Study of Serum Biochemical Metabolites during Late Laying Phase of Layer Chicken

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ABSTRACT

An experiment was conducted ontwelve adult white leg horn layers birds in their late phase of laying cycle from the poultry farm unit No. 3, department of Poultry Science, Bombay Veterinary College Goregaon, Mumbai 65.Blood sampling was done early in the morning at 8am by using 5ml syringe. Blood sample from the experimental birds was collected at weekly intervals. Biochemical parameters viz total protein, albumin, glucose, triglyceride, cholesterol, calcium, phosphorus were estimated by using pri-test diagnostic ready made kit supplied by [ROBONIC (INDIA) PVT .LTD. Plot No .A 374, TTC, MIDC industrial Area, Navi Mumbai- 400710, INDIA] .The parameters which were evaluated include total protein, albumin, serum glucose, cholesterol,triglyceride, calcium and phosphorus. The glucose content of the birds decreased with the age and were: 297 ± 4.45; 245 ± 11.42; 171.59±6.58 and 165.13 ± 8.33 mg/dl at 74th, 75th, 76th and 77th week of age respectively while the other parameters like cholesterol, total protein, albumin and triglyceride increased with age. The total protein levels were: 6.43 ± 0.13gm/dl; 7.26 ± 0.53gm/dl; 8.08 ± 0.51 g/dl and 8.28 ± 0.51 g/dl at 74th week, 75th week, 76th week and 77th week of age respectively.Non statistical difference was found in calcium but variation in level of calcium was seen with increase of age during egg laying season. Phosphorus showed non significant variation during lying season but was found statistically significant (p<0.05).

Key words: *Layers chicken; Total protein; Albumin; Glucose;Triglyceride; Total cholesterol; Ca & P;*

Chicken belongs to the *Galliformes* order and is believed to have originated from the red jungle fowl (*Gallus gallus*). Poultry industry forms a major portion of the agriculture sector in developing countries including India. India is the third largest producer of eggs in the world next only to China and USA and producing 45 billion eggs in 2006. The states such as Andhra Pradesh, Haryana, Maharashtra and Tamil Nadu are the major egg producing states that account for more than 60 per cent of the eggs produced in the country. The poultry meat production in India increased 18 folds from 81,000 tonnes in 1961 to 2.1 million tonnes in 2006. During the last two decade, India has a remarkable growth in poultry industry. India's egg production was 2 million tonnes in the year 2002 and remained amongst the top 5 of egg producing countries in the world. India has vast resource of livestock and poultry, which play a vital role in improving the socio-economic conditions of rural masses. Chicken are the most popular among the domesticated poultry species and account for 92 per cent of the total poultry population in India (*Chaudhary and Yousuf, 2008*).

The progress of poultry industry depends on feed industry. India is in a lucky position as regards agro-climatic conditions, availability of load and irrigation. India can produce feed to meet domestic demand as well as for exports. As of today, India exports millions of tonnes of soyameal cake to Europe where it is converted to feed. Feed costs represent about 70% of the total production cost (*Jadhavet al., 2007*).The blood biochemical analysis is a valuable tool for evaluating the state of health and indispensable preliminary knowledge of the biological material chosen for research. Its evaluation indicates the extent of damage in various vital organs and status of the disease. The

biochemical constituent of blood are relatively constant whereas fluctuation occurs due to age, sex, breed, climate, geographical locations, nutritional status, seasons and present status of individuals. The evaluation of blood biochemistry in birds allows for the identification of metabolic alteration due to many endo- and exogenous factors (*Bowes et al., 1989*). The clinical chemistry can be a useful adjunct in the development of a diagnosis and prognosis in avian disease (*Lewandowski et al., 1986*).

METHODOLOGY

Experimental birds: Twelve adult white leg horn layers birds in their late phase of laying cycle were used in this study in the poultry farm unit No. 3 Department of Poultry Science Bombay Veterinary College Goregaon Mumbai 65. The layer birds were kept in the cage system of housing cages unit measuring 1.5x1.5x0.3m and were fed with commercially prepared adult poultry feed and large amount of water. All the layer birds were kept on standard feeding and management conditions.

Blood sampling: Blood sampling were done early in the morning at 8.00 am by using 5ml syringe. About 3ml of blood was collected through jugular vein from each birds. Blood sample from the experimental birds was collected at weekly intervals. Blood was allowed to clot and then clear serum was separated by centrifugation at 3000 rpm for 15 minutes and stored at 20^0 C until analysed.

Biochemical parameters viz total protein, albumin, glucose, triglyceride, cholesterol, calcium, phosphorus were estimated by using pri-test diagnostic ready made kit supplied by [ROBONIC (INDIA) PVT .LTD. Plot No .A 374, TTC, MIDC industrial Area, Navi Mumbai-400710, INDIA]. The activity and concentration were estimated by using pri-test robonic spectrophotometer at different wave length as per pri-test diagnostickit.

Statistical analysis: Analysis of variance of the data of the concentration of total protein, albumin, glucose, total cholesterol, triglyceride, calcium and phosphorus were done according to *Snedecor and Cochran (1994)* using Completely Randomized Design (CRD). Differences in means were tested using critical difference (CD) test.

RESULTS AND DISCUSSION

Total protein: The total protein concentration in serum of layers bird are presented in Table 1, its analysis of variance in Table 2 and the mean value of total protein in serum were : 6.43 ± 0.13; 7.26 ± 0.53; 8.08 ± 0.51 g/dl; 8.28 ± 0.51 g/dl at 74th week, 75th week, 76th week and 77th week of age respectively.

Significant difference ($p<0.05$) in serum total protein were found and significant increase in levels of total protein were found in 74th to 76th week of age and the level remain almost the same from 75th to 77th week of the bird. This is in agreement with *Ayubaliet al. (2012)* who studied the total protein concentration in different age groups and reported that significant variation in total protein concentration increases with age. Our findings are also in agreement with *Laperaet al. (2010)* showed higher total protein concentration in late egg laying phase in female bronze turkey and *Arzour-Lakehalet al. (2013)* who reported increase in total protein concentration with age in broilers but contradictory to the findings of *Albokhadaim**et al.** **(**2013)* in female indigenous chickens which showed low concentration of total protein in adult age as compared with younger age. Blood protein concentration may vary due to breeding season, indicating that the egg production is associated with a marked increase in total protein concentration induced by estrogens.

Albumin: The serum albumin concentration in serum of layers bird are presented in Table 1, its analysis of variance in Table 2The mean values of albumin in serum, were : 1.37 ± 0.05; 1.94 ± 0.09; 2.08 ± 0.01; 2.10 ± 0.07 g/dl at 74th, 75th, 76th, 77th week of age respectively. The lowest concentration was observed in 74th week (1.37 ± 0.05) and highest concentration was observed in 77th week (2.08 ± 0.01). There was significant difference ($p<0.05$) in the levels of albumin concentration among the different weeks of layers. This is in agreement with *AyubAliet al. (2012), Laperaet al. (2010)* who studied the albumin concentration in different age groups and reported that significant variation in total protein concentration increases with age.

The increase in albumin might be due to the estrogen secretion at the onset of egg production which comes in agreement with the findings of *Uristet al. (1958)*. As regards to relationship of level of Albumin concentration in serum, our study showed that there is significant difference in levels of total albumin in serum and revealed that albumin concentration increases as age increases. Our findings are in agreement with *Ayubaliet al (2012), Bhattiet al. (2001), Laperaet al. (2010)* and *Piotrowskaet al. (2011)* in broilers.

Glucose: The glucose concentration in serum of layers bird is presented in Table 1, its analysis of variance in Table 2. The mean values of glucose in serumwere: 297

Table 1: Mean ± S.E of biochemical metabolites during late phase of laying in layers (Biochemical Metabolites)

Age of layers	Total Protein (g/dl)	Albumin (g/dl)	Glucose (mg/dl)	Triglycerides (mg/dl)	Total Cholesterol (mg/dl)	Calcium (mg/dl)	Phosphorus (mg/dl)
74th	6.43b	1.37b	297.0a	349.98b	41.70d	17.84	5.87ab
week	±0.13	± 0.05	±4.45	± 60.6	± 3.82	± 1.04	± 0.26
75th	7.26ab	1.94a	245.45b	401.93B	86.83c	19.36	6.21a
week	± 0.53	± 0.09	±11.42	± 57.43	± 3.25	± 0.94	± 0.43
76th	8.08a	2.08a	171.59c	614.43a	99.42b	20.87	6.39a
week	± 0.51	± 0.01	±6.58	± 17.02	± 4.13	± 0.72	± 0.27
77th	8.28a	2.10a	165.13c	630.41a	217.15a	21.07	4.87b
week	± 0.51	± 0.07	±8.33	± 47.07	±2.26	± 1.53	± 0.46

Table 2: ANOVA table for biochemical metabolites during late phase oflayers.

ANOVA → Biochemical Metabolites ↓	Source of Variation	Degree of Freedom	Sum of Squares	Mean sum of squares	F cal	C.D.
Total Protein(g/dl)	Groups	3	25.815	8.605	3.411	-
	Error	44	110.994	2.523		(0.05)= 1.307
	Total	47	-	-		
Albumin (g/dl)	Groups	3	4.291	1.430	25.079	(0.01) = 0.262
	Error	44	2.510	0.057		(0.05) = 0.196
	Total	47	-	-		
Glucose (mg/dl)	Groups	3	143167.17	47722.39	105.047	(0.01) = 23.42
	Error	44	19989.038	454.296	-	(0.05) = 17.53
	Total	47	-	-	-	
Triglycerides(mg/dl)	Groups	3	746675.43	248891.810	8.034	(0.01) =93.437
	Error	44	1363120.2	30980.005	-	(0.05) =144.79
	Total	47	-	-	-	
Total cholesterol(mg/dl)	Groups	3	201472.47	67157.49	501.44	(0.01) = 12.71
	Error	44	5892.857	133.93	-	(0.05) = 9.52
	Total	47	-	-	-	
Calcium (mg/dl)	Groups	3	81.618	27.206	1.767	Non Significant
	Error	44	677.450	15.397	-	Non Significant
	Total	47	-	-	-	
Phosphorus(mg/dl)	Treatments	3	16.701	5.567	3.426	
	Error	44	71.487	1.625	-	(0.05)= 1.049
	Total	47	-	-	-	

± 4.45; 245 ± 11.42; 171.59±6.58; 165.13 ± 8.33 mg/dl at 74th, 75th, 76th, 77th week of age respectively. The glucose concentrations in serum in present study are almost similar to the levels reported by *Bhattiet al. (2001), Abdi-Hachesooet al. (2011)*. However, *Albokhadaimet al. (2012)* reported higher level of serum glucose.

In our study serum glucose level was highest at 74th week (197 ± 6.85 mg/dl) has significant difference ($p<0.05$) with the other groups of the age. There was a significant decrease in the level of glucose from 74th week (197 ± 6.85 mg/dl) onwards to 75th week (245.45b ±11.42) but the level remain almost same from the 76th week to 77th week of age of layer birds and the content decreased with age. The decrease in trend of glucose as age progress reported by *Ayubaliet al. (2012), Arzour-Lakehalet al. (2013)* is in agreement with our study. The decrease in the serum glucose level in the present investigation may be due to increased secretion of steroid hormones with age of the birds for the preparation of the laying. The decreasing trend of glucose level was accompanied by increased liver glycogen indicating a stimulated pancreatic activity

which comes in agreement with the findings of *Schulz (1940)* who had reported that in pigeons, the pancreatic islets of langerhans increased in size and number during the laying period of female.

Triacylglyceride: The triglyceride concentration in serum of layers bird are presented in Table 1, its analysis of variance in Table 2The mean values of triglyceride in serum, were : 349.9 ± 60.6; 401.93 ± 57.43; 614.4± 17.02 ; 630.4± 47.07 mg/dl at 74th, 75th, 76th, 77th week of age respectively. Lipid metabolite are strongly associated to energy metabolism and reflects its fluctuation occuring during the growth period. In the present study, the serum triglyceride concentration peaked on the 77thweek of life, reflecting intensive lipid metabolism and transport in the experimental layers chicken. Age related changes, with the tendency of serum triglyceride to increase significantly with age, were also observed in chickens by *Albokhadaimet al (2013).* There was significant variation ($p< 0.05$) in the levels of triglyceride among the different age groups of layers but contradictory to the findings of *Suchy et al. (2010), Kececi and Colet al. (2011)* where they have reported that triglyceride levels were significantly higher in young pheasants than in adults. The triglyceride content in yolk also reflects on lipogenetic function of liver which may be considered in future studies.

Total cholesterol: The cholesterol concentration in serum of layers bird is presented in Table 1, its analysis of variance in Table 2.The mean values of cholesterol in serum, were : 41.70 ± 3.82 ; 86.83 ± 3.25; 99.42 ± 4.13; 217.1 ± 2.26 mg/dl at 74th, 75th, 76th, 77th week of age respectively. The levels of total cholesterol wereincreasing as the age groups increase with statisticallysignificant variation ($p< 0.05$). Cholesterol is considered as the precursor of all steroid hormones therefore the levels of the total cholesterol is important for the transformation of the cholesterol to sex steroids. The level of cholesterol showed increasing trend as the age of the layers increases and the variation are statistically significant. The observation that total cholesterol concentration increases with the age groups is in agreement with *Ayubaliet al. (2012)* but in contrast decrease in levels of cholesterol was observed significantly during fattening period in chickens by *Piotrowskaet al. (2011).*

Calcium: The calcium concentration in serum of layers bird are presented in Table 1, its analysis of variance in Table 2.The mean values of calcium in serum, were : 17.84 ± 1.04; 19.36 ± 0.94; 20.87 ± 0.72; 21.07 ± 1.53 mg/dl at 74th, 75th, 76th, 77th week of age respectively. The calcium concentration in serum in present study are nearly similar to the levels reported by *Ayubaliet al. (2012).*The levels of calcium showed increasing trends as the age of layers increases but the variation was not statistically significant. These finding is in agreement with the reports of *Albokhadaimet al. (2013)* in indigenous chickens where they found that no significant variation were detected in the calcium levels with the increase in age. The serum calcium level in the present investigation increased at 77th week of age. The values on 74th week (17.84 ± 1.04 mg/dl) increased to 20.87 ± 0.72 mg/dl at 77th week of age.

Calcium is mainly needed for the ossification of bones, regulation of muscle activity and catalyzation of enzymes and hormone system. Increasing the levels of calcium with the age might be due to the increased secretion of steroid hormones. The steroid hormones are implicated in the regulation of Ca metabolism in laying hens through several modes of action. A considerable increase in plasma Ca levels at the beginning at laying period of hens and subsequent gradual increase were also observed by *Pavliket al. (2009)*

Phosphorus: The phosphorus concentration in serum of layers bird are presented in Table 1, its analysis of variance in Table 2.The mean values of phosphorus in serum, were: 5.87± 0.26; 6.21± 0.43 ; 6.39± 0.27 ; 4.87± 0.46 mg/dl at 74th, 75th, 76th, 77th week of age, respectively.

Phosphorus is an important constituent of nucleic acids and phospholipids. There was significant variation ($p< 0.05$) in the levels of phosphorus among the different age groups of layersbut the variationdid not show any definite trend. The levels was highest in 76th week (6.39 ± 0.27mg/dl) and lowest within 77th week (4.87± 0.46mg/dl). In contrast to our findings are reports of *Abdi-Hachesooet al. (2011), Albokhadaimet al. (2013)* where they have reported increases in phosphorus level with increase of age. In our findings the non significant increase in phosphorus levels from 74th week to 76th week of age and then significant ($p<0.05$) decrease at 77th week of age of layer birds. Our findings are in agreement with *Piotrowskaet al. (2011)* where they reported phosphorus is not age dependent and found non significant change upto 76th week of age.

CONCLUSION

Twelve adult white leg horn layers birds in their late phase of laying cycle were used in farm unit No. 3 department of Poultry Science Bombay Veterinary College Goregaon Mumbai 65. Blood sampling were done early in the morning at 8.00 am by using 5ml syringe. About 3ml of blood was collected through jugular vein from each birds. Blood sample from the experimental birds was collected at weekly intervals. Blood was allowed to clot and then clear serum was separated by centrifugation at 3000 rpm for 15 minutes and stored at 20^0 C until analysed. The parameters which were evaluated include serum glucose, cholesterol, total protein, albumin, triglyceride, calcium and phosphorus.

The glucose content of the birds decreased with the age while the other parameters like cholesterol, total protein, albumin and triglyceride increased with age and was statistically significant ($p<0.05$). Non statistical difference was found in calcium but variation in level of calcium was seen with increase of age during egg laying season. Phosphorus showed non significant variation during lying season but was found statistically significant ($p<0.05$). Extensive study of various biochemical constituents from the initiation of laying upto the end of laying cycle is required. Also the relation of the levels of various biochemical profile, mineral profile and endocrine profile during the different egg laying phase in chicken are to be studied.

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