

Effects of L-T₃ on the plasma glucose, protein and amino acid levels of the tree sparrow, *Passer montanus malaccencis*

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Abstract

The two major thyroid hormones are L-thyroxine (L-T₄) and Tri-iodothyronine (L-T₃) which they take an important role in the oxidative and metabolic process of the body of homoiothermous animals. In birds, L-T₄ and L-T₃ are important in the regulation of the body metabolism performing a number of diverse functions. The dose dependent effects of L-T₃ on the levels of glucose, proteins and amino acids of plasma were studied on the tree sparrow, *Passer montanus malaccencis*. Administration of different doses of T₃ to normal intact birds produced a significant dose dependent elevation in plasma glucose level as compared to their respective controls. The low dose of T₃ decreased the plasma amino acid level from 16.43±3.77 to 12.04±0.97 mg/ml but at mid and high doses of L-T₃, the plasma amino acids of the bird increased from 87.33±2.60 mg/ml for control to 105±3.46 mg/ml for high dose and 102±4 mg/ml for mid dose. The plasma protein level was considerably increased at the low dose of T₃, 20.99±2.6 mg/ml for control to 29.20±1.7 mg/ml. On the contrary, the mid and high doses of T₃ caused decreased of plasma protein levels from 24.14±1.5 mg/ml for control to 19.45±1.13 mg/ml for mid dose and 19.22±0.6 mg/ml for high dose. Plasma glucose levels at mid dose was found to be 1.13±0.06 mg/ml and at high (5 µg) dose was 1.21±0.1 mg/ml and for the control was 0.921±0.05 mg/ml. The results suggested that administration of both mid and high doses of L-T₃ to normal intact birds caused hyperglycemia as showed by the increased in plasma glucose levels. The energy demand imposed upon by the prolong administration of high dose of L-T₃ caused the utilization

of plasma protein to meet the energy requirements of the birds and therefore, this may also be considered for increasing the free amino acid levels in the plasma.

Key words : L-T₃, tree sparrow, plasma, glucose, protein and amino acid

L-thyroxine (L-T₄) and Tri-iodothyronine (L-T₃) are the two major thyroid hormones in birds^{18,30,31,32,37}. In birds L-T₄ and L-T₃ perform a number of diverse functions in the regulation of the body metabolism^{4,17,39,44,48,50}, tissue respiration^{28,36,47,48,49}, molting^{1,28}, pigmentation^{17,22,44}, erythropoiesis^{10,33,35,48,50}, reproduction^{1,21,22,27,45,54}, hatching process^{2,14}, and light responses^{5,8,15,27,38,46}, etc.

The plasma L-T₃ level was found to be better related to the metabolic rate²⁶, heat²⁰, O₂ consumption³, etc. It was also reported that L-T₃ had a greater calorogenic and lipogenic effect in bird^{30,31}. L-T₃ therefore, seems to be thyroid hormone active in energy metabolism of birds. However, there is a general paucity of information on the effect of L-T₃ at cellular level. The reports on the possible involvement of L-T₃ in regulation of intermediary metabolism of carbohydrates are limiting. The thyroid also does an important role in the metabolism of protein^{34,40,42,51}, L-T₄ was reported to be protogenic in a number of vertebrates^{43,53}. Reports also suggested that depending on the dose administered L-T₄ produced either a protein anabolic or protein catabolic effects⁴². L-T₄ also caused rapid degradation of protein to meet the energy requirements of the animal, when insufficient carbohydrates and fats were available to supply the energy for various metabolic

process. However, the precise role of T₃ in regulating the protein metabolism in birds has received relatively little attention. Therefore, the present study on the dose dependent effects of L-T₃ on plasma levels of glucose, proteins and amino acids was taken on the tree sparrow, *Passer montanus malaccensis*.

The tree sparrow, *Passer monanus malaccensis* undertaken during the present study is a non-migratory passerine, omnivorous birds distributed widely in Indian subcontinent (The experiment was conducted in year 1990). For each set of experiment adult species of approximately equal size weighing 17±3 gm was acquired time to time from the local supplier and released in the outdoor aviary exposed to ambient condition of temperature and light. The birds were laprotomised, weighed and tagged. All the experimental birds were then divided into different groups and after a week of acclimatization, they were given different hormonal treatment.

For studying the dose dependent effect of tri-iodothyronine (L-T₃) on intermediary metabolism of carbohydrates and protein, experimental birds were divided into 3 groups of 5 number each. Birds of group I, group II, and group III were given 1 µl (low dose), 2.5 µl (mid dose), and 5 µl (high dose) of L-T₃ respectively, in 0.1 ml of normal saline solution. In all the experiments a parallel control group injected with equivalent quantities of saline was

also studied. All injections were made intramuscularly over a period of ten days and daily doses of the hormone (s)/drug (s) given was in 0.1 ml of respective vehicle. The hormone (s)/drug (s) was given at the same hours of the day in order to avoid complication in terms of diurnal changing response²⁴. Twenty-four hours after the last injection, birds were sacrificed and blood was collected immediately in tubes, previously rinsed with anticoagulant.

Biochemical analysis :

The plasma portion of blood was separated by centrifugation. The blood was centrifuge for fifteen minutes at 3000 rpm and plasma was obtained. Then, glucose, amino acids and proteins of the plasma were estimated.

Plasma glucose :

Plasma was deproteinized with 5 % trichloro acetic acid (TCA) and centrifugation was performed at 3000 rpm for ten minutes to remove precipitated proteins. The supernatant was neutralised to pH 7.0±0.2 with 10 % NaOH for glucose estimation. Determination of the glucose levels of the deproteinized plasma were using the method of Dubois *et al.*¹¹.

Total plasma protein :

Lowry *et al.*²³ method was followed for the estimation of total amount of in plasma. For precipitating the protein, TCA was utilized. The precipitate after centrifuging was dissolved in aqueous solution of normal NaOH. The change in the colour by the addition of Folin-

Ciocalteau reagent to the solution was measured at 680 nm.

Plasma amino acid :

Free amino acid levels of plasma were estimated by colorimetric method¹⁹. Aliquots of plasma amino acid solution were treated with ninhydrin reagent and ethanol and the colour produced was measured at 550 nm.

For all the experiments, 3 replicates were processed for each sample. The values were expressed as mg/ml. Statistical analysis of significance of quantitative data between the control and treated group was calculated using student "t" test¹³.

Effects of tri-iodothyronine (L-T₃) on plasma levels of glucose, protein and amino acids of tree sparrow, *Passer montanus malaccensis* are presented in Table-1. Administration of different doses of T₃ to normal intact birds produced a significant dose dependent elevation in plasma glucose level as compared to their respective controls, free amino acid levels in the plasma was also found to be subsequently elevated when both the mid high dose of T₃ were administered. The low dose of T₃ on the other hand caused insignificant depletion of the plasma amino acids of the bird. The low dose (1 µl) of T₃ caused depletion (16.43±3.77 to 12.04±0.97 mg/ml) of the plasma amino acid levels of the bird (Table-1). And at mid dose (2.5 µg) and high dose (5 µg) of L-T₃, the plasma amino acids of the bird also increased from 87.33±2.60 mg/ml for control to 105±3.46 mg/ml for high dose and 102±4 mg/ml for mid dose (Table-1). The plasma protein level was considerably

Table-1. Effect of Tri-iodothyronine (L-T₃) on plasma levels of glucose, protein and amino acids of tree sparrow, *Passer montanus malaccensis*

Treatment for 10 days	Dose (µg/bird/day)	Body weight (gm)		Plasma (mg/ml)		
		Initial	Final	Glucose	Protein	Amino acid
T ₃	1 µl	17±0.57	18.16±0.44	3.40±0.45	29.20±1.7 _b	12.04±0.97
Normal saline	0.9 % NaCl	17.15±0.12	18.07±0.14	2.99±0.06	20.99±2.6	16.43±3.77
T ₃	2.5 µg	16.33±0.7	15.5±0.7	1.13±0.06	19.45±1.13	102±4 _b
	5 µg	18.2±0.3	17.3±0.3	1.21±0.1	19.22±0.6 _b	105±3.46 _a
Normal saline	0.9 % NaCl	16.83±1.11	19±1	0.921±0.05	24.14±1.5	87.33±2.60

Values are expressed as M ± SE

a,P<0.01; b,P<0.05

increased at the low dose of T₃ (1 µl), 20.99±2.6 mg/ml for control increased to 29.20±1.7 mg/ml (Table-1). The mid and high doses of T₃ on the contrary caused depletion of plasma protein levels. At mid (2.5 µg) and high (5 µg) doses of T₃, the levels of plasma protein decreased from 24.14±1.5 mg/ml for control to 19.45±1.13 mg/ml for mid dose and 19.22±0.6 mg/ml for high dose (Table-1). Plasma glucose levels at mid (2.5 µg) dose was 1.13±0.06 mg/ml and at high (5 µg) dose was 1.21±0.1 mg/ml and for the control was 0.921±0.05 mg/ml (Table-1). The present study did not use L-T₄ but L-T₃ was used, earlier studies were also reported where L-T₃ was used in passerine birds, T₃ may be the hormone utilized at the cellular level^{7,29}. Many reports have showed the plasma L-T₃ level was found to be better related to the metabolic rate, heat, O₂ consumption, etc and L-T₃ had a greater calorogenic and lipogenic effect in bird and thus, L-T₃ may be the thyroid hormone active in energy metabolism of birds^{3,20,26,30,31}.

The results of the present study suggested that the administration of both mid and high doses of L-T₃ to normal intact birds caused hyperglycemia as indicated by an increased in plasma glucose levels as compared to their respective controls. L-T₃ was observed to play important role in the metabolism of protein^{16,34,43,51,53}. In tree sparrow, the low dose of L-T₃ was found to have a protein anabolic effect. The increased in the plasma protein level may be due to the greater retardation of catabolic than that on anabolic process⁵¹. This, in tree sparrow like as in other vertebrates, the low dose level of T₃ was found to be more pronounced on anabolic process of protein rather than its catabolic process^{16,41,42}. This view may be further accompanied by the depletion of total amino acid of plasma. The energy demand imposed upon by the prolong administration of high dose of L-T₃ was great enough and thus, the plasma protein was utilised to meet the energy requirements of the birds. Moreover,

increased oxidation destruction of plasma protein may also be considered to raise the free amino acid levels in the plasma. Thus, depending on the dose administered L-T₃ may be either protein anabolic or protein catabolic. Similar results of thyroid hormones were also observed in a number of other vertebrates' species^{9,34}. In general thyroid hormones were known to modify plasma proteins^{12,25}. As in tree sparrow, a dose dependent effect of L-T₄ on protein metabolism was also reported in other vertebrates^{35,52}.

In birds, two major thyroid hormones, L-thyroxine (L-T₄) and Tri-iodothyronine (L-T₃) are important in the regulation of the body metabolism performing a number of diverse functions. The dose dependent effects of L-T₃ on plasma levels of glucose, proteins and amino acids on the tree sparrow, *Passer montanus malaccencis* were studied and reported. Different doses of T₃ to normal intact birds produced a significant dose dependent elevation in plasma glucose levels. The low dose of T₃ decreased the plasma amino acid level and at mid and high doses of L-T₃, the plasma amino acids of the bird increased. The plasma protein level was increased at the low dose but decreased at the mid and high doses. From the results of the study, it may be concluded that at mid and high doses of L-T₃ to normal intact birds may cause hyperglycemia as the levels of plasma glucose increased. The long exposure of the birds at the high dose of L-T₃ may cause the utilization of plasma protein to meet the energy requirements and therefore, may have increased the plasma free amino acid levels.

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