



## SHORT COMMUNICATION

### Efficacy of Protein, Symbiotic and Probiotic Supplementation on Body Performance and Organs Weight in Molted Layers

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#### ABSTRACT

Two hundred White Leg Horn Layers (70 week age) were arranged and brought to the poultry research station, Department of Physiology and Pharmacology, University of Agriculture, Faisalabad. Four groups were made (n=50 each) into keeping G1 as control (CP 16%, No other supplement), G2 (CP 18% diet), G3 (CP16% diet; symbiotic @ 85 mg L<sup>-1</sup>/day) and G4 (CP 16% diet; probiotic @ 85 mg L<sup>-1</sup>/day). The body and organs (heart, liver, spleen, kidney, brain and pituitary) weight from fifteen birds in each group at 5% (5P), peak (PP) and end (EP) of post molt production stage were determined. The overall mean heart weight in G2 and pituitary weight in G2 and G3 reduced (P≤0.05) as compared to G1. The mean kidney weight found increased (P≤0.05) in G3 and G4 as compared to G1. The results show metabolic relation of protein and probiotics with body organs.

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#### INTRODUCTION

Molting program in poultry is based on the natural molting process as the wild birds undergo annually. Induced molting is used in the poultry industry to increase the reproductive lifespan of layers leading to new laying cycles (Laurentiz *et al.*, 2005). The supplementation of molted birds with additional protein and probiotics is an innovative approach in this regard. Togun *et al.* (2004) studied the positive effects of crude protein addition and follicle stimulation on the recovery and performance of one hundred post molted, aged, Nera Black Hens. Johnson and Lohman (2003) suggest that as a result of breeding specialized hens, which lay more frequently, the protein requirements of laying hens has increased. Traditional diets are no longer good enough on their own.

Improved digestion and nutrient utilization is also a contributing factor for this. In a study by Yoruk *et al.* (2004), the use of probiotic in the late laying period of hens shows the improvement in feed conversion with reduction in the mortality. In this process, the fat was actually mobilized from the muscles and different tissues of the body including body organs. The current study was thus planned to seek the dynamics of body and different body organs weight in molted laying hens in response to protein, symbiotic and probiotic supplementation.

#### MATERIALS AND METHODS

Spent commercial White Leg Horn hens (n=200) of 70 week old with similar weight range were procured from the commercial layer farm. The birds were induced to molt through high dietary zinc @ 3g/kg of feed (100g, CP 16%, 2795 Kcal energy)/day/bird. The birds were equally distributed to four groups (n=50 each) and allocated with their respective treatments keeping G1 as a control (CP 16%; no other supplement), G2 (CP 18% diet, no other treatment), G3 (CP 16%; Symbiotic (Perfectin®) in water @ 85mg L<sup>-1</sup> of drinking water daily), G4 (CP 16%; Probiotic (Protexin®) @ 85mg L<sup>-1</sup> of drinking water daily) for the whole experiment period. The feed, Perfectin and Protexin formulation has been presented in the Table 1. The temperature of the poultry house was maintained at 27±2°C throughout the experiment. The record of body weight was maintained throughout the experiment by randomly selecting 15 birds from each group. Fifteen birds were slaughtered at 5% production level (5P), peak of production (PP) and end of the production (EP) to collect the organs, including heart, liver, spleen, kidney, brain and pituitary for determining their weight. The data were subjected to two-way analysis of variance technique using two factorial completely randomized designs. Duncan multiple range test was applied to seek difference between means.

**Table 1:** Composition of feed and supplements (g/100gm) for molted white leghorn hen

Feed Ingredients	CP 16% Energy = 2795 Kcal g/100g	CP 18% Energy = 2800 Kcal g/100g
Corn	40	40
Rice Tips	10	10
Rice Polishing	11	11
Maize Gluten 30 %	6	-
Maize Gluten 60 %	-	4
Canola Meal	10	10
Soyabean Meal	8	10
Fish Meal	6	6
D.C.P	1.5	1.5
Limestone Powder /0 Chips	7	7
Vitamin Premix <sup>1</sup> + Amino acid <sup>2</sup>	0.5	0.5
Total	100	100
Composition of Perfectin (Per Kg)	Composition of Protexin	
Probiotic: Viability: 1×10 <sup>4</sup> cfu ml <sup>-1</sup>	Viability: 1 × 10 <sup>6</sup> cfu ml <sup>-1</sup>	
Lactobacillus acidophilus	Lactobacillus plantarum	
Bifidobacterium thermophilus	Lactobacillus plantarum	
Bifidobacterium longum	Lactobacillus bulgaricus	
Streptococcus faecium	Lactobacillus acidophilus	
Prebiotics:	Lactobacillus rhamosus	
VitA 4,000,000 IU	Lactobacillus acidophilus	
Vit. D3 800,000 IU	Lactobacillus rhamosus	
Vit. E 500 IU	Lactobacillus rhamosus	
Vit. K 200 mg	Bifidobacterium bifidum	
Vit. B1 200mg	Bifidobacterium bifidum	
Vit B2 2,000mg	Streptococcus thermophilus	
Vit B6 600mg	Streptococcus thermophilus	
Vit B12 2,000mg	Enterococcus faecium	
Vit C 2,000mg	Enterococcus faecium	
Folic acid 100mg	Aspergillus oryzae	
Niacin 10,000mg	Aspergillus oryzae	
L-lysine 5,000mg	Candida pintolopesi	
DL-methionine 15,000mg	Candida pintolopesi	
Iron 7,500mg	Candida pintolopesi	
Copper 1,000mg	Candida pintolopesi	
Zinc 7,500mg	Candida pintolopesi	
Manganese 10,000mg	Candida pintolopesi	

Study Groups: G1- (CP16 %, no other supplement), G2- (CP18%, no other supplement), G3- (CP16%, Symbiotic @ 1g/4L drinking water), G4- (CP16%, Probiotic @ 1g/4L drinking water); <sup>1</sup> Composition per kg of diet: vitamin A, 8,300 IU; cholecalciferol, 2,200 ICU; vitamin E, 8 IU; vitamin B12, 0.02 mg; riboflavin, 5.5 mg; D-calcium pantothenic acid, 15 mg; niacin, 36 mg; choline, 500 mg; folic acid, 0.5 mg; vitamin B1, 1 mg; pyridoxine, 2.2 mg; biotin, 0.05 mg; vitamin K, 2 mg. <sup>2</sup> Composition per Kg of diet: Methionine 0.143 g, Lysine 0.72g, Threonine 0.35g.

## RESULTS AND DISCUSSION

The results are given in the Table 2, which shows high ( $P \leq 0.05$ ) body weight in G2 and G3 at 5P as compared to control (G1), while overall mean body weight did not differ significantly between groups irrespective of the production stage. The increase ( $P \leq 0.01$ ) in liver weight was observed in G2 and G4 as compared to G1 at 5P, however, the liver weight then decreased significantly in G3 and G4 as compared to G1 at EP. The mean heart weight did increase significantly in G3 and G4 at PP and G4 at EP as compared to other groups. The overall mean high ( $P \leq 0.01$ ) heart weight was recorded in G4 as compared to G1, which was irrespective of the production stage demarcation (Table 2). The data regarding the spleen weight in different groups at any of production stage was found statistically non-significant. The overall mean kidney weight was found to be high ( $P \leq 0.01$ ) in G3 and G4 as compared to G1 regardless of the production stage (Table 2). The data regarding brain weight found statistically non-significant. However, irrespective of the production span overall mean weight of pituitary found decreased significantly in G2 and G3 as compared to G1 and G4 (Table 2) this decrease in G2

**Table 2:** The body and organs weight (mean±SE) in different groups at 5% production (5P), peak of production (PP) and end of production (EP)

Group	5P	PP	EP	Overall Mean
<b>Body Weight</b>				
G1	1.36±0.03 <sup>d</sup>	1.42±0.05 <sup>abcd</sup>	1.44±0.04 <sup>abcd</sup>	1.41±0.02
G2	1.48±0.06 <sup>abc</sup>	1.44±0.04 <sup>abcd</sup>	1.37±0.03 <sup>cd</sup>	1.43±0.03
G3	1.49±0.04 <sup>ab</sup>	1.51±0.05 <sup>a</sup>	1.33±0.04 <sup>d</sup>	1.44±0.03
G4	1.44±0.04 <sup>abcd</sup>	1.39±0.03 <sup>bcd</sup>	1.41±0.04 <sup>abcd</sup>	1.42±0.02
<b>Liver Weight</b>				
G1	30.04±1.20 <sup>cd</sup>	30.92±0.98 <sup>bcd</sup>	31.89±1.77 <sup>bcd</sup>	30.95±0.77
G2	34.80±1.16 <sup>ab</sup>	33.72±1.55 <sup>bc</sup>	29.52±1.46 <sup>de</sup>	32.68±0.91
G3	33.67±0.86 <sup>bc</sup>	32.03±1.75 <sup>bcd</sup>	24.85±1.31 <sup>f</sup>	30.18±1.09
G4	38.35±1.16 <sup>a</sup>	32.86±1.30 <sup>bcd</sup>	26.14±1.72 <sup>ef</sup>	32.45±1.30
<b>Heart Weight</b>				
G1	6.64±0.30 <sup>ab</sup>	5.11±0.16 <sup>d</sup>	6.05±0.30 <sup>bc</sup>	5.93±0.19 <sup>BC</sup>
G2	6.40±0.23 <sup>ab</sup>	4.98±0.16 <sup>d</sup>	5.52±0.32 <sup>cd</sup>	5.63±0.18 <sup>C</sup>
G3	6.65±0.29 <sup>ab</sup>	6.12±0.20 <sup>bc</sup>	5.52±0.26 <sup>cd</sup>	6.10±0.17 <sup>AB</sup>
G4	6.25±0.20 <sup>ab</sup>	6.23±0.21 <sup>ab</sup>	6.82±0.21 <sup>a</sup>	6.43±0.13 <sup>A</sup>
<b>Spleen Weight</b>				
G1	1.60±0.16	0.95±0.05	0.75±0.04	1.10±0.09
G2	1.57±0.06	0.76±0.06	0.73±0.03	1.02±0.09
G3	1.49±0.08	1.06±0.06	0.84±0.11	1.13±0.07
G4	1.54±0.13	1.11±0.09	0.70±0.04	1.11±0.09
<b>Kidney Weight</b>				
G1	8.81±0.36	8.13±0.43	8.27±0.41	8.40±0.23 <sup>C</sup>
G2	8.97±0.58	8.22±0.19	9.02±0.54	8.73±0.27 <sup>BC</sup>
G3	9.22±0.34	9.64±0.34	8.82±0.37	9.23±0.20 <sup>AB</sup>
G4	9.60±0.47	10.07±0.35	8.89±0.28	9.52±0.23 <sup>A</sup>
<b>Brain Weight</b>				
G1	2.79±0.13	2.72±0.04	2.60±0.05	2.70±0.05
G2	2.74±0.05	2.78±0.07	2.56±0.08	2.69±0.04
G3	2.52±0.07	2.85±0.07	2.48±0.11	2.61±0.06
G4	2.90±0.05	2.82±0.09	2.59±0.05	2.76±0.04
<b>Pituitary Weight</b>				
G1	7.90±0.27 <sup>bc</sup>	7.80±0.45 <sup>bc</sup>	8.30±0.32 <sup>b</sup>	7.91±0.20 <sup>A</sup>
G2	6.01±0.50 <sup>d</sup>	7.55±0.59 <sup>bc</sup>	6.60±0.33 <sup>cd</sup>	6.72±0.29 <sup>B</sup>
G3	5.81±0.34 <sup>d</sup>	6.65±0.72 <sup>cd</sup>	5.71±0.43 <sup>d</sup>	6.10±0.30 <sup>B</sup>
G4	8.70±0.46 <sup>b</sup>	5.69±0.23 <sup>d</sup>	9.10±0.80 <sup>a</sup>	8.62±0.59 <sup>A</sup>

G1- (CP16 %, no other supplement), G2- (CP18%, no other supplement), G3- (CP16%, Symbiotic @ 1g/4L drinking water), G4- (CP16%, Probiotic @ 1g/4L drinking water); abcd similar alphabets on means do not differ significantly ( $P \leq 0.05$ ) for each parameter; AB similar alphabets on over all means in a column do not differ significantly ( $P \leq 0.05$ ).

and G3 was seen at 5P and EP as compared to G1 and G4. A 5.5, 2, 3 and 3.5% mortality was recorded in G1, G2, G3 and G4 respectively in the post molt production phase.

Body weight gain in G2 and G3 after 5P shows an earlier weight gain and recovery in response to protein and symbiotic supplementation from the depressing impact of molting. Anwar *et al.* (2012) have reported a non-significant change in the growth hormone producing cells of the pituitary in molted White Leg Horn Layers which could also be responsible for a non-significant difference in the weight gain of different post molt groups in the current study. It was previously reported that intestinal microbiota is generally considered important for its beneficial role in host nutrition, health, and immunity (Sohail *et al.*, 2010) as the supplementation of symbiotic and probiotic helps to alleviate the heat stress in broiler birds. It was seen in current experiment that body weight was reduced in G2 and G4 at PP as compared to their body weight at 5P and inverse in G1, however, that decrease was non-significant, which could be attributed to the better post molt production performance and decreased size of prolactin producing lactotrophs (Anwar *et al.*, 2012).

The liver weight was decreased in symbiotic (G3) and probiotics (G4) supplement groups at the end of production. It shows the remobilization of liver glycogen and lipids in

these groups which may be used for the synthesis of egg components. Sohail *et al.* (2011) have reported a decrease in the liver generated lipid peroxidation enzymes in the symbiotic supplemented heat stressed broiler birds, which shows the decreased availability of lipids in liver in symbiotic supplemented birds. Over all liver weight irrespective of the production stage did not differ significantly among the groups, which is due to the non-significant change of body weight among the groups. Again the better production in protein and probiotic supplemented molted layers could be related to the remobilization of fats and glucose from liver (Anwar *et al.*, 2012). Spleen and brain weight did not differ significantly among the groups. However, heart and kidney weight did increase in the probiotic supplementation group (G4) comparatively. In contrast to this Shareef and Al-Dabbagh (2009) reported a non-significant change in the heart and kidney weight after feeding with various level of yeast as probiotic; however they reported a significant increase in the body weight of yeast feeded groups. Islam *et al.* (2004) have also reported a non-significant change in the heart and kidney weight of broiler chickens fed on different levels of probiotic (Protexin®) as compared to the control. The results of Islam *et al.* (2004) were also supported by the findings of Anjum *et al.* (2005) who also found non-significant change in the weight of heart and kidney after probiotic feeding in the diet of broiler chickens as compare to the control birds. In their study increased body weight and better feed conversion ratio was noticed by feeding the lactobacillus (probiotic) in the feed of broiler chickens. In current experiment contrary to the results of literature reports body weight did not differ significantly among groups. A non-significant change of heart and kidney weight in previous experiments reported in the literature could be related to the increase in the body weight which may be due to the muscle proliferation or fat deposition in other tissues rather than heart and kidney tissue.

The post molt increase in body weight of protein and symbiotic supplemented group at 5% production stage shows an early recovery in these groups as compared to control after the body weight loss during the process of

molt. The decrease of liver weight in symbiotic and probiotic supplemented layers shows the remobilization of liver fat and lipids, which could be the reason of strong immunity and low mortality in supplemented groups. The increase of heart and kidney weight in probiotic supplemented birds suggests the relation of these organs with metabolism and body clearance of probiotics.

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