

Role of *Spirulina platensis* Supplementation on Growth Performance, Blood Metabolites and Antioxidative properties of Fattening Lambs

ABSTRACT:

Twenty healthy lambs weighing (45.0±0.5 kg. body weight) were randomly selected from a herd of 250 lamb, left two weeks for acclimation, then divided into two equal groups; the first group (Control) received a commercial diet, while the second group (Sp) received same commercial diet but with *Spirulina platensis* powder at the rate of 1g/10kg B.W/day. The basal diet was formulated to meet all lambs' nutritional requirements. Lambs received a diet twice every single day at 6.0am and 4.0 pm with full access to water all the day. Animals were weighed on days 0, 14, 28 & 42 of the experiment, after fasting for a twelve hours after the evening meal. *Spirulina platensis* increased body weight gain, hemoglobin, white blood cells, total protein, albumin, globulin, A/G ratio, vitamin A, blood GSH and serum MDA, while ALT, AST, glucose, Cholesterol and triglyceride were decreased. *Spirulina platensis* showed antioxidant properties that led to improving growth performance of lambs.

Keywords: *Spirulina platensis*, Fattening lambs, Growth performance, Daily weight gain, Daily feed intake, Vitamin A, Blood GSH, Serum MDA.

INTRODUCTION:

Spirulina is a blue-green microalga that is considered the oldest life form on earth. *Spirulina platensis* proved to contain a valuable protein content of 55% to 70% with plenty of essential amino acids content like Lysine, methionine, tryptophan and cysteine. Moreover, ruminants can consume *Spirulina* [1]. *Spirulina platensis* is a proven source of protein and other beneficial nutritional constituents [2]. *Spirulina* can be found in salt lakes, soil, marshes, fresh water, brackish water and seawater. It is an obligate photoautotroph, therefore cannot grow in the dark, even when other factors are available. The optimum temperature for growth is between 35°C and 39°C [3].

Sheep as a substitution for livestock requires more attention, not only for supporting the rural community's economy, but also for the benefits that would result to the farmers and the whole society as well. Sheep offer a nice eating experience due to the quality of the meat. Most farmers and breeders still use the conventional methods when it comes to feeding their sheep herds; therefore, forage is still the first choice for sheep-fattening strategies.

Extensive farm animal manufacturing systems may be related to a couple of stressful incidents that negatively affect immune response and the overall animal's performance. The excessive metabolic rate for the duration of in-depth feeding is accompanied by an increased manufacturing of free radicals, and any imbalance between production of these molecules and their secure disposal can culminate in oxidative stress, which can damage cells and tissues [4 & 5].

There is a continual demand for antioxidants to reduce the serious effects of free radicals that negatively affect the immune system especially under extensive oxidative stress situations [6]. Therefore, the substitution of synthetic for natural feeding leads to better results for animal welfare and consumer safety, due to the presence of natural antioxidants. This is of great importance for animal welfare and consumer health [7&, 8]. *Spirulina platensis* is a perfectly cost-effective natural antioxidant and immune booster for humans and animals with almost no side effects [9, 10&, 11]. The present study was held to investigate role of *Spirulina platensis* supplementation on growth performance, blood metabolites and antioxidative properties of fattening lambs.

2. Materials and Methods:

Animal welfare and ethics clearance

This study was undertaken at a private lamb herd, California, USA, and the use of animals and procedures adopted in this study were in accordance with the Animal Welfare Act of Practice for the Care and Use of Animals for Scientific Purposes.

2.1. *Spirulina platensis*:

Spirulina platensis powder was purchased from a local store located in California; chemical composition of *Spirulina platensis* was represented in table 1.

Table (1): Chemical Composition of *Spirulina platensis*.

Item	% Content
Total Protein	68
Total Carbohydrates	30.75
Total Fat	3.3
Fiber	6.0
Moisture	4.5
Ash	9.0

2.2. Animals and Diets:

The experimental work done on a lamb farm, California (USA) during the period from October 2020 to December 2020. Chemical analyses was done in a chemistry laboratory near Anaheim, California, USA. Twenty healthy lambs weighing (45.0 ± 0.5 kg body weight) were randomly selected from a herd of 250 lamb, left two weeks for acclimation, then divided into two equal groups; the first group (Control) received a commercial diet, while the second group (Sp) received same commercial diet with *Spirulina platensis* powder at the rate of 1g/10kg B.W. day. The basal diet was formulated to meet all lambs' nutritional requirements. Lambs received a diet twice every single day at 6.0am and 4.0 pm with full access to water all the day.

Animals were weighed on days 0, 14, 28&42 of the experiment, after fasting for a twelve hours after the evening meal. The constituents of the basal diet were presented in Table 2.

Table (2): Nutrient Analysis of Lamb Feed*

Item	Content
Crude Protein, min	14.0%
Crude Fat, min	5.0%
Crude Fiber, max	11.0%
Acid Fiber Detergent, max	14.0%
Calcium, min	0.9%
Calcium, max	1.4%
Phosphorus, min	0.3%
Salt, min	0.1%
Selenium**	0.2
Vitamin A***	4,250
Vitamin D***	830
Vitamin E***	50

*Includes not more than 0.85% equivalent protein from non-protein nitrogen

Ppm*IU/lb

2.3. Sampling and Analysis

Blood samples were collected from Jugular vein of lambs on first day of the experiment (day0) and each consecutive two weeks thereafter for a period of three intervals. Hemoglobin (Hb) and White blood cell counts (WBC) were analyzed according to Linne&Ringsrud[12]; reduced glutathione was analyzed according to Beutler et al. [13]. Total protein (TP), Albumin, alanine amino transferase (ALT), aspartate amino transferase (AST), cholesterol (CHO), triglyceride (TG) and blood urea nitrogen (BUN) were analyzed according to Young[14]. Globulin was estimated by subtracting albumin value from total protein value, in the same way albumin/globulin(A/G) ratio was calculated. Malondaldehyde (MDA) concentration was done according to Ohkawa et al. [15]. Vitamin A (Retinol) concentration was done as described by Suzuki & Katoh, [16].

3. Statistical Analysis

Statistical analysis of the obtained data was carried out for analysis of variance according to [17]. Using SAS software version 9.4, 2020 USA. Means were compared by the L.S.D. values at 5% level.

4. Results and Discussion:

Spirulina platensis acquired great success as a growth promoter and antioxidant agent for animals, due to its rich content of numerous beneficial organic compounds, which boost the immunity system and enhance growth performance. Spirulina platensis was used throughout this study in fattening lambs.

4.1. Growth Performance:

Table 3 summarized the growth performance of lambs, Spirulina platensis caused highly significant increased ($P < 0.01$) for daily weight gain (kg/day) and final weight (kg), while Spirulina platensis significantly increased ($P < 0.05$) daily feed intake (Kg/day). On the other hand, feed conversion ratio highly significant decreased ($P < 0.01$) compared to the control group. The protein content of Spirulina platensis used in this study is 68%, which considered the main factor enhancing the growth performance, this finding is in a full agreement with that obtained by [18, 19, 20], all illustrated that in order to have better growth, lambs should have a high-quality feed with a valuable rich-protein supplement. The increase in body weight and average daily weight gain could be due to the increased rumen microbial crude protein production [21, 22]. Spirulina platensis contains valuable nutrients, like protein, vitamins, essential fatty acids, minerals, amino acid as well others, which could boost the growth rate. [23]. Spirulina may boost and improve the growth performance by improving the development and the morphological structure of the digestive tract that lead to an increase in total tract digestibility of protein and dry matter [24]. Feed Conversion Ratio was 11.5 ± 1.45 and 7.45 ± 0.32 for control and Spirulina platensis feed group respectively, the same results was also proposed by [25].

Table (3): Effect of treatment on Growth Performance

Item	Treatment	
	Control	Sp.
Initial Weight (Kg).	45 ± 0.5	45 ± 0.5
Daily Weight Gain (Kg/Day).	0.135 ± 0.02	$0.245 \pm 0.016^{**}$
Daily Feed Intake (Kg/day).	1.58 ± 0.002	$1.69 \pm 0.01^*$
Final Weight (Kg).	51.6 ± 0.89	$54.75 \pm 2.00^{**}$
Feed Conversion Ratio	11.5 ± 1.45	$7.45 \pm 0.32^{**}$

Con= Control, Sp= Spirulina platensis

4.2. Hematology

The Hb concentration and WBC count of lambs fed with Spirulina platensis were higher than that fed with the commercial lamb diet (Table 4). The Hb concentration and WBC count were significantly higher in Spirulina platensis group compared to the control group. The increase in hemoglobin concentration in spirulina platensis supplementation compared to the control may be due to the increased iron content in Spirulina platensis and presence of phycocyanin. The increase in WBC count may be due to presence of phycocyanin and polysaccharides levels in spirulina platensis [26]. Spirulina platensis supposed to boost the immunity in fish [27], and in chickens [28].

Table (4): Effect of treatment on Hemoglobin Concentration and total WBC count

Item	1 st Sample(0 day)	2 nd Sample(14 day)	3 rd Sample(28 Day)	4 th Sample(42 day)
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	Control	Sp.	Control	Sp.	Control	Sp.	Control	Sp.
Hemoglobin (Hb)	11.12 e	11.23 d	11.24 d	11.54 c	11.43 c	11.69 b	11.51 c	11.85 a
White Blood Cells	10356 b	10638 a	8340 f	8623 e	7890 g	8865 d	7675 h	9140 c

Sp= Spirulina platensis Hb= Hemoglobin WBC= White blood cells

4.3. Biochemical Parameters

Table 5 showed the biochemical parameters; it is clear from the table that there was a significant increase for TP, albumin, globulin and A/G ratio. The increase in TP, albumin, globulin and subsequently A/G ratio may be due to the increased protein content in Spirulina platensis used in this study. The same finding was proposed by [29]. The increased plasma globulin is always indicates higher individual resistance and thought to be associated with a strong innate response [30]. Concerning ALT, AST, there was significant decrease in Spirulina platensis supplementation group by the end of the experiment at 42 day. Spirulina platensis possess a protective action against liver dysfunctions [31]. Spirulina platensis caused a significant decrease in glucose level of lambs by the end of the experiment. Spirulina platensis possess anti-diabetic properties [32]. Spirulina platensis caused a significant decrease for both Cholesterol and triglycerides compared to the control group by the end of the experiment at 42 day. The same finding is in consistent with that proposed in rabbits by [33], in rats [34], in hamsters [35] and in human [36]. The decrease in cholesterol may be due to the increase in lipase and hepatic triglyceride lipase action [37]. [38] Proposed another finding for the decrease of Cholesterol concentration who referred the decrease in cholesterol concentration with Spirulina platensis to the ileal bile acid absorption and inhibition of both jejunal CHO absorption. Spirulina platensis reduced triglyceride and Cholesterol [39].

Table (5): Effect of treatment on biochemical parameters

Item	1 st Sample(0 day)		2 nd Sample(14 day)		3 rd Sample(28 day)		4 th Sample(42 day)	
	Control	S.p.	Control	S.p.	Control	S.p.	Control	S.p.
TP (g/dl)	4.44 f	4.56 e	4.67 d	4.86 c	4.78 c	4.98 b	4.93 b	5.45 a
Albumin (g/dl)	2.10 e	2.19 d	2.18 d	2.37 c	2.33 c	2.56 b	2.46 b	2.74 a
Globulin (g/dl)	2.24 f	2.43 d	2.36 e	2.49 d	2.42 d	2.68 b	2.58 c	2.76 a
A/G ratio	0.90 c	0.92 c	0.88 d	0.95 c	0.95 c	1.06 a	1.00 b	1.01 b
ALT(U/ml)	20.90 c	20.44 e	21.31 b	20.74 d	21.54 a	20.46 e	21.34 b	19.39 a
AST(U/ml)	51.10 a	50.00 e	50.89 b	50.73 c	50.65 c	50.49 d	50.54 d	49.15 a
Glucose (mg/dl)	40.40 c	39.60 e	42.3 b	40.23 c	42.66 b	40.00 d	43.3 a	37.77 a
Cholesterol (mg/dl)	46.45 c	45.8 d	47.32 b	46.23 e	47.98 b	45.90 d	48.44 a	45.55 e
Triglycerides (mg/dl)	40.40 c	39.76 d	40.78 c	40.10 cd	41.56 b	39.78 d	42.30 a	37.90 e

S.p= Spirulina platensis TP= Total Protein. A/G ratio= Albumin/Globulin ratio. ALT= Alanine amino transferase. AST = Aspartate amino transferase

4.4. Antioxidant Activity:

Table 6 represent vitamin A, Blood GSH and Serum MDA concentrations. It is clear from the results that there was a significant increase in Vitamin A and Blood GSH concentrations in the group fed with *Spirulina platensis* compared to the control group. On the other hand there was a significant decrease in Serum MDA in the group fed with *Spirulina platensis* powder compared to the control group. The increase in vitamin A and blood GSH with a decrease in serum MDA are true indicators of enhanced oxidative defense of animal tissues [40]. These results are in full agreement with that obtained by [41] who reported that *Spirulina platensis* supplementation leads to significantly increase in superoxide dismutase activity and catalase enzyme in the erythrocytes along with a concomitant increase in reduced tripeptide glutathione content in chicken. The antioxidative effect of *Spirulina* spp. Is referred to the presence of Phycocyanin [42]. Moreover, [34] stated that the antioxidative effect of *Spirulina* is related to the presence of many valuable ingredients that has distinct antioxidant activities alone or in synergy such as phycocyanin, β -carotene and polysaccharide. The antioxidant activity of phycocyanin is about 20 times more than vitamin C [29].

Table (6): Effect of treatment on Vitamin A, GSH and MDA concentrations

Item	1 st Sample(0 day)		2 nd Sample(14 day)		3 rd Sample(28 Day)		4 th Sample(42 day)	
	Control	S.P	Control	S.P	Control	S.P	Control	S.P
Vitamin A (g/dL)	61.20 f	61.65 f	63.6 e	66.5 d	67.9 d	72.8 b	68.3 c	86.3 a
Blood GSH (M/L)	2.6 e	3.3 d	2.8 e	4.3 c	3.6 d	6.1 b	4.2 c	9.4 a
Serum MDA (nM/ml)	7.3 f	7.45 f	8.3 d	7.8 e	11.9 b	9.9 c	13.6 a	12.4 b

5. CONCLUSION:

Spirulina platensis increased body weight gain, hemoglobin, white blood cells, total protein, albumin, globulin, A/G ratio, vitamin A, blood GSH and serum MDA, while ALT, AST, glucose, Cholesterol and triglyceride were decreased. *Spirulina platensis* showed antioxidant properties that led to improving growth performance of lambs. Future experiments will be done soon to explore the nature and mode of action of the antioxidant properties of *Spirulina platensis*.

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