

Fig 5 a & b Expression of LAT4 (aatransporter), C-Control T1-0.3ppm, T2-0.6ppm, a J-Jejunum (MP), b M- Magnum. EP- Early Laying period, *** P<0.001, *P<0.05

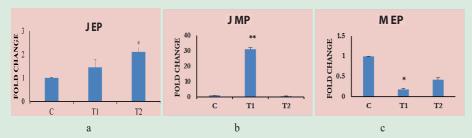


Fig 6 a, b & c-Expression of GHLR (Ghrelin receptor), C- Control T1-0.3ppm, T2- 0.6ppm a, b, J-Jejunum b- M-Magnum, EP-Early laying period, MP- Mid Laying period, *P<0.05, **P<0.001

Hormones

From the standard curves for each hormone and based on absorbances the estimation of concentration of respective hormones in plasma samples was conducted. The concentration of melatonin increased significantly (P<0.01, Fig 1) of both T1 and T2 groups at EP and MP periods of study (Fig 3). The either of the treatments during any of the time period, did not cause significant change in the concentration of ghrelin except at MP (Fig 4), where the concentration decreased upon treatment with 0.3 ppm of selenium (Fig 4). The concentration of EST increased (P<0.05) at EP and MP of the T1 group (Fig 5), whereas increase in concentration (P<0.01) was observed only at MP upon treatment with 0.6ppm (Fig 5). The either of the treatments did not cause significant difference in the concentration of progesterone between the groups and time periods.

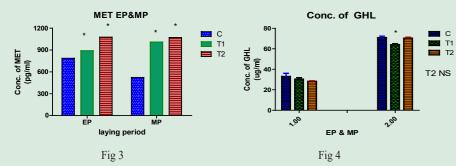


Fig 3 Effect of supplementation of Se (T1-0.3ppm, T2-0.6ppm) on concentration of plasma MET during EP and MP phase of laying period. C- Control, * P<0.01;

Fig 4 Effect of supplementation of Se (T1-0.3ppm, T2-0.6ppm) on concentration of plasma GHL during EP and MP phase of laying period. C- Control, * P<0.01

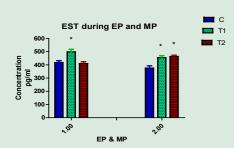


Fig 5

Fig 5 Effect of supplementation of Se (T1-0.3ppm, T2-0.6ppm) on concentration of plasma EST during EP and MP phase of laying period. C- Control. * P<0.01

The concentration of plasma amino acids of the Control group decreased significantly (P<0.05) when the birds passed from EP to MP stage (Table 3). The concentration of plasma amino acids was higher at EP compared to plasma concentration at MP. At EP, out of 14 amino acids estimated, concentration of 12/14 amino acids decreased upon supplementation of either 0.3/0.6ppm of organic selenium. Out of these the ten amino acids decreased significantly upon treatment with lower dose. Whereas with higher dose, concentration of all 12 amino acids decreased significantly. Only five essential amino acids were estimated (Table 3). Essential amino acids Valine, Threonine, increased significantly upon supplementation with either of the dose. The amino acid Phenylalanine increased with lower 0.3ppm dose only. Whereas at MP, 12/14 increased significantly upon treatment with 0.3ppm of selenium. Where as concentration of only 8/14 amino acids increased upon supplementation of 0.6ppm of organic selenium. Levels of essential amino acids Lysine, Valine, Threonine increased compared to the plasma concentrations of Control group. With lower dose 0.3ppm, also concentration of Methionine and Phenylalanine also increased when compared to Control group. At MP more number of amino acids and essential amino acids inceased at MP upon supplementation of either dose at MP (Table 3).

Table 3- Effect of organic selenium treatment on the mean concentration (ng/ml) of plasma amino acids

		EP			M	IP .
	С	T1	T2	С	T1	T2
Lysine	3417±15	1245±14	2155±12	2209±30	2852±31	2698±25
Alanine	35711±20	18876±23	25564±20	28596±25	38417±26	28709±21
Valine	11004±22	13216±12	12622±13	11562±15	13354±16	8778±15
Serine	28158±32	12285±20	15840±19	21351±20	27760±22	22775±12
Threonine	6190±13	7670±15	8265±18	9168±8	13696±12	12160±13
Glutamine	32769±26	17762±19	21796±22	35629±42	30410±35	31755±36
Asparagine				1955±15	1654±12	3734±14
Methionine	3036±11	2271±13	2421±10	3970±14	5488±15	3576±13
Glutamic acid	9678±18	5406±15	6937±12	8587±12	13238±20	8724±12
Phenyl alanine	4025±14	4822±10	3707±15	4089±10	6248±11	3534±15
Proline	12718±21	8144±18	8287±15	9149±15	12221±18	9716±12
Glycine	94.8±10	76.5±9	69.99±8	88.9±7	82.47±8	73.94±7
Cysteine	84.7±8	81.13±4	58.89±6	89.44±5	116.39±10	78.11±6
Tryptophan	6044±15	5561±14	5409±16	5179±19	7561±12	6209±10
Tyrosine	7654±19	6627±23	6418±25	9902±23	12421±20	9291±21

C- Control; Supplementation of Se (T1-0.3ppm, T2-0.6ppm), N=8, *P<0.05, **P<0.01 The blocks in row in yellow colour, are in the same trend when compared to control at EP and MP respectively.

Conclusion

The concentration of plasma hormones differed between the two laying periods, being less at MP indicating inversely proportional to egg production, which could be increased by supplementation of organic selenium which could increase egg production and marginally body weight of treated groups. The effects were different with different dose of selenium. Basic and applied work, related to relation between hormones, jejunum parameters and productivity, and their modulation by appropriate dose of organic selenium can be utilized by other researchers and farmers for increasing productivity related to poultry rearing.



Relation Between Different Physiological Parameters and Production Performance in White Leghorn Layers













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Objectives:

- To estimate Melatonin and Ghrelin in plasma, and their receptor expression in jejunum and magnum of White Leghorn during early (24-28 weeks) and mid laying period (32-36 weeks).
- To study relationship between level of hormones, amino acids, expression of amino acid transporters and differential egg production parameters at two different phases of laying period.
- To observe the effect of supplementation of organic selenium in modulation of different parameters mentioned and their relation with egg production.

Methodology

Experiments were conducted in White Leghorn layers at Directorate of Poultry Research farm, Rajendranagar, Hyderabad. Birds were selected at 16 weeks of age for the experiment. Actual experiment started at 24 weeks of age. Experiments were conducted for eight weeks during early (24-28 weeks) and mid laying period (32-36 weeks) respectively. A fifty number each of 24 week old White Leghorn breed of chickens of uniform body weight (Mean±200g) were selected and randomly allotted to three groups of fifty birds each and physiological parameters like body weight, feed intake, egg production and weight, Histology of jejunum were conducted. Experiments were conducted for eight weeks during early (24-28 weeks) and mid laying period (32-36 weeks) respectively for some other physiological parameters like plasma melatonin, ghrelin, estrdiol, progesterone by EIA, Quantification of fold change in gene expression of hormone receptors and amino acid transporters of jejunum tissue by Real Time PCR assay and plasma amino acids were estimated by UHPLC. One group served as Control which was offered feed devoid of organic selenium supplement but with routine premix, and the other groups were supplemented with additional selenoyeast, @0.15g/Kg of the product i e 0.3ppm(T1) and 0.6ppm (T2) respectively to the treatment groups.0ppm to the Control (C) group. It was a commercial product. From 16-20th week, grower feed was offered and from 21st-36th week, layer feed was offered. Feed was mainly based on maize and soybean as per the composition recommended by NRC, 1994.

Temperature in the shed during March till May varied from 29-38oC. Relative Humidity varied between 46-59%. Bodyweight (15d interval) and feed intake (weekly interval) were estimated. Blood samples were collected at weekly intervals. Egg production parameters like egg nos., egg weight, egg production percentage were estimated. Transfer of selenium to egg and breast muscles was estimated by Inductively coupled plasma - optical emission spectrometry.

Results and Discussion

For Experiment No. I:

When compared between the control groups of EP and MP, the body weight and egg weights and egg production percentage increased significantly as the birds passed from EP to MP stage. The two different doses of selenium had different effects. The lower concentration of the dose increased egg production significantly (P< 0.05) compared to control and T2 group at EP and Higher dose at MP (P<0.01) when compared to C and T1 (Table 1c). The feed intake of birds of T1 and T2 group increased significantly at MP (Table 1b). The higher concentration of the dose, increased body weight and transport of selenium significantly to muscles and egg at 36 weeks of age (Table 1a&e). Both the concentrations had beneficial effect (P<0.01) on the histomorphology of the jejunum (portion of digestive tract) the T1 being more efficient (Fig 1).

Table 1a

	EP Body weight (g) of WLH hens MPWeeks							
	25	27	29	32	34	36	Gain	
С	1342.1ª±1	1346.7ª±13	1356°±13	1404.6°±15.4	1414.4±13.2	1417.4°±12	75	
T1	1342.1ª±14	1354.3°±15	1374·69 ^a ±13.7	1427.4a±16.8	1434a ±12.4	1417.4ª±12	133	
T2	1334.6ª±12	1360.7ª±13	1424·22b±12.30	1449.0°±14	1478 ^b ±12	1517.2 ^b ±13.5	183	

Values are represented as Mean±SE. Values with different superscripts in a column are significantly different at least at P<0.05. C-Control, T1 and T2-Treatment C-Control, T1, T2- Treatment 24-28 weeks-35 birds, 32-26 weeks-25 birds

Table 1b

EP Egg production (%) of WLH MPWeeks						
	24	26	28	32	34	36
С	54.4*±0.5	65.1±0.7	82.1±1.0	76.5±0.3	68.84±1.2	64.06±0.9
T1(0.3ppm) 51±0.5 67.4±0.8 86.08*±0.6 79.8*±0.4 77.85**±1.1 72.1**±1						72.1**±1.05
T2(0.6ppm) 51±0.5 67.8±0.3 84.78±0.5 76.69±0.3 79.71**±1. 03 76.6±**1.04						
C-Control, T1, T2-Treatment 24-28 weeks (EP)- 35 birds 32-26 weeks (MP)- 25 birds * P<0.05;** P<0.01						

Table 1c

EP FI (g/bird/day) of WLH hens MP						
	24	26	28	32	34	36
С	99ª±1.4	88a±1.8	94.75°±0.85	99.1ª±0.4	75.8°±1.2	81.6°±0.6
T1	97ª±1.7	97 ^b ±1.09	93.8°±1.1	99.1ª±1.5	89.1 ^b ±1.4	90.1 ^b ±0.7
T2	97ª±1.2	101 ^b ±1.07	95°±1.71	96.3°±0.95	89.8 ^b ±1.4	89.83 ^b ±1.1

Values are represented as Mean±SE. C-Control, Treatment-T1, T2. Mean values with different superscripts are significantly different from each other.

Table1d

EP Egg weights (g) of WLH MP						
	24	26	28	32	34	36
С	43.1	46.15	47.8	49.2	49.75	51.1
T1	43.2	45.9	47.8	48.4	49.49	51.8
T2	43.1	46.4	47.9	49.9	49.91	51.28

C-Control, T1, T2- Treatment

Table 1e

Transfer of Selenium					
Conc. Of Selenium/ egg Conc. Of Se in breast meat/kg					
C (0)	11-12 ug	ND			
T1(0.3 ppm)	11-13 ug	ND			
T2 (0.6 ppm)	15-17 ug*	0.976±0.2 mg			

C- CONTROL, T1, T2-TREATMENT Total number of samples or eggs = 10 in each group; at MP i e 36 weeks of age. Average weight=51g for C and T2, T1-51.8g Inductively coupled plasma - optical emission spectrometry

Histomorphology of Jejunum

	c	71		T2	EP	
EP	Villus Height (um)	Crypt Depth (um)	МР	Villus Height (um)	Crypt Depth (um)	
Control	865.76±100	79.14±10	Control	962.38±120	400±30	
Treatment 1	1254±89	109.45±5	Treatment 1	1387±95	538±60	
Treatment 2	1039.2±105	103±5.8	Treatment 2	1500±100	450±50	
C T1 T2						

Fig 1 The villus height and crypt depth of both T1 and T2 were significantly (P<0.01) different from Control.

Studies on Gene expression

The fold change expression of CAT and b°AT genes when compared between control (C), and treatment (T1), (T2) groups, it was observed that expression of CAT and b°AT significantly increased (P<0.05) by T1 treatment in jejunum tissue for CAT gene (Fig 2a) and decreased (P<0.05) in magnum tissue at EP (Fig 2c) whereas fold change expression of b°AT increased (P<0.05) by T2 treatment also in jejunum tissue of both EP and MP periods (Fig 3a&b). The fold change expression of LAT2 gene increased (P<0.05) by T1 treatment in jejunum of EP group (Fig 4a) but decreased (P<0.05) in magnum tissue of EP (Fig 4b) and MP (Fig 4c) by the same treatment. The fold change expression of LAT 4 amino acid transporter gene when compared between control (C) and treatment (T1), (T2) groups it was observed that with T1 treatment, expression of gene for jejunum tissue increased at MP (P<0.001, Fig 5a) and decreased (P<0.05) in magnum tissue at EP (Fig 5b). The expression of the ghrelin receptor of jejunum increased significantly (P<0.01) at EP in the T2 group (Fig 6a) and at MP in the T1 group (P<0.001, Fig 6b). Whereas the expression of gene in magnum tissue decreased (P<0.05) at EP only (Fig 6c).

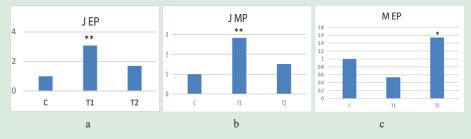


Fig 2 a, b, c-Fold change expression of CAT gene (aa transporter) in jejunum (J, a) and magnum (M, c) tissues of early period (EP, 26 weeks), jejunum tissue of mid period (J, b, 34 weeks). C-Control, T1-Treatment 1(0.3ppm Organic selenium enriched yeast), T2-Treatment2(0.6ppm Organic selenium enriched yeast). *P<0.05; **P<0.01



Fig 3 a & b- Fold change expression of boAT (aa transporter) gene in jejunum (J, a) tissues of early period (EP, 26 weeks), jejunum tissue of mid period (J, b, 34 weeks). C-Control, T1-Treatment 1(0.3ppm Organic selenium enriched yeast), T2-Treatment2 (0.6ppm Organic selenium enriched yeast). * P<0.05; P<0.01

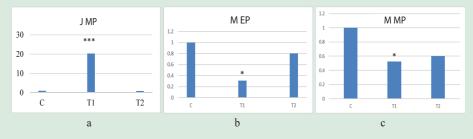


Fig 4 a, b & c- Fold change expression of LAT 2(aa transporter) gene in a jejunum (J,) tissues of mid period (MP, 34 weeks), b, c- magnum tissue of early and mid period (M, 26 & 34 weeks). C- Control, T1-Treatment 1(0.3ppm Organic selenium enriched yeast), T2-Treatment2 (0.6ppm Organic selenium enriched yeast). * P<0.05; ***P<0.001