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# SEX AND AGE-DEPENDENT CHANGES IN LIPID PROFILE AND ANTIOXIDANT ENZYMES IN IRAQI PATIENTS WITH HYPOTHYROIDISM

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## Abstract

A common endocrine condition called hypothyroidism is brought on by inadequate thyroid hormone production. It affects approximately 0.6% of the population. Thyroid hormones have physiological and clinical effects on carbohydrate and lipid metabolism. The purpose of this study is to evaluate, by age and sex, how thyroid hormone functions affect lipid profile parameters as well as antioxidant enzyme levels in Iraqi hypothyroidism patients. Using spectrophotometric enzymatic techniques based on absorbance measurements in the formation of a colored complex, serum levels of catalase (CAT), superoxide dismutase (SOD), and glutathione peroxidase (GPX), the lipid profile (triglycerides (TG), total cholesterol (TC), high-density lipoproteins (HDL), low-density lipoproteins (LDL), and very low-density lipoproteins (VLDL). Results indicated that patients with hypothyroidism had significantly higher CAT concentration (*P<0.0001) than healthy people from control group in all age and sex categories. Glutathione peroxidase (GPX) & superoxide dismutase (SOD), two antioxidant enzymes of hypothyroid people, were dramatically reduced by exposure to oxidative stress (P<0.0001). Lipid profile studies found TC, TG, LDL, and VLDL in patients with hypothyroidism were significantly higher among both younger and older age groups. whereas the levels of HDLa dropped. Age was found to be strongly positively correlated with both GPX and fat (TC, LDL, and VLDL). Higher cholesterol and antioxidant enzyme deficiencies were shown to be statistically significant differences between the patient group and the control group among age- and sex-matched groups, indicating the clinical importance of oxidative stress.

**Keywords**: *Hypothyroidism, Total Cholesterol, Glutathione peroxidase, Catalase, HDL, Age variable.*

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## 1. Introduction

A decrease in thyroid function brought on by a reduction in thyroxine (T4) and triiodothyronine (T3) production is the hallmark of hypothyroidism. Low levels of T3 and T4 stimulate secretion of thyroid-stimulating hormone (TSH). The basal metabolic rate is reduced by other clinical processes and is influenced by thyroid hormones (1,2). The essential reasons for hypothyroidism cases are different, such as surgical removal of the thyroid, treatment with radiation, autoimmune disease and inborn effects. Given the importance of hypothyroidism disease, specialists classify the biochemical disorder according to the organ of origin into three types: primary, secondary and tertiary. The primary hypothyroidism is the most widespread disorder, and this process takes place when the thyroid gland is infected by physiological damage leading to clinical unrest or turmoil, problems; the hormones in this gland will not be secreted. The primary hypothyroidism is medically diagnosed by anabolism of immune cells and their capability to anabolize the thyroid hormones (3,4). The pituitary gland's inability to create enough TSH, which stimulates the thyroid to make T3 and T4, is the cause of the second kind of hypothyroidism. Other reasons for secondary hypothyroidism include damage to the pituitary gland, tumours, and radiation, as well as tertiary hypothyroidism, which occurs when thyrotropin-releasing hormone (TRH) is not functioning properly, causing the pituitary gland to fail to produce TSH. The type of thy hypothyroidism is very rare (5, 6).

Numerous biochemical indicators, including antioxidants and enzymes like glutathione peroxidase (GPX), catalase (CAT), and superoxide dismutase (SOD), have been clinically linked to the onset of hypothyroidism. It is crucial to examine and confirm the levels of these enzymes from hypothyroid patients' blood serum in order to monitor the severity of this dangerous condition. Lipid profile is also required in case of hypothyroidism disease since the assorted classes of lipids. The effect on the biological system of the living cell of the human being infected with hypothyroidism. Triglycerides (TG), total cholesterol (TC), high-density lipoproteins (HDL), low-density lipoproteins (LDL), and very low-density lipoproteins (VLDL) are among the lipid profiles that are subject to change. Additionally, the pre-studies verified the biochemical connection between enzymatic antioxidants and the lipid profile, as well as the onset and severity of hypothyroidism (7,8,9). The biochemical and clinical roles of antioxidant enzymes, such as CAT, GPX, and SOD, as well as lipid profiles as chemical markers to track hypothyroidism disease activity based on age, smoking, sex, marital status, blood groups, family history, as well as body mass index have been shown in a number of preliminary studies (10, 11, 12). The goal of the current study was to ascertain the activity levels of antioxidant enzymes (CAT, GPX, & SOD) and lipid profile markers (TG, TC, HDL, LDL, and VLDL) in the blood serum of hypothyroid patients by sex.

### Materials and Methods

A total of 193 persons (102 male and 92 female) participated in this study, and they were divided into 95 hypothyroidism patients and 98 healthy individuals, who were considered the control group. All participants underwent clinical examination, and they were divided into four age categories: the first (ages 11–22), second (ages 23–35), third (ages 36–48), and fourth (ages 49–67).

### Ethical Approval

The official order was issued by the Basrah Health Directorate, the unit for training and human management within the knowledge centre department of research.

### Blood Sampling Location

Al-Fayhaa Teaching Hospital in Basrah Governorate, South Iraq, collected complete blood samples at 9:00 AM from a variety of hypothyroidism patients and healthy individuals serving as control groups. A qualified nurse collected entire samples from both healthy individuals and people with hypothyroidism. Patients with hypothyroidism as well as control groups had five milliliters of venous blood taken; all samples were then placed in vacutainer tubes. Then they underwent centrifugation at 5000 revolutions per minute (rpm) for 7 minutes. While the remaining blood was put in special tubes to separate the plasma, all sera were collected and kept at 27°C for the evaluation of clinical biochemical characteristics. After thoroughly cleaning the erythrocytes with sodium chloride (9% w/v), the mixture was lysed using deionized water at a ratio of 1:1/v/v (13,14).

### Assessment of Biochemical Variables

Using an enzymatic kit and ELISA, the activity of antioxidant enzymes (GPX, CAT, and SOD) in whole-blood samples from hypothyroid patients and healthy individuals was assessed (15). The lipid profile (TG, TC, HDL, LDL and VLDL) were assessed by spectrophotometric enzymatic methods as absorbance measurement in light of the color formation complex three species with all blood samples from hypothyroid patients and healthy persons (16, 17).

### Statistical analysis

SPSS version 25 (IBM, Armonk, New York) was the program used. Descriptive statistics, such as mean and standard deviation (SD), were used to extract the results. Mann-Whitney and Kruskal-Wallis tests were used to examine for differences in quantitative data between the two parameter sets. The r-value was computed using Spearman's correlation coefficient. P-values less than 0.05 were regarded as statistically significant.

**Results**

Hypothyroidism is a complex clinical disorder that affects most people in the world. The biochemical significance of clinical investigation of various chemical parameters confirms the value of monitoring any alterations that may occur during the progression of the dangerous hypothyroidism disease. To determine the function of the oxidants, measurements of antioxidant enzymes, such as CAT, GPX, and SOD activity, in the blood sera of hypothyroid patients should be evaluated (18). The activity of enzymatic antioxidants (SOD, GPx, and CAT) in the blood serum of hypothyroid patients as well as regulate groups in relation to the age factor (Table 1).

**Table (1): Antioxidant enzyme activities (CAT, GPx, as well as SOD) in hypothyroidism and healthy individuals according on age.**

| Age | Groups | CAT(K/ml) | GSH-px(U/L) | SOD(U/ml) |
|---|---|---|---|---|
| 11-22 | Control(N=24) | 4.06±10.28 | 36.85±10.66 | 46.60±34.59 |
| | Patient(N=28) | 43.58±14.84 | 13.78±3.98 | 12.70±3.81 |
| P-value | | <0.0001 | <0.0001 | 0.002 |
| 23-35 | Control(N=24) | 3.20±1.38 | 50.87±30.07 | 67.58±30.07 |
| | Patient(N=28) | 56.15±19.06 | 12.34±2.75 | 12.02±2.75 |
| P-value | | 0.031 | 0.010 | 0.002 |
| 36-48 | Control(N=17) | 3.89±0.61 | 40.76±18.32 | 71.72±31.97 |
| | Patient(N=16) | 72.45±56.30 | 15.53±4.26 | 10.56±1.73 |
| P-value | | 0.031 | 0.010 | 0.001 |
| 49-67 | Control(N=22) | 6.09±10.11 | 30.39±22.37 | 68.84±36.04 |
| | Patient(N=26) | 40.46±15.33 | 19.29±1.58 | 10.71±1.92 |
| P-value | | <0.0001 | 0.152 | 0.001 |

**The results were shown as Mean ±SD, ***P<0.0001, **P<0.001, *P<0.05, and N.S.: Non-significant.**

Assorted concentration values were obtained for the CAT, GPX, and SOD enzymes across age groups. They are represented by (43.53 ± 14.84, 13.78 ± 3.98, 12.7 ± 3.81), respectively, in the first category, but the concentration values were (56.15 ± 19.06, 12.34 ± 2.75, 12.02 ± 2.75) in the second age category. Various enzyme concentrations in the third category: (72.45±56.30, 15.53±4.26, 10.56±1.73). at the same time, the values of concentration of these enzymes are equal to (40.46±15.33,19.29 ± 1.58,10.71±1.92) at the fourth category. In the first, second, third, and fourth categories, all antioxidant enzyme concentrations were significantly lower in patients than in control groups (***p <0.0001). However, compared to healthy individuals in the fourth age group, the same enzyme concentration was considerably greater (p<0.001) in the blood sera of hypothyroid patients.

The action of sex variable is very important to follow the severity, progress and hazard of hypothyroidism disease, especially in old patients. Antioxidant enzyme activity (CAT, GPX, and SOD) in hypothyroid individuals by sex is displayed in Table 2.

**Table (2): Activities of antioxidant enzymes (CAT, GPX, as well as SOD) in the blood sera of hypothyroid and healthy individuals according to sex factor.**

| Sex | Groups | CAT(K/ml) | GSH-px(U/L) | SOD(U/ml) |
|---|---|---|---|---|
| Male | Control(N=48) | 6.54 ±10.47 | 42.11±19.66 | 64.71±36.09 |
| | Patient(N=53) | 47.10±16.04 | 14.75±4.72 | 11.99±3.09 |
| P-value | | <0.0001 | <0.0001 | <0.0001 |
| Female | Control(N=50) | 3.69±1.41 | 39.45±18.85 | 64.54±30.20 |
| | Patient(N=42) | 53.06±32.62 | 14.70±3.63 | 11.67±3.02 |
| P-value | | <0.0001 | <0.0001 | <0.0001 |

**Mean ± SD was used to express the concentrations. *P<0.05, **P<0.001, ***P<0.0001, and N.S.: Not significant.**

Catalase, glutathione peroxidase, and superoxide dismutase showed concentrations of 47.10±16.04, 14.75±4.72, and 11.99±3.09, respectively, in male patients with hypothyroidism. On the other hand, according to sex, the blood sera of female patients with hypothyroidism had concentrations of the same antioxidants of 53.06±32.62, 14.70±3.63, and 11.67±3.02, respectively.

Age and lipid profiles are clinically associated in hypothyroid individuals. Therefore, Table (3) shows aspirating to clarify the serum levels of TG, TC, HDL, LDL, and VLDL according to two age groups in hypothyroid individuals.

**Table (3): lipid profile concentrations (TG, TC, HDL, LDL, & VLDL) with age in both healthy people and hypothyroid patients.**

| Age | Groups | TC (mg/dl) | TG (mg/dl) | HDL (mg/dl) | LDL (mg/dl) | VLDL (mg/dl) |
|---|---|---|---|---|---|---|
| 11-22 | Control (N=22) | 108.26±57.056 | 163.45±39.915 | 43.67±22.306 | 114.72±46.56 | 24.04±19.19 |
| | Patient (N=28) | 200.11±92.985 | 225.93±40.977 | 22.17±12.524 | 140.81±51.082 | 45.47±18.60 |
| P-value | | 0.001 | 0.0001*** | 0.001** | 0.011** | 0.035 |
| 23-35 | Control (N=26) | 108.26±57.056 | 163.45±39.915 | 43.67±22.306 | 114.72±46.56 | 24.04.±19.19 |
| | Patient (N=24) | 200.11±92.985 | 225.93±40.977 | 22.17±12.524 | 140.81±51.082 | 45.47±18.60 |
| P-value | | 0.001 | 0.0001*** | 0.001** | 0.011** | 0.035 |
| 36-48 | Control (N=17) | 108.26±57.056 | 163.45±39.915 | 43.67±22.306 | 114.72±46.56 | 24.04±19.19 |
| | Patient (N=16) | 200.11±92.985 | 225.93±40.977 | 22.17±12.524 | 140.81±51.082 | 45.47±18.60 |
| P-value | | 0.001 | 0.0001*** | 0.001** | 0.011** | 0.035 |
| 49-67 | Control (N=22) | 108.26±57.056 | 163.45±39.915 | 43.67±22.306 | 114.72±46.56 | 24.04±19.91 |
| | Patient (N=26) | 200.11±92.985 | 225.93±40.977 | 22.17±12.524 | 140.81±51.082 | 45.47±18.60 |
| P-value | | 0.001 | 0.0001*** | 0.001** | 0.011** | 0.035 |

**The concentration data were shown as mean ± SD, *** p<0.0001, **p<0.001, *p<0.05, and N.S: Non-significant.**

Different concentration values were measured for the profile (TC, TG, HDL, LDL, and VLDL) in patients with hypothyroidism, with values of (200.11±92.985, 225.93±40.977, 22.17±12.524, 140.81±51.082, 45.47±18.60) in the first age category. In contrast, the same lipid classes showed concentrations of (200.11±92.985, 225.93±40.977,22.17±12.524,140.81±51.082 and 45.47±18.60) in the second category. The concentration values for TC, TG, HDL, LDL, and VLDL were estimated to be equal in the third age group (200.11±92.985,225.93±40.977,22.17±12.524,140.81±51.082, and 18.60±45.47), whereas the same lipid types recorded different concentrations in the fourth age group (200.11±92.985,225.93±40.977,22.17±12.524,140.81±51.082, and 18.60±45.47). The great clinical significance of lipid profiles in diagnosis lies in assessing the severity and acuity of hypothyroidism; therefore, the biochemical correlation between lipid profiles and sex was studied in patients with hypothyroidism, as shown in Table 4.

**Table (4): Concentration values of lipid profiles (TG, TC, HDL, LDL and VLDL) in blood sera of hypothyroidism patients as well as healthy individuals, depending on sex variable.**

| Sex | Groups | TC(mg/dl) | TG(mg/dl) | HDL(mg/dl) | LDL(mg/dl) | VLDL(mg/dl) |
|---|---|---|---|---|---|---|
| Male | Control(N=48) | 108.26±57.056 | 163.45±39.915 | 43.67±22.306 | 114.72±46.56 | 24.04±19.199 |
| | Patient(N=53) | 200.11±92.985 | 225.93±40.977 | 22.17±12.524 | 140.81±51.082 | 45.47±18.60 |
| P-value | | 0.0001*** | 0.0001*** | 0.001** | 0.011** | 0.05* |
| Female | Control(N=50) | 110.26±57.056 | 160.45±39.915 | 43.67±22.306 | 110.72±46.56 | 19.919.24.04± |
| | Patient(N=42) | 206.11±92.985 | 229.93±41.977 | 20.17±12.524 | 150.81±51.082 | 47.47±18.60 |
| P-value | | 0.0001*** | 0.0001*** | 0.001** | 0.011** | 0.035 |

**Mean ± SD ***p<0.0001, **p<0.00, *p<0.05, N.S: Non-significant were used to represent concentration levels.**

Table (4) showed all classes of lipids (TC, TG, HDL, LDL, and VLDL) various Concentrations, represented by (200.11±92.985, 225.93±40.977, 22.17±12.524, 140.81±51.082 and 45.47±18.60), respectively, in male patients with hypothyroidism. Also, the same lipid profile showed different concentrations:( 206.11±92.985,229.93±41.977,20.17±12.524,150.81±51.082,47.47±18.60) respectively, in female patients with hypothyroidism.

**Table (5) correlation coefficient (r) among age, GPX, LDL, TC, and VLDL. HDL**

| Variables | (r ) | p (value) |
|---|---|---|
| **(Sex-age)** | **0.8** | **0.001****** |
| **(TC- HDL)** | **-0.357** | **0.013*** |
| **(TC- VLDL)** | **0.908** | **0.001****** |
| **(GPX-TC)** | **0.318** | **0.028*** |
| **(GPX- LDL)** | **0.379** | **0.008****** |
| **(TG - HDL)** | **-0.441** | **0.002 ****** |
| **(LDL-HDL)** | **-0.500** | **0.001****** |
| **(Age-GPX)** | **0.465** | **0.001****** |
| **(Age-TC)** | **0.60** | **0.001****** |
| **(Age-LDL)** | **0.60** | **0.001****** |
| **(Age- VLDL)** | **0.362** | **0.01****** |
| **** The correlation is significant at the 0.01 level (2-tailed).<br>*The correlation is significant at the 2-tailed 0.05 level.** | | |

Figures (1-4) and Table 5 On the other hand, TC exhibited a somewhat negative association with HDL (r=-0.3, p=0.013*), but a highly significant positive correlation with sex and age (r=0.8, p=0.001). Additionally, TC exhibited a moderately positive association with GPX (r = 0.318, p = 0.028) and a very high positive correlation with VLDL (r = 0.908, p = 0.001). Additionally, there was a somewhat favorable connection (r = 0.379, p = 0.008*) between GPX and LDL. Additionally, the data revealed a high negative association between LDL and HDL (r = -0.500, p = 0.001**) and a moderately negative correlation between TG & HDL (r = -0.441, p = 0.002**). Age also shown a strong positive association with TC (r = 0.60, p = 0.001**), a moderate positive correlation with VLDL (r = 0.362, p = 0.01**), a strong positive correlation with LDL (r = 0.60, p = 0.001), & a moderate positive correlation with GPX (r = 0.4, p = 0.001**).

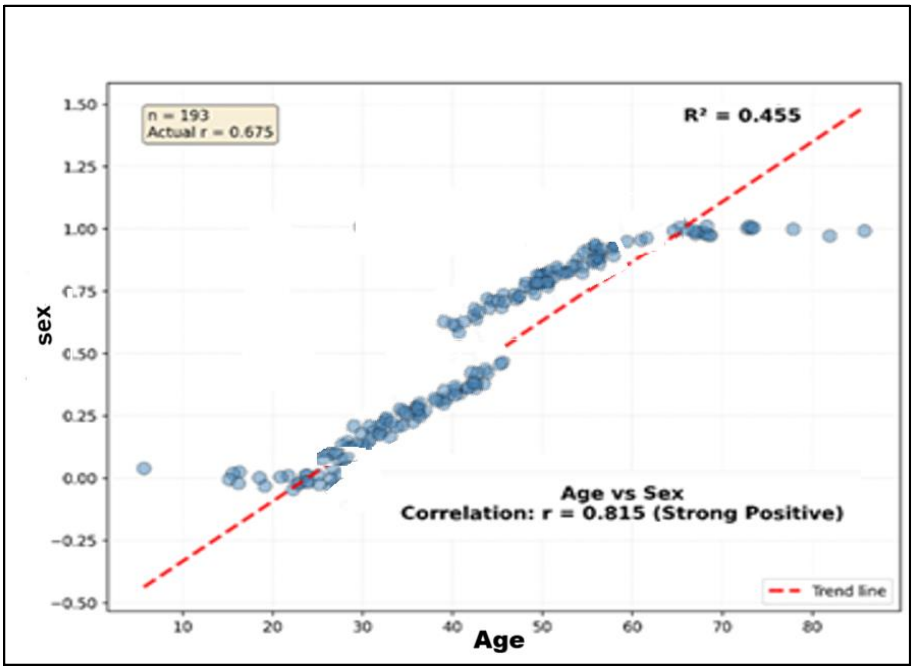


**Fig.(1) Correlation between sex & age**

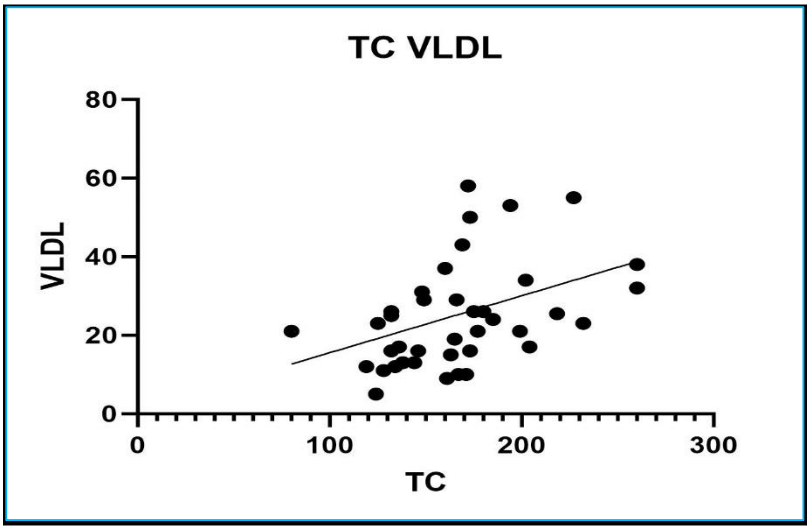


**Fig.(2) Correlation between VLDL & TC**

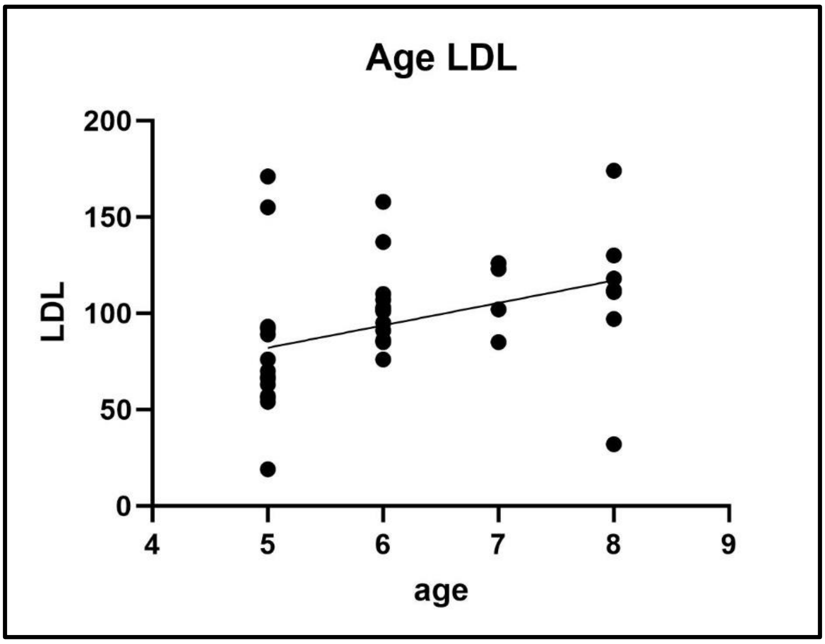


**Fig.(3) Correlation between Age & LDL**

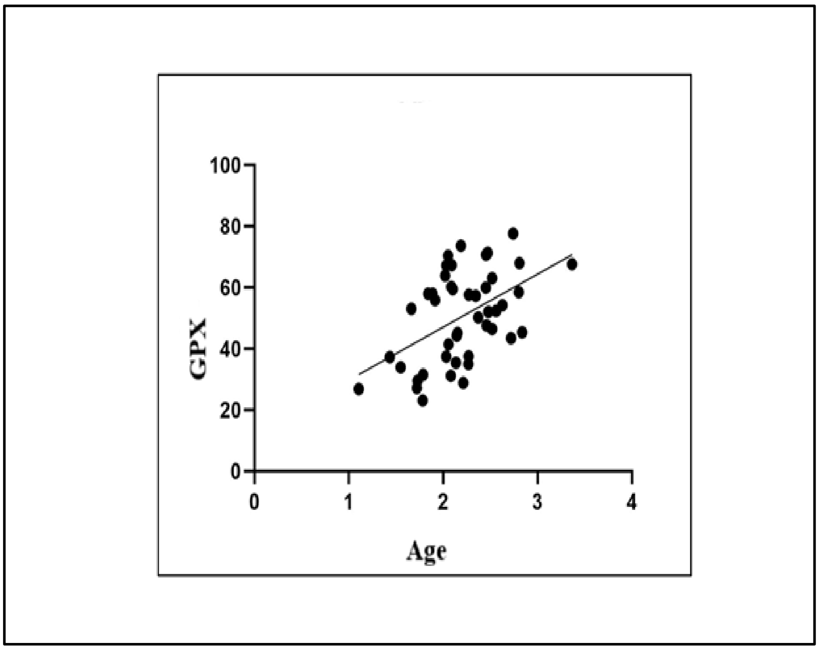


**Fig .(4) Correlation between Age & GPX**

**Discussion**

Triglycerides, total cholesterol, HDL cholesterol, LDL cholesterol, and very low-density lipoprotein cholesterol were measured using age and sex-dependent scales. Different concentrations of the antioxidant enzymes were recorded, with the highest catalase concentration in the first and last age groups and the lowest glutathione peroxidase concentration in the first age group. Therefore, age is a key factor in studying and monitoring the severity of hypothyroidism (21). Catalase (CAT) levels were considerably higher (p <0.0001) than controls, but GPX and SOD levels were lower than those of healthy persons, with the declines being more noticeable in the first as well as third age groups (22, 23). A marked increase in antioxidant enzyme (CAT) activity was also observed in hypothyroid patients of both sexes. The levels of these enzymes were significantly lower ($p<0.0001$) than in the control groups. The current study's findings are consistent with data (24). The decrease in SOD and GPX concentrations can be explained by a reduced ability of the body to detoxify or eliminate free radicals. CAT, GPX, and SOD levels are biochemically related to reactive oxygen species; for example, hydrogen peroxide is crucial in the synthesis of thyroid hormones (25). Decreased glutathione peroxidase (GPX) levels in the liver, muscles, heart, and lymphatic organs, along with decreased superoxide dismutase (SOD) levels, have been studied in hypothyroid patients across various age groups and both sexes, consistent with previous research (26-27). Lipid profile is a key biomarker for monitoring hypothyroidism activity. Several studies have confirmed a biological correlation between lipid profile and this disease (28, 29). The highest triglyceride concentration was observed in the first age group, while the lowest high-density lipoprotein (HDL) concentration was observed in hypothyroid patients across all age groups. When comparing them with controls, all the lipid profiles in a hypothyroid individual were significantly higher ($p<0.0001$) but at it highest in second age group. Conversely, almost all the lipid profiles of the third age group revealed a statistically significant decrease ($p<0.0001$). Lipid problems are more prevalent in overt hypothyroidism, according to one research. They therefore have a higher chance of developing cardiovascular disease (30). Higher levels of total cholesterol, LDL (bad cholesterol), & VLDL (very-bad cholesterol) are caused by reduced cholesterol clearance, which results from poor conversion of cholesterol to bile acids in the liver. As a result, people with hypothyroidism may also have increased triglyceride levels, particularly those linked to very low-density lipoprotein (VLDL) cholesterol (31). Although females had greater triglyceride levels, both male and female hypothyroid patients showed a substantial increase ($p<0.0001$) in height values, indicating the clinical impact of sex in hypothyroidism. This could be possibly explained by females having higher TSH (thyroid-stimulating hormone) than males (32). We also found substantial positive correlations between right atrial remodeling and age and sex, total cholesterol, LDL, as well as VLDL levels, as well as an inverse correlation between HDL cholesterol and triglycerides, which is consistent with a prior research (24).

## Conclusions

The present study provided confirmation and exposition of the biochemical and clinical correlation between age and sex factors pertaining to hypothyroidism patients, antioxidant enzymes (CAT, GPX, and SOD), and lipid profile parameters (TG, TC, HDL, LDL, and VLDL) levels. Moreover Biochemical follow up of hypothyroidism disease severity based on the concentrations values of antioxidant enzymes and lipid profile according to age and sex characteristics. Also, the enzymes (CAT, GPX, and SOD) are biochemically associated with lipid profile levels, and age is positively correlated with sex.

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