total FC g/chick	Pric/kg (L.E)	Spirulina Cost (L.E)	Total feed cost (L.E)	Weight gain	Pric/kg (L.E)	Total Revenue (L.E)	Net Revenue (L.E)	Economic efficiency	Relative Economic efficiency (%)
Effect of Stocking Density										
Control (C) 10 bird/m2	3436.67	3.2	0.44	11.44	2019.5	15.2	30.69	19.25	168.27	100
12 bird/m2	3606.75	3.2	0.47	12.01	1899.3	15.2	28.87	16.86	140.38	83.52
14 bird/m2	3623.25	3.2	0.48	12.07	1719.1	15.2	26.13	14.06	116.49	69.23
Effect of *Spirulina platensis* Supplementation Levels										
Control (C) 0.00 g/Kg	3671.25	3.2	0.00	11.75	1804.5	15.2	27.43	15.68	133.45	100
0.15 g/ Kg	3560.27	3.2	0.53	11.92	1846.2	15.2	28.06	16.14	135.40	101.46
0.25 g/Kg	3435.15	3.2	0.86	11.85	1987.3	15.2	30.21	18.36	154.94	116.10
Effect of Interaction between Stocking Density and *Spirulina platensis* Supplementation										
0.00 g/Kg	3637.77	3.2	0.00	11.64	1925.6	15.2	29.27	17.63	151.46	100
10 birds *	3450.26	3.2	0.52	11.56	1944.4	15.2	29.55	17.99	155.62	102.75
0.15 g/Kg 0.25 g/Kg	3221.98	3.2	0.81	11.12	2188.6	15.2	33.27	22.15	199.19	131.51
0.00 g/Kg	3670.86	3.2	0.00	11.75	1828.9	15.2	27.79	16.04	136.51	100
12 birds *	3623.59	3.2	0.54	12.14	1860.3	15.2	28.28	16.14	132.95	97.39
0.15 g/Kg 0.25 g/Kg	3525.80	3.2	0.88	12.16	2008.6	15.2	30.53	18.37	151.07	110.67
0.00 g/Kg	3705.12	3.2	0.00	11.86	1659.0	15.2	25.22	13.36	112.65	100
14 birds *	3606.97	3.2	0.54	12.08	1733.8	15.2	26.35	14.27	118.13	104.86
0.15 g/Kg 0.25 g/Kg	3557.66	3.2	0.89	12.27	1764.6	15.2	6.82	14.55	118.58	105.26

Net revenue = Price of Wight gain / chick – feed cost.
Economic efficiency = net revenue / feed cost × 100
Relative economic efficiency (%) assuming the Control treatments = 100 %

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تأثير الكثافة العددية وإضافة طحلب الاسبيريولينا بلاتينسيز كإضافة علفية علي الأداء الانتاجي والحالة الفسيولوجية لدجاج التسمين

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أجريت هذه التجربة لدراسة التأثيرات النافعة لطحلب الاسبيريولينا كإضافة علفية تحت مستويات مختلفة من الكثافة العددية علي الاداء الانتاجي والحالة الفسيولوجية والكفاءة الاقتصادية لدجاج التسمين عند عمر 6 أسابيع. استخدم في هذه الدراسة عدد 540 كتكوت غير مجنس عمر يوم واحد من سلالة الروس وقسمت الي 9 مجموعات ناتجة من تجربة عاملية 3*3 بكل معاملة 60 كتكوت. وزعت الطيور علي 3 مجموعات طبقا لمستوي الكثافة العددية (10 ، 12 ، 14 طائر/ م2). كل مجموعه من المجموعات الثلاثة السابقة قسمت داخليا الي 3 مجموعات فرعية طبقا لمعدلات اضافة طحلب الاسبيريولينا إلي العلائق (صفر : 0.15 ، 0.25 جم/ كجم عليقه). تم تسجيل ودراسة وزن الجسم ، معدل الزيادة في وزن الجسم ، معدل استهلاك العلف ، الكفاءة التحويلية ، نسبة النفوق ، قياسات الذبيحة ، بعض قياس الدم البيوكيميائية بالإضافة إلي الكفاءة الاقتصادية علي مدار فترة التجربة. بينت النتائج أن جميع القياسات التي تم تقديرها كانت أفضل معنويا ($P \leq 0.05$) عند مستوي كثافة 10 طيور / م2 مقارنة بباقي الكثافات العددية ، تم ملاحظة نفس الاتجاه مع الطيور التي غذيت علي عليقة بها 0.25جم طحلب اسبيريولينا / كجم عليقه. كذلك فان هناك تحسن بنسبة كبيرة في معدل الاداء ومقاييس الحالة الفسيولوجية والكفاءة الاقتصادية نتيجة للتداخل بين الكثافة العددية (10 طيور/ م2) ومعدل اضافة الاسبيريولينا (0.25جم /كجم). وعلي ذلك فان اضافة طحلب الاسبيريولينا عند مستوي 0.25جم/كجم عليقة تحت كثافة عددية 10 طيور / م2 تعتبر آمنه ولها تأثيرات إيجابية علي كلا من الكفاءة الانتاجية والحالة الفسيولوجية والكفاءة الإقتصادية لدجاج التسمين .

الكلمات الدالة: *الكثافة العددية ،اسبيريولينا بلاتنسيز ، الأداء الانتاجي ، ، خصائص الذبيحه ،الحالة الفسيولوجية.*