

## Effects of Propylthiouracil-Induced Hypothyroidism on Plasma Lipid Table in Rabbits

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**Abstract:** Hypothyroidism is commonly associated with hyperlipidemia. The aim of this study was to investigate the effect of Propylthiouracil-induced hypothyroidism on the plasma lipid profile in rabbits. This investigation was performed on male, white New Zealand (*Oryctolagus cuniculus huxley*) rabbits. Twenty rabbits were used; each of the two groups (hypothyroidism and control) consisted of ten animals. Hypothyroidism was induced by oral administration of Propylthiouracil 50 mg.kg<sup>-1</sup> for three weeks.

The mean values of Triiodothyronine (T<sub>3</sub>), thyroxine (T<sub>4</sub>), free T<sub>3</sub> (FT<sub>3</sub>) and free T<sub>4</sub> (FT<sub>4</sub>) were 121.7±32.7 ng/dl, 1.81±0.60 µg/dl, 2.36±9b92 ng/dl and 0.43±0.22 p mol/dl in the control group, and 88.4±19.2 ng/dl, 1.15±0.37 µg/dl, 1.48±0.44 ng/dl and 0.18±0.11 p mol/dl in the hypothyroidism group, respectively (p<0.05 for all). Levels of total plasma triglyseride (TG), high-density lipoprotein cholesterol (HDL-C), total cholesterol (TC) and apolipoprotein-B (Apo-B) were recorded as 44.2±8.57, 12.2±2.7, 40.4±14.9 and 87.2±15.2 mg.dl<sup>-1</sup> in the control group, and 97.9±29.8, 23.2±2.9, 67.2±13.5 and 147.4±55.7 mg.dl<sup>-1</sup> in the hypothyroidism group, respectively (p<0.001 for all parameters). The results showed that T<sub>3</sub>, T<sub>4</sub>, FT<sub>3</sub> and FT<sub>4</sub> decreased in the hypothyroidism group, but lipid fractions were higher than in the control group. In addition, a significant negative correlation was found between T<sub>4</sub>-TC (r=-0.739, p<0.05) and T<sub>4</sub>-Apo-B (r=-0.666, p<0.05).

We concluded that lipids may be an indicator for hypothyroidism, but that thyroid hormones are better indicators that lipids.

**Key Words:** Hypothyroidism, lipid, rabbit.

### Tavşanlarda Propiltiourasil ile Oluşturulan Deneysel Hipotiroidizmin Plazma Lipid Profili Üzerine Etkisi

**Özet:** Hipotiroidizm genellikle hiperlipidemi ile ilişkilidir. Bu çalışmanın amacı, Propylthiouracil ile oluşturulan hipotiroidizmin plazma lipidleri üzerine etkisini araştırmaktır. Çalışmada Yeni Zelanda ırkı, beyaz, erkek tavşanlar (*Oryctolagus cuniculus huxley*) kullanıldı. On tavşan kontrol, 10 tavşan da hipotiroidizm grubu olarak ayrıldı. Hipotiroidizm, oral yoldan üç hafta süre ile 50 mg.kg<sup>-1</sup> vücut ağırlığına Propylthiouracil verilerek oluşturuldu.

Deney sonunda kontrol grubu hayvanların (T<sub>3</sub>), (T<sub>4</sub>), FT<sub>3</sub> ve FT<sub>4</sub> değerleri sırasıyla 121.7±32.7 ng/dl, 1.81±0.60 µg/dl, 2.36±0.92 ng/dl, 0.43±0.22 p mol/dl iken hipotiroidili hayvanlarda 88.4±19.2 ng/dl, 1.15±0.37 µg/dl, 1.48±0.44 ng/dl, 0.18±0.11 p mol/dl idi (p<0.05). Ayrıca, plazma total TG, HDL-C, TC ve Apo-B seviyelerinin ise, kontrol grubunda sırasıyla 44.2±8.57, 12.2±2.7, 40.4±14.9, 87.2±15.2 mg.dl<sup>-1</sup>, hipotiroidi grubunda 97.9±29.8, 23.2±2.9, 67.2±13.5, 147.4±55.7 mg.dl<sup>-1</sup> olduğu belirlendi (tüm parametreler için; p<0.001). Sonuçlar gösterdi ki, hipotiroidizmde T<sub>3</sub>, T<sub>4</sub>, FT<sub>3</sub> ve FT<sub>4</sub> değerleri azalmıştır. Ayrıca T<sub>4</sub>-TC (-0.739, p<0.05) ve T<sub>4</sub>-Apo-B (-0.666, p<0.05) arasında önemli oranda negatif ilişki bulunmuştur.

Sonuç olarak, lipidler hipotiroidizm için bir indikatör olabilirler ancak, tiroid hormonları lipidlerden daha iyi birer indikatördürler.

**Anahtar Sözcükler:** Hipotiroidizm, lipid, tavşan.

### Introduction

It is well-known that hypothyroidism is associated with hypercholesterolemia and increases the risk of atherosclerotic disease (1-3). There is general agreement that total and LDL-C and triglyceride levels increase in hypothyroid (3). However, the influence of hypothyroidism on HDL-C has been variable (4-6). In addition, clinical and epidemiological studies have

indicated a number of risk factors for atherosclerosis. Among these, elevated TC and LDL-C levels were positively correlated with the prevalence of coronary artery disease (7-11).

Hyperlipidemia, observed in hypothyroidism, is a metabolic result currently treatable with thyroid hormones. Before the availability of sensitive thyroid hormone analysis, increased serum or plasma cholesterol

levels were accepted as important evidence supporting the diagnosis of hypothyroidism (12), but classical signs and symptoms of clinical hypothyroidism may not be observed when it is mild or moderate (13).

In a widespread lipid screening program, patients with thyroid deficiency may be found in increasing numbers among hyperlipidemic individuals rather than in the general population, and blood lipid levels may be affected by ethnic factors.

Nonetheless, there is still great contradiction between results. Therefore, the purpose of this study was to assess the predictive value of various lipids, lipoprotein and apolipoprotein in discriminating between experimental hypothyroidism and euthyroidism.

### Material and Method

**Material:** The chemicals, Propylthiouracil from Sigma Chem. Co., EDTA from E. Merck, AG. and kits from Diagnostic Products Corporation were obtained.

This investigation was performed on male, white New Zealand (*Oryctolagus cuniculus huxley*) rabbits weighing 2.5 to 3 kg. Twenty rabbits were studied, each of two groups consisting of ten animals. They were maintained under a temperature of  $20\pm 2^{\circ}\text{C}$  and a daily light/dark cycle, and supplied with normal food water ad libitum.

**Induction of Hypothyroidism:** Hypothyroidism was induced orally with Propylthiouracil (50 mg.  $\text{kg}^{-1}$  per day) for three weeks (14). Propylthiouracil has an antithyroid effect on animals. Because the compound prevents the transformation of iodines to iodine, iodine was not able to enter the thyroid gland. Thus, since the residues of thyrosine and iodine are not bound, monoiodothyronine and diiodothyronine do not form. Furthermore, propylthiouracil prevents the formation of  $\text{T}_3$  into  $\text{T}_4$  (15).

After feeding for three weeks, the animals were anesthetized for half an hour. As anesthetics, 5 mg xylazine hydrochloride (Rompun, Bayer) and 20 mg ketamine hydrochloride (Ketalar, Parke-Davis) were used (14). Five milliliters of blood was collected by cardiac puncture, and immediately placed into tubes containing EDTA. The plasma samples were obtained by centrifuging blood samples at 3,000 rpm for 30 minutes at  $4^{\circ}\text{C}$ . Measurements of lipid and hormone levels were carried out in these plasma samples.

**Measurement of Lipid and Hormone Levels:** Plasma lipid levels were measured by autoanalyzer (Hitachi 705, Japan) using a kit (DPC; Diagnostic

Products Corporation, CA 90045, USA). Furthermore, plasma  $\text{T}_3$ ,  $\text{T}_4$ ,  $\text{FT}_3$  and  $\text{FT}_4$  levels were measured with RIA commercial kits (DPC; Diagnostic Product Corporation USA).

**Analysis of Data:** Means and standard deviations were calculated according to the standard methods. The Mann Whitney U-Test was used for differences between means, Pearson's Correlation Test for correlations. The significance level was 0.05 for all tests.

### Results

In this study, the effect of hypothyroidism on plasma lipid levels were revealed in hypothyroid rabbits and compared to control rabbits. Hypothyroidism was confirmed by  $\text{T}_3$ ,  $\text{T}_4$ ,  $\text{FT}_3$  and  $\text{FT}_4$ , which were significantly lower than those of the control group. Body weight and plasma  $\text{T}_3$ ,  $\text{T}_4$ ,  $\text{FT}_3$  and  $\text{FT}_4$ , concentrations in the two groups are shown in Table 1. There was no difference in body weight between the groups at the beginning. However, in the hypothyroidism group, weight increased at the completion of the study, but the increase was not significant. Plasma TG, HDL-C, TC and Apo-B levels in the two groups of rabbits are shown in Table 2.

As seen in Tables 1 and 2, in the hypothyroidism group, there was a significant decrease in plasma  $\text{T}_3$ ,  $\text{T}_4$ ,  $\text{FT}_3$  and  $\text{FT}_4$ , whereas there was a decrease in plasma TG, HDL-C, TC and Apo-B.

In addition, a significant negative correlation was found between  $\text{T}_4$ -TC ( $r=-0.739$ ,  $p<0.05$ ) and  $\text{T}_4$ -Apo-B ( $r=-0.666$ ,  $p<0.05$ ).

### Discussion

$\text{T}_3$ ,  $\text{T}_4$ ,  $\text{FT}_3$  and  $\text{FT}_4$  values were significantly lower, but all kinds of lipid values were higher in the hypothyroidism group than in the controls.

According to these results, it seems that all the lipid types examined in this study provide additional information on hypothyroidism. We also found that plasma hormones, which are associated with thyroid function levels, are better indicators of hypothyroidism than plasma lipids. These findings are in accordance with other studies, in which humans were generally used as study subjects (16-24).

The result of this study indicated that, in rabbits with hypothyroidism, plasma lipid levels significantly increased above normal values. To date, no study examining plasma lipids in the hypothyroid state has been done on rabbits,

	Control	Hypothyroidism	P
Number of animals	10	10	
Body weight* (g)	2760 ± 155	2846 ± 137	>0.05
T <sub>3</sub> (ng.dl <sup>-1</sup> )	121.7 ± 32.7	88.4 ± 19.2	<0.05
T <sub>4</sub> (µg.dl <sup>-1</sup> )	1.81 ± 0.60	1.15 ± 0.37	<0.05
FT <sub>3</sub> (ng.dl <sup>-1</sup> )	2.36 ± 0.92	1.48 ± 0.44	<0.05
FT <sub>4</sub> (pmol.dl <sup>-1</sup> )	0.43 ± 0.22	0.18 ± 0.11	<0.05

\* : at the beginning of the study.

Table 1. Body weights of the rabbits and plasma hormone levels (Mean±SD).

	Control	Hypothyroidism	P
HDL-C (mg.dl <sup>-1</sup> )	12.2 ± 2.7	23.2 ± 2.9	<0.001
TG (mg.dl <sup>-1</sup> )	44.2 ± 8.57	97.9 ± 29.8	<0.001
TC (mg.dl <sup>-1</sup> )	40.4 ± 14.9	67.2 ± 13.5	<0.001
Apo-B (mg.dl <sup>-1</sup> )	87.2 ± 15.2	147.4 ± 55.7	<0.001

Table 2. Plasma lipid levels (Mean±SD).

so we were not able to compare our results with those of previous studies. Although a number of studies have been carried out on patients with hypothyroidism comparing lipids with regard to lipoprotein and apolipoprotein levels, the results mostly conflict with each other and with our findings. Parle et al. (17) observed no significant difference in fasting plasma lipids between patients with hypothyroidism and controls. These results are thus contradictory to ours. Hoogerbrugge et al. (18) indicated that TC and LDL-C were higher in hypothyroid rats than in controls. The serum levels of TC, HDL-C, LDL-C were found to be higher in hypothyroidism subjects than in controls, decreasing after treatment (19). Although the materials of the studies are different, this finding is in agreement with ours. Lanol et al. (20) found a statistically significant difference in the increase in TC and TG, while HDL-C was normal in subjects with overt hypothyroidism.

Muis et al. (21) found that the plasma concentrations of TG, TC, HDL-C and Apo-B increased in hypothyroid states, supporting our results. Lopez and Morales (22) reported that total serum lipids decreased in hypothyroid pregnant rats. This report is in contradiction with our findings and those of other investigators. Muis et al. (24) and Aviram et al. (25) demonstrated in humans, and Ibrahim et al. (24) in Nubian goats (*Capra hircus*), that lipids and their fractions were increased in hypothyroid state. These conflicting data may be due to the use of

different animals as study subjects or different drugs for hypothyroidism.

Even though several theories have been proposed to explain the lipid mechanism lipoprotein and apoprotein differences in thyroid dysfunction are not well understood. Some investigators have suggested that the increase in kinds of lipid levels in hypothyroidism patients results from the hormones of thyroid influence on lipid metabolism (24). Other possibilities for the metabolism of lipids in hypothyroidism are the presence of lipid activating factors in hypothyroidism, and a decrease in hepatic lipase activity. This study also brought out the fact that plasma lipids and their fractions are significantly higher in hypothyroid rabbits than in the control group. Thus our results support these possibilities. However, further studies will be required to elucidate the metabolic pathways of lipids and their fractions.

In conclusion, this study demonstrated that plasma TG, HDL-C, TC and Apo-B levels were elevated in hypothyroid rabbits with 3 weeks of Propylthiouracil-induced hypothyroidism in comparison with the control group. However, our data indicate that further experiments should be performed to investigate what is responsible for the elevation of resting plasma lipids and their fractions in hypothyroid rabbits treated 3 weeks previously with Propylthiouracil.

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