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INVESTIGATION INTO GROWTH DYNAMICS AND PHYSIOLOGICAL CHANGES IN BROILER CHICKENS FED WITH *SPIRULINA PLATENSIS* ALGAE-SUPPLEMENTED DIET

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Abstract

Spirulina platensis is a good candidate as an in-feed antibiotics substitute for broilers. However, its use seems impractical owing to its high price, especially when being administered throughout the whole rearing period. Spirulina platensis is a good candidate as an in-feed antibiotics substitute for broilers. However, its use seems impractical owing to its high price, especially when being administered throughout the whole rearing period. The prohibition of feed grade antibiotic growth promoters and rising demand for sustainable protein sources have necessitated the exploration of new feed ingredients that offer a safe and nutritious alternative for poultry. This study aimed to assess the impact of dietary supplementation with spirulina on the performance, hematological parameters, and serum profiles of broiler chickens. Four hundred 1-d-old Broiler chicks were randomly assigned to 1 of 4 dietary treatments, consisting of 2 replicates of 50 birds each. Commercial mash starter and finisher diets were supplemented with 0.5%, 1%, and 1.5% of *Spirulina platensis.* The Body Weights, Feed Conversion Rate, Villi length, Carcass yield percentage were significantly increased by the dietary inclusion of the 1% of *Spirulina platensis* as compared to the control fed broilers. In conclusion, 1% of *Spirulina platensis* supplementation significantly increased Body weight, decreased Feed Consumption Ratios and increases the villi height. The *Spirulina platensis* offers a good alternative to improve poultry production.

Keywords: *Spirulina platensis* - Performance- Broilers – Histopathology.

INTRODUCTION

The growing requirement for food sources high in human protein has led to a need for new feed materials that give poultry and animals a safe source of nutrients. Additionally, consumers are putting more and more pressure on producers of animal feed, particularly poultry feed, to cut back on the usage of antibiotic growth promoters as feed additives and develop alternatives for poultry diets (Humphrey, B.D., N. Huang and K. C,Klasing, 2002). Growing consumer demand for human protein sources has spurred a necessity for novel feed ingredients that ensure safe nutrient provision for poultry and livestock. Poultry feed manufacturers, particularly, face mounting pressure from consumers to minimize the use of antibiotic growth promoters in feed additives and explore viable alternatives for poultry diets. The intestinal epithelium functions naturally as a barrier, safeguarding against pathogenic bacteria and toxins present in the intestinal lumen. The normal microflora or the intestinal epithelium are disturbed by a variety of factors, including pathogens, chemicals, and stresses. Such disruptions can increase the permeability of this natural barrier, allowing pathogens and detrimental substances to penetrate more easily.

These disruptions can alter metabolism, impair the body's capacity to digest and absorb nutrients, and provoke prolonged inflammatory responses in the intestinal mucosa. (Hofstad, 1972; Podolsky, 1993; Oliveira, 1998). As a consequence, the villus diminishes in size, cell turnover rates rise, and the effectiveness of digestion and absorption lessens. (Visek, 1978).

The effects of herbs and plant extracts include altering appetite and intestinal microflora, promoting pancreatic secretions to increase endogenous enzyme activity, and stimulating the immune system (Jamil *et al,* 2015). In the last decade, the beneficial nutritional properties of microalgae have been extensively advertised worldwide, resulting in a growing interest among producers in algae businesses (Fathi et al., 2018).

The beneficial effects of probiotics are explained by mechanisms like the production of antimicrobial substances and organic acids, protection of villi and absorptive surfaces from pathogenic toxins, and immune system stimulation (Shanmugapriya et al., 2024; Dobrogosz et al., 1991; Ewing & Cole, 1994; Walker & Duff, 1998; Pelicano et al., 2002). The histopathological findings suggest that the broilers' overall immunity has increased. Consequently, supplementing their diet with *Spirulina platensis* at 0.5% may be a valuable natural feed additive, boosting productivity (Samar H. Abdelfatah et al., 2024)

As a high-quality natural feed additive, Spirulina (blue-green alga) provides significant benefits in animal and poultry nutrition, with protein levels between 55-65% and a complete profile of essential amino acids (Nikodémusz et al., 2010). Research has demonstrated that hens on Spirulina-supplemented diets show enhanced productive performance compared to control hens. Additionally, the inclusion of *Spirulina platensis* in the diet has been associated with better feed conversion, increased body weight gain, and improved carcass yield (Kharde et al., 2012).

On the other hand, prebiotics effects are based on reduction of the growth of many pathogenic or nonpathogenic intestinal bacteria by means of the pH reduction that results from increased lactic acid levels in the ceca (Choi et al., 1994). Some bacteria may recognize binding sites in such molecules as if they were on the mucosa surface, and the intestinal colonization by pathogenic bacteria is thus reduced.

Therefore, there is lower incidence of infectious processes, and the functions of secretion, digestion and absorption of nutrients can be appropriately performed by the mucosa (Iji & Tivey, 1998). The present study evaluated the effects of different levels of *Spirulina platensis* and their association on the histological of the intestinal mucosa of broilers.

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MATERIALS AND METHODS

Four hundred day-old male broiler chicks (Ross) assigned to 50 chicks of 2 treatment groups, randomly. The experimental design was completely random, consisting of three dietary levels (0.5%, 1% and 1.5%) of *Spirulina platensis* and a control group (without *Spirulina platensis*) were formulated. Each treatment had two replicates of 25 birds. Chicks fed three basal of Maize-soybean diets during three periods of 0-10 days birds fed with broiler Pre-starter, 11-20 birds fed with broiler Starter I, 21–30 days birds fed with broiler Starter II, 31-36 days birds fed with broiler Finisher. The diets supplemented with amino-acids, minerals, and vitamins to meet all the Ross requirements.

Body Weight and Feed Intake Measurement:

Birds were group weighed by cage at 1, 10, 20, 30 and 36 d of age. Feed intake was monitored by cage at 10, 20, 30 and 36 d of age. Cage was the experimental unit for performance was used to calculate feed/gain ratios.

Table 1: Ingredients Composition (%) of Experimental Pre-Broiler Starter rations

Ingredients	Control (without *Spirulina platensis*)	*Spirulina platensis* @ 0.5%	*Spirulina platensis* @ 1%	*Spirulina platensis* @ 1.5%
Maize	48.33	48.33	48.33	48.33
Soya DOC 48%	35.45	35.45	35.45	35.45
Crushed fish 45%	6	6	6	6
Meat-cum-Bone Meal	4	4	4	4
Ricebran oil	3.6	3.6	3.6	3.6
Di-calcium Phosphate	0.05	0.05	0.05	0.05
DL-Methionine	0.29	0.29	0.29	0.29
L-Lysine HCl	0.9	0.9	0.9	0.9
L-Threonine	0.1	0.1	0.1	0.1
Sodium bi-carbonate	0.1	0.1	0.1	0.1
Salt	0.07	0.07	0.07	0.07
Choline chloride 60%	0.15	0.15	0.15	0.15
Additives	0.96	0.96	0.96	0.96
Spirulina platensis	Nil	0.005	0.01	0.015
Total	**100**	**100**	**100**	**100**

Table 2: Ingredients composition (%) of experimental Broiler Starter-I rations

Ingredients	Control (without *Spirulina platensis*)	*Spirulina platensis* @ 0.5%	*Spirulina platensis* @ 1%	*Spirulina platensis* @ 1.5%
Maize	55.4	55.4	55.4	55.4
Hypo Soya DOC	29.52	29.52	29.52	29.52
Crushed fish 45%	6	6	6	6
Meat-cum-Bone Meal	4	4	4	4
Ricebran oil	2.9	2.9	2.9	2.9
Di-calcium Phosphate	0.4	0.4	0.4	0.4
DL-Methionine	0.26	0.26	0.26	0.26
L-Lysine HCl	0.1	0.1	0.1	0.1

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L-Threonine	0.03	0.03	0.03	0.03
Sodium bi-carbonate	0.2	0.2	0.2	0.2
Choline chloride 60%	0.17	0.17	0.17	0.17
Salt	0.09	0.09	0.09	0.09
Additives	0.91	0.91	0.91	0.91
Spirulina platensis	Nil	0.005	0.01	0.015
Total	**100**	**100**	**100**	**100**

Table 3: Ingredients Composition (%) of experimental Broiler Starter-II rations

Ingredients	Control (without *Spirulina platensis*)	*Spirulina platensis* @ 0.5%	*Spirulina platensis* @ 1%	*Spirulina platensis* @ 1.5%
Maize	59.54	59.54	59.54	59.54
Hypo Soya DOC	25.3	25.3	25.3	25.3
Crushed fish 45%	4.8	4.8	4.8	4.8
Meat-cum-Bone Meal	4.5	4.5	4.5	4.5
Ricebran oil	3.86	3.86	3.86	3.86
Di-calcium Phosphate	0.2	0.2	0.2	0.2
DL-Methionine	0.3	0.3	0.3	0.3
L-Lysine HCl	0.11	0.11	0.11	0.11
L-Threonine	0.03	0.03	0.03	0.03
Sodium bi-carbonate	0.2	0.2	0.2	0.2
Choline chloride 60%	0.17	0.17	0.17	0.17
Salt	0.08	0.08	0.08	0.08
Additives	0.91	0.91	0.91	0.91
Spirulina platensis	Nil	0.005	0.01	0.015
Total	**100**	**100**	**100**	**100**

Table 4: Ingredients Composition (%) of Experimental Broiler Finisher rations

Incredients	Control (without *Spirulina platensis*)	*Spirulina platensis* @ 0.5%	*Spirulina platensis* @ 1%	*Spirulina platensis* @ 1.5%
Maize	57.00	57.00	57.00	57.00
Ricebran oil	5.90	5.90	5.90	5.90
Soya DOC 48%	20.50	20.50	20.50	20.50
Meat-cum-Bone Meal	5.00	5.00	5.00	5.00
Crushed fish 45%	10.00	10.00	10.00	10.00
DL-Methionine	0.26	0.26	0.26	0.26
L-Lysine HCl	0.12	0.12	0.12	0.12
L-Threonine	0.03	0.03	0.03	0.03
Sodium bi-carbonate	0.20	0.20	0.20	0.20
Salt	0.19	0.19	0.19	0.19
Choline Chloride 60%	0.17	0.17	0.17	0.17
Additives	0.62	0.62	0.62	0.62
Spirulina platensis	Nil	0.005	0.010	0.015
Total	100	100	100	100

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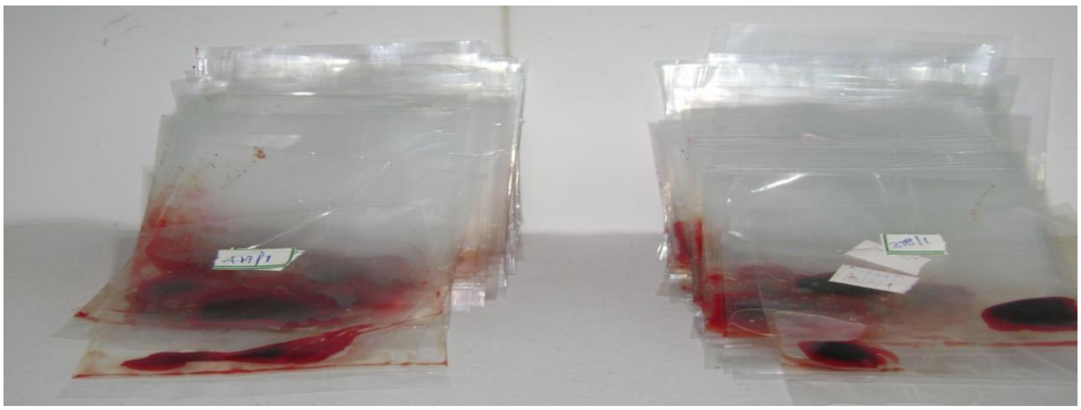
Body Weight and Feed Consumption

Chicks were weighed individually and the feed consumption was measured 10 days once of experimental period. Cumulative Weight Gain and Feed Consumption were determined, from which, once in 10 days and cumulative Feed Conversion Ratio was calculated.

Sample Collection



Tissue Sampling and Measurement of villus height:

Tissue Sampling

On day 36, 6 chicks from each treatment were killed by cervical dislocation for measurement of intestinal villus height by method of Sun *et al.,* (2005). Five centimeter a section of jejunum (medial portion posterior to the bile ducts and anterior to Meckel's diverticulum) was removed, rinsed in Tris-buffered saline, cut into 5 equal pieces, and and fixed in 10% neutral buffered formalin. Each intestinal pieces was subsequently cut into 5-mm sections and placed into tissue cassettes. Cassettes were embedded in paraffin, cut into thicknesses of 5µm, and mounted onto slides. Tissue slides were stained using hematoxylin and eosin. Sun *et al.,* (2005)

Measurement of Villus Height

In the jejunum (4 sections for each segment per bird), the villus height was measured from the villus tip to the bottom, not including the intestinal crypt. The measurement was done with the Scion Image Program (Scion Corporations, Frederick, MD). The mean villus heights from 15 birds were expressed as a mean villus height for 1 treatment group. Zentek *et al.,* (2002)

Carcass Characteristics:

At the end of the experimental period, 10 chicks from each treatment were randomly selected weighed, slaughter and dressed to determine the carcass weight and liver, gizzard, breast muscle, fat.

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Statistical Analysis

All data were analyzed by analysis of variance (ANOVA) procedures (Steel and Torrie, 1980) appropriate for a completely randomized design by the GLM procedure of SAS (1995). When the effect of *Spirulina platensis* on performance, blood constituents was the main effect. The level of statistical significance was preset at P_0.05.

RESULTS AND DISCUSSION

Growth Performance:

The present study showed Body Weight Gain (Table 5), Feed Intake (Table 6) and Feed Conversion Ratio (Table 7) of broiler chicks fed different levels of *Spirulina platensis* at 10^{th}, 20^{th}, 30^{th} and 36^{th} days of age. Results showed that chicks fed with 1% of *Spirulina platensis* had the higher body weight gain and improved feed conversion ratio compared with control group or other dietary treatments. The 1% of *Spirulina platensis* (Table 5), - supplemented group had a greater Body weight gain (2162.14 ± 11.27a) compared with control birds (1847.32 ± 94.94b), Feed Intake (Table 6) was lower for birds supplemented with 1% *Spirulina platensis* (3207 ±20.6ab) compared with the control (3497.92 ±24.05b). Feed Conversion Rate (Table 7) was lower for birds supplemented with 1% *Spirulina platensis* (1.716 ±0.09a) than control birds (1.864 ±0.01a), The obtained results confirmed the previous findings of several researchers Razafindrajaona *et al.,* (2008), Also in agreement with our study, Toyomizu *et al.,* (2001) reported that *Spirulina platensis* confirmed these results when *Spirulina* was introduced at the rates of 40 and 80g/kg in broiler diets. Birds feeding with *Spirulina platensis* shows significant at (P<0.01). This result was agreed with Hussein and Kaoud (2012). However,Ross and Dominy (1990), Mariey *et al.,* (2012) and Nikodémusz et al. (2010) reported that birds fed dietary *Spirulina* had benefit effects on productive performance.

Table 5: Effect of Growth Performance on Broiler Chicks fed with *Spirulina platensis* (g)

	10^{th} day	20^{th} day	30^{th} day	36^{th} day
T1	271.44±14.126a	794.7 ±41.12a	1468.62 ±75.67b	1847.32 ±94.94b
T2	307.04 ±4.03a	870.5 ±5.70a	1671.34 ±3.97a	2065.86 ±10.16a
T3	310.06 ±2.85a	908.92 ±5.26a	1715.34 ±7.68a	2162.14 ±11.27a
T4	393.70 ±13.95a	804.5 ±49.83a	1510.08 ±91.01b	1921.82±116.16b
Trt	892.06**			
Days	10.59**			
Txd	1.33 ns			

(T1-Control, T2-5g/kg of *Spirulina,* T3-1g/kg of *Spirulina,* T2-15g/kg of *Spirulina*

**and ns, Significant at P< 0.01 and not significant respectively.

Mean in a column followed by a same letter (s) are not significantly (P<0.05) different according to Duncan's Multiple Range Test. # Mean ±S.E

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Table 6: Effect of Feed Intake on Broiler Chicks fed with *Spirulina platensis* (g):

	10th day	20th day	30th day	36th day
T1	319.24±13.14a	1154.60 ±6.57a	2362.56 ±12.11b	3497.92 ±24.05b
T2	343.0 28.21a	1238 ±6.9a	2682 ±10.47a	3465 ±23.75a
T3	318 ±3.99a	1144.96 ±96a	2530 ±15.9ab	3207 ±20.6ab
T4	324.28 ±4.63a	1184.649±17.25a	2595.84 ±26.01ab	3260.80±96.64ab
Trt	985.28**			
Days	2.85**			
Txd	<1			

(T1-Control, T2-5g/kg of *Spirulina,* T3-1g/kg of *Spirulina,* T2-15g/kg of *Spirulina*

** and ns, Significant at P< 0.01 and not significant respectively.

Mean in a column followed by a same letter (s) are not significantly (P<0.05) different according to Duncan's Multiple Range Test. # Mean ±S.E

Table 7: Effect on Feed Conversion Rate on Broiler Chicks fed with *Spirulina platensis*:

	10th day	20th day	30th day	36th day
T1	1.157±0.05a	1.420 ±0.06a	1.582 ±0.05a	1.864 ±0.01a
T2	1.110± 0.01a	1.416 ±0.04a	1.604 ±0.01a	1.868 ±0.09a
T3	1.022 ±0.01a	1.256 ±96b	1.472 ±0.09a	1.716 ±0.09a
T4	1.085 ±0.06a	1.455±0.07a	1.575 ±0.09a	1.897±0.07ab
Trt	70.32**			
Days	5.47**			
Txd	<1			

(T1-Control, T2-5g/kg of *Spirulina,* T3-1g/kg of *Spirulina,* T2-15g/kg of *Spirulina*

**and ns, Significant at P< 0.01 and not significant respectively.

Mean in a column followed by a same letter (s) are not significantly (P<0.05) different according to Duncan's Multiple Range Test. # Mean ±S.E

Histopathology

Effects of dietary Spirulina supplementation on villi height is presented in Table 8. Feeding dietary Spirulina had a significant (P<0.01) increase in the height of villi as Spirulina levels in diets increased up to 1%. The increased height of villi length may be related to the high protein contents in Spirulina (with values ranging from 55-65% and includes all of the essential amino acids).

Table 8: Effect of Supplementation of Different Levels of *Spirulina platensis* on Villi Length of Broiler Chicks on Day 15, 22 and 36.

	control	0.5%	1%	1.5%
Day 36th	270.9548±14.363c	331.6767±14.78b	362.0058±14.53a	306.2682±4.84b
Treatment X days		16.16**		

** Significant at P< 0.01. Mean in a column followed by a same letter (s) are not significantly (P<0.05) different according to Duncan's Multiple Range Test. # Mean ±S.E

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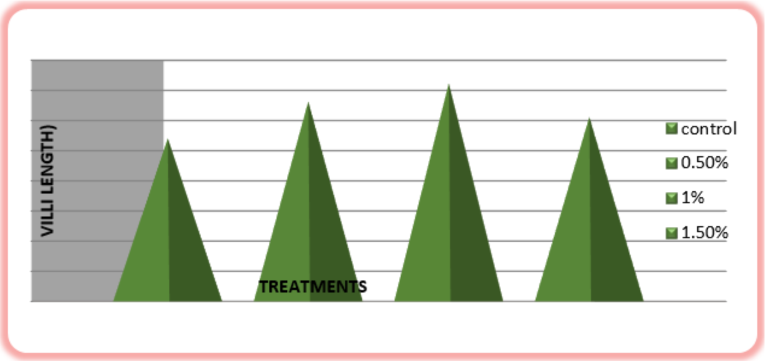

Plate 1: Control Plate 2: 0.5% *Spirulina platensis*

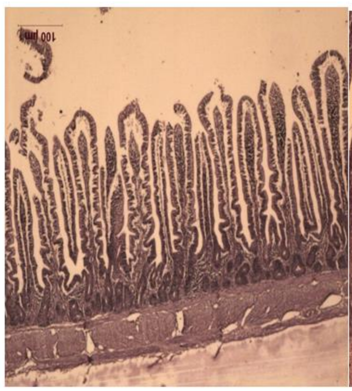

Plate 3: 1% of *Spirulina platensis* Plate 4: 1.5% of *Spirulina platensis*

Shows Increase Height

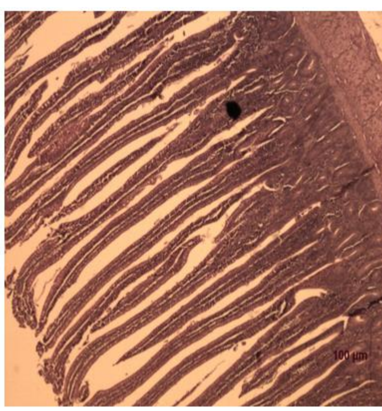

Figure 1: Effect of Supplementation of Different Levels of *Spirulina platensis* on Villi Length

Intensive livestock production systems may be associated with multiple stressful incidents that negatively impact immune response and animal performance. The high metabolic rate during intensive feeding is accompanied by an increased production of free radicals, and any imbalance between production of these molecules and their safe

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disposal may culminate in oxidative stress, which can damage cells and tissues (Miller et al., 1993; Lykkesfeldt & Svendsen, 2007). Therefore, under oxidative stress conditions, there is an increased demand for antioxidants to reduce the deleterious effects of free radicals on the immune system (Carroll & Forsberg, 2007). Interestingly, feeding natural, rather than synthetic, antioxidant could be advantageous to animal welfare and consumer safety (Call et al., 2008; Makkar et al., 2007). The blue-green algae, *Spirulina platensis*, have been considered as a suitable natural antioxidant and immune-stimulant to humans and animals with fewer side effects and more cost effectiveness than synthetic products (Abdel-Daim et al., 2013; Belay, 2002; Khan et al., 2005). Recently, the impact of dietary Spirulina supplementation on animal health and productivity has been reported (Holman & Malau-Aduli, 2012).

In the present research work it was found that the villi height increased at the level of 1gm/kg of *Spirulina platensis* in diet supplementation of broiler chicken. Our observation is sustained by Zhang *et al.,* (2005) who reported the positive role of yeast cell wall in ileal mucosal development of broiler chicks. It was found that the different yeast products contain varying amounts of cell wall fraction and therefore the prediction of response to various yeast products may require additional data regarding levels of *β*-glucans and *α*-mannans. From this result it can be deduced that increase in the villus height suggests an increased surface area capable of greater absorption of available nutrients (Caspary, 1992). Likewise, greater villus height increases the activity of enzymes secreted from the tip of the villi resulting in improved digestibility (Hampson, 1986).

Carcass Weight (g)

The dressing weight of Meat, Breast, Leg, Gizzard, Neck, Liver, Heart and Abdominal fat (percent live body weight) of broilers are presented in Table 9 to 10. The mean dressing weight of broilers supplement with *Spirulina platensis* was statistically significant at (P<0.01) in trail 1. The total weight (2172.33±14.26), meat weight (1588.66±13.56), breast weight (713±4.863), leg weight (453.5±7.448), gizzard (66.5±0.9), neck (62.66± 2.39), liver (91.16± 2.82) and heart (17.66± 0.334) of broilers on 36th day of age was significantly increased but fat weight (20± 1.63) was decreased in 1% of *Spirulina platensis* than the control (table 9 and 10).

Table 9: Effect of Supplementation of Different Levels of *Spirulina platensis* on Carcass Weight (g) of Broiler Chicks on Day 15, 22 and 36.

	Total weight	Meat	Breast	Leg
control	2035.83±14.163c	1393.5±8.58c	571.33±6.707c	350±2.779c
0.5%	2071.5±16.824bc	1491.5±16.126b	637.33±3.223b	412.83±2.367b
1%	2172.33±14.26a	1588.66±13.56a	713±4.863a	453.5±7.448a
1.5%	2094.5±17.246b	1423.83±28.744c	623±2.978b	443.5±2.627b
Treatment X days	13.68**	22.45**	157.17**	115.79**

** Significant at P< 0.01. Mean in a column followed by a same letter (s) are not significantly (P<0.05) different according to Duncan's Multiple Range Test.# Mean ±S.E

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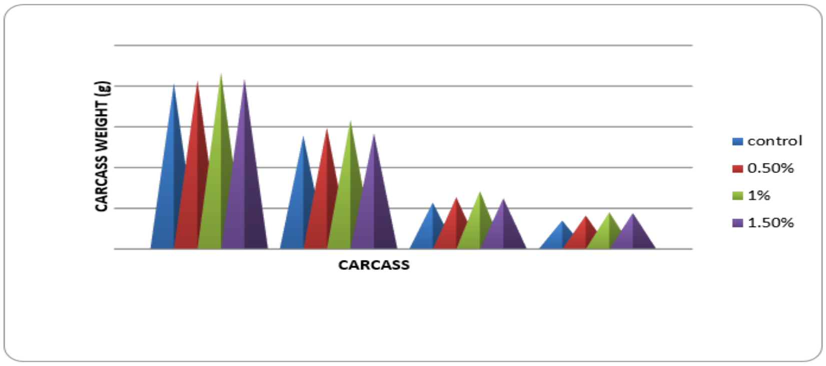

Figure 2: Effect of Supplementation of Different Levels of *Spirulina platensis* on Carcass Weight (g)

Table 10: Effect of Supplementation of different Levels of *Spirulina platensis* on Carcass Weight (g) of Broiler Chicks on Day 15, 22 and 36

	Gizzard	Neck	Liver	Heart	Fat
control	51.166±1.6b	57.5± 0.43b	78.17± 3.125b	14.66 ±0.496b	38± 0.819a
0.5%	57.66±1.39b	59.66± 2.84b	84.83± 1.564b	16.83± 0.544b	30.33± 0.92b
1%	66.5±0.9a	62.66± 2.39a	91.16± 2.82a	17.66± 0.334a	20± 1.63b
1.5%	58±2.5b	58.66± 0.95b	82.16± 2.128b	16.5± 0.995b	28.66± 1.789b
Treatment X days	13.57**	1.32**	15.07**	6.01**	47.94**

** Significant at P< 0.01. Mean in a column followed by a same letter (s) are not significantly (P<0.05) different according to Duncan's Multiple Range Test. # Mean ±S.E

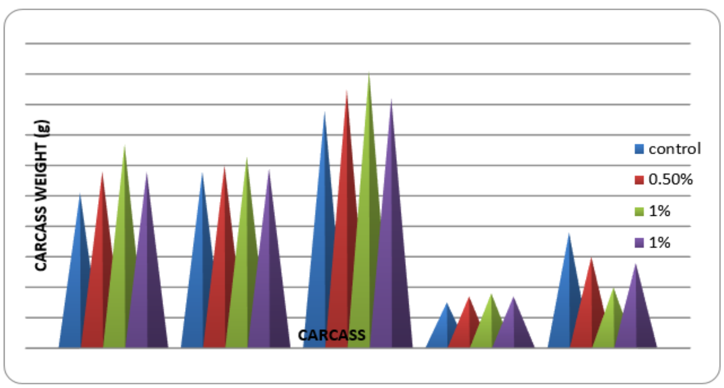

Figure 3: Effect of Supplementation of Different Levels of *Spirulina platensis* on Carcass Weight (g)

CONCLUSION

Studies demonstrate that including *Spirulina platensis* in the diet can enhance the condition of intestinal villi and epithelial cells, leading to better growth performance. The synergistic benefits seen with 1% *Spirulina platensis* on both growth performance and intestinal histology suggest it is a beneficial addition to probiotic supplementation.

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