Profile of thyroid hormone in male Layer chickens given by testosterone

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Abstract. The aim of the study was to evaluate the administration of Testosterone toward growth, levels of Triiodothyronine (T3) and Thyroxine (T4) in male Layer chicken. Forty male Layer chickens strain Isa Brown was used in this study. Chickens were divided into 2 groups, control and treatment groups (given commercial testosterone) at a dose of 3 mg/kg body weight/day for 5 weeks. The results showed the growth of chickens treated by Testosterone increased significantly from week 1 to week 5 with a range between 59.43 ± 0.76 grams to 332.50 ± 0.79 grams, (P < 0.05). However, due to treatment differences, the results obtained that the administration of Testosterone cannot accelerate the growth of male Layer chickens (P > 0.05). Even in the week 3 and week 4, the growth rate is lower than the control. The levels reduction of T3 start from 4.91 ± 0.98 ng/mL to 1.38 ± 0.09 ng/mL, while T4 levels start from 1.01 ± 0.85 ug/mL to 0.84 ± 0.21 ug/mL. It could be concluded that the administration of testosterone can reduce T3 and T4 levels but cannot accelerate the growth rate of male Layer chickens.

1. Introduction
The growth of male chickens involves a variety of factors and hormones, one of which is Testosterone and- Thyroid hormones [1]. The two hormones are related to supporting growth. In fact, on the growth of Emberiza bruniceps's feathers, thyroid activity is strongly influenced by gonadal hormones [2]. Because of its importance and seen as beneficial, breeders often give Testosterone to get faster growth, longer crowing, and get more beautiful feathers color in their poultry.

Previous research has mentioned that chemical aromatase blockers such as Fadrozol, can increase Testosterone levels as much as 248% on the 4th day after administration. On the same day, there was also an increase in T3 and T4 hormones [3]. Added by Astuti et al. [4] that natural aromatase blockers can stabilize testosterone in mice. Although the effect of continuous testosterone administration on DOC is not known certainly, it is feared that it can lead to negative effects such as Down-Regulation. Therefore, this research will be conducted on male Layer chickens that are given synthetic Testosterone continuously in relation with the growth and activity of the thyroid hormone. The
specific research objective was to evaluate the growth rate of Layer chickens and thyroid hormone levels after the chickens were given synthetic Testosterone for 35 days.

2. Materials and methods
The research animals used in the study were 40 DOC Isa Brown male Layer chickens. Each group of animals kept in a breeder cage size 2 x 2 x 1 m, given partitions and lights following the development of age. The feed given was commercial product starter feed. Furthermore, the animals were divided into 2 groups: the control group was given NaCl 1 mL orally and the treatment group was administered a commercial synthetic Testosterone was given by the subcutaneous injection of 0.1 mL (3 mg/day/head) for 35 days.

Blood collection of 0.5 to 1 mL is carried out through the brachial vein after the animal is anesthetized using ketamine and xylazine at a dose of 100 mg/kg body weight and 10 mg/kg body weight, respectively. The blood collection was done just before the treatment and repeated in the first week to the fifth week. The blood that has been drawn was then centrifuged at a speed of 3000 rpm for 20 minutes. The serum formed was taken into a microtube, stored in a freezer at -20 ºC until the hormone T3 and T4 were assayed. Assay T3 and T4 hormones were carried out using the Enzyme Link Immunosorbent Assay (ELISA) method.

The data obtained were analyzed using Analysis of Variance (ANOVA). In case there are significant differences, the test is continued using Tukey HSD. All methods implemented have received approval from the Ethics Commission of Integrated Research and Testing Laboratory with certificate number 00059 / 04 / LPPT / XI / 2019.

3. Results and discussion
3.1. Profile of Body Weight Gain in 5 Weeks Treatment
Chicken growth involves a variety of hormones and several other factors. In this study, chicken weight increased significantly from week 1 to week 5 (Table 1 and Figure 1).

Andriyanto et al. [5] states that chicks will grow rapidly in their first 3 weeks, then decrease slightly, and will be stable when they reach adulthood. Since traditionally kept, the chicken growth in this study is anticipated to be lower than the growth of Layers that are grown in a modern way, as stated by Choo et al. [6]; Simeon [7].

| Treatment        | Week 1  | Week 2  | Week 3  | Week 4  | Week 5  |
|------------------|---------|---------|---------|---------|---------|
| Control          | 62.04±0.17<sup>a</sup> | 91.60±0.50<sup>d</sup> | 161.30±0.47<sup>c</sup> | 226.8±0.90<sup>b</sup> | 323.90±0.76<sup>a</sup> |
| Testosterone     | 59.43±0.76<sup>c</sup> | 118.15±0.71<sup>d</sup> | 139.50±0.39<sup>c</sup> | 200.5±6.63<sup>b</sup> | 332.50±0.79<sup>a</sup> |

<sup>a–e</sup> Different letters in the same row show a significant difference (P < 0.05).

![Body Weight (g)](image_url)

**Figure 1.** Profile of male Layer chickens body weight gain treated by Testosterone. The body weight of treatment lower than control group at the week 3 and 4.
Comparison between the control group and treatment (given Testosterone) shows there was no significant difference (P > 0.05), even at the 3rd and 4th weeks, body weight in the control group was higher than that of the Testosterone group (Figure 1). Similar to Andriyanto et al. [5] who reported that giving Testosterone to broilers at the age of 18-21 days reduced body weight.

3.2. Profile of Triiodothyronine (T3) Levels during the Treatment
In poultry, the thyroid hormone has important functions. Aside from metabolism and growth, this hormone also functions oxygen consumption, especially for chicks [8]. Based on the results, the profile of hormone Triiodothyronine (T3) level has decreased from week 1 to week 5. Statistically, significant differences in the treatment group (P < 0.05) occurred at week 1 and week 2 as well as week 4 and week 5 (Table 2). Although T3 levels continuously go down, male Layer chickens body weight rises gradually from week 1 to week 5 (Table 1).

![Figure 2. Level of Triiodothyronine (T3) during the treatment. The profile decreased from week 1 up to week 5.](image)

This is a match with the statement of Elwahesh et al. [9] that an increase in chicken body weight is related to a decrease in the level of the hormone Triiodothyronine (T3). Moreover, Ritchie [10] explained that the fall of T3 that circulates under metabolic requirements, stimulates the anterior pituitary to release TSH. Unlike the adult roosters, where the Thyroid Releasing Hormone (TRH) produced by the hypothalamus does not stimulate TSH secretion.

3.3. Profile of Hormone Thyroxine (T4) Levels during the Treatment
The levels of Thyroxine (T4) during giving testosterone can be seen in Table 3 and Figure 3.

![Figure 3.](image)

**Table 2.** Profile of T3 levels (ng/mL) during the treatment

| Treatment      | Week 1      | Week 2      | Week 3      | Week 4      | Week 5      |
|----------------|-------------|-------------|-------------|-------------|-------------|
| Control        | 4.53±0.89a  | 3.18±0.31bc | 3.93±0.69ab | 2.35±0.90d  | 1.44±0.24d  |
| Testosterone   | 4.91±0.98a  | 3.05±0.76b  | 2.15±0.20bc | 1.95±0.22c  | 1.38±0.09c  |

a–e Different letters in the same row show significant difference (P < 0.05).

**Table 3.** Profile of hormone T4 levels (ug/mL) during the treatment

| Treatment      | Week 1      | Week 2      | Week 3      | Week 4      | Week 5      |
|----------------|-------------|-------------|-------------|-------------|-------------|
| Control        | 0.91±0.14ab | 0.73±0.15b  | 1.21±0.30a  | 0.87±0.11ab | 0.91±0.24ab |
| Testosterone   | 1.01±0.85a  | 0.73±0.11a  | 0.88±0.32a  | 0.72±0.11a  | 0.84±0.21a  |

a–e Different letters in the same row show significant difference (P < 0.05).
The decrease of hormone T4 levels has been started since the 1st and 2nd weeks and then increase in the 3rd week. The increase of T4 levels only occurs in the 3rd week then drops again in the 4th to 5th week, in line with the rapid improvement of body weight. Based on its function, T4 has more roles in thermoregulation. It is proven that in winter, T4 concentrations shoot up than summer [10]. This happens as during the winter the thyrotrop cells in pars tuberalis are secreted, that way TSH stimulates epidermal cells (tanisit) in the 3rd ventricle to express the type 2 deiodinase (DIO2) enzyme which converts the prohormone thyroxine (T4) into bioactive component triiodothyronine (T3).

![Figure 3. Level of Thyroxine (T4) during the treatment.](image)

Furthermore, Hanon et al. [11]; Kasiyati [12] reported that T3 stimulates the release of GnRH neuroendocrine at the end of the hypothalamic media eminasia (ME). TSH affects hypothalamic epidermal cells to stimulate the activity of DIO2 enzymes resulting in the conversion of T4 to T3. T3 hormones are responsible for regulating the hormones of the GnRH system [13].

4. Conclusion
From the results and discussion, it appears that in 35 days the body's metabolic rate is increased. This is confirmed by the decrease of T3 and T4 levels as one of the hormones that function to carry out the body's metabolism. However, the decrease of T3 and T4 as an indicator of increased metabolism is anticipated to not only be focused on growth. This is proven even though continuous testosterone is given for 35 days, it cannot accelerate the growth rate of the male Layer chickens.

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