Ibn Al-Haitham Journal for Pure and Applied Sciences
Journal homepage: jih.uobaghdad.edu.iq
PISSN: 1609-4042, EISSN: 2521-3407
IHJPAS. 2024, 37(4)

Hepcidin and CRP Levels in a Sample of Iraqi Hypothyroid Patients Treated with Levothyroxine

Haider D. Abdullah[1,*] and Suha Abdul-Khaliq Al-Jowari[2]
[1,2]Department of Biology, College of Science, University of Baghdad, Baghdad, Iraq.
*Corresponding Author.

Received: 10 May 2023 Accepted: 4 July 2023 Published: 20 October 2024
doi.org/10.30526/37.4.3480

Abstract

Study of hypothyroidism and its relationship to the levels of the hormone hepcidin and C-reactive protein in people treated with levothyroxine. This study was carried out between November 2022 and March 2023. The study had a total of 90 male and female participants with an average age of 25 to 50 years old. All samples were obtained from both hospitals in Baghdad and the Medical Lab. The diagnosis of hypothyroidism based on symptoms and the hypothyroidism control test score led to the collection of 30 samples from healthy individuals, 30 from those with untreated hypothyroidism, and 30 additional samples. From people with hypothyroidism who were treated with levothyroxine, where the necessary tests were conducted for the three groups, Blood samples were collected in a gel tube, and serum was separated for use in their respective measurements of (TSH, T3, T4, CRP, and hepcidin). According to the results, there were no significant differences in the mean patient's age ($p=0.419$) or gender distribution ($p=0.638$) between the three studied groups. When thyroid function tests were done on the groups that were studied, the hypothyroid patients had significantly lower levels of triiodothyronine (T3) and thyroxine (T4) and higher levels of thyroid-stimulating hormone (TSH) before treating them ($p\leq0.001$). And the highest serum hepcidin and CRP levels (11.77 ± 0.98 and 14.61 ± 2.49 respectively) were found in group 1 patients. On the contrary, the lowest levels (7.61 ± 1.56 and 3.19 ± 0.29, respectively) were present in group 2 patients. In healthy subjects, the hepcidin and CRP levels were 8.45 ± 0.53 and 3.51 ± 0.91, respectively. Accordingly, there was a significant difference in hepcidin and CRP levels between the three studied groups, and the results showed a decrease in hepcidin and C-reactive protein after treatment.

Keywords: Hypothyroidism, hepcidin, levothyroxine, C-Reactive protein, thyroid-stimulating hormone, triiodothyronine, thyroxine.

1. Introduction

Hypothyroidism is a common condition where the thyroid doesn't create nor release enough thyroid hormone into the bloodstream. Hypothyroidism is defined as an elevated thyroid-stimulating hormone (TSH) concentration in combination with a free thyroxine (fT4)

concentration below the reference range. This makes the metabolism slow down. Also called underactive thyroid, hypothyroidism can make us feel tired, gain weight, and be unable to tolerate cold temperatures. The main treatment for hypothyroidism is hormone replacement therapy. Levothyroxine (LT4) is the mainstay of treatment and is one of the most commonly prescribed drugs worldwide [1]. Hypothyroidism has several causes. They include Hashimoto's disease, an autoimmune disorder where the immune system attacks your thyroid. This is the most common cause. Thyroiditis, inflammation of the thyroid, Congenital hypothyroidism refers to hypothyroidism that is present from birth. A surgical procedure may involve the removal of part or all of the thyroid. Radiation therapy for the thyroid with certain medicines may cause, in rare cases, a pituitary disease, or too much or too little iodine in your diet [2].

Hypothyroidism can be caused by autoimmune Hashimoto's disease and subclinical hypothyroidism. When the thyroid gland experiences inflammation, it triggers a response in interleukins. An increase in IL6 leads to a rise in hepcidin concentration [3], which in turn raises the hormone hepcidin in the body. This, in turn, regulates the body's iron levels. Additionally, a rise in interleukin 6 also causes a rise in CRP, which is considered an indicator of increased inflammation in the body [4].

Studies also show that there is a correlation between high levels of CRP and people with subclinical hypothyroidism, as this protein is elevated and stimulated by interleukin 6, which is considered an indicator of inflammation in the body. The hormone hepcidin, representing an inflammatory response, controls hypothyroidism and iron levels, leading to elevated IL-6 and CRP [5].

Patients with hypothyroidism from Hashimoto's disease demonstrated the use of levothyroxine as a treatment for hypothyroidism, as it serves as a substitute for thyroid hormone, which is deficient. The treatment showed a decrease in inflammatory indicators in the body, as levothyroxine treatment is considered an anti-inflammatory in the thyroid gland and thus reduces signs of inflammation [6].

2. Materials and methods

2.1. Study Design, Timing, and Ethical Consideration

This study was conducted during the period from November 2022 to March 2023. For each patient participating in the study, written informed consent was obtained. The ethical committees of the Department of Biology, College of Science, University of Baghdad approved the study. The study had a total of 90 male and female participants with an average age of 25 to 50 years old. All samples were obtained from both hospitals in Baghdad and the Medical Lab. They were diagnosed with hypothyroidism according to symptoms and a hypothyroidism control test score. The patient questionnaire in Appendix 1 directly provided the clinical information. The consultant supervised the process and secured approval for both patient and control sampling.

2.2. Study groups

The study groups are as follows: Group 1: 30 patients (9 males and 21 females) with hypothyroidism with no treatment. Group 2: 30 patients (9 male and 21 female) diagnosed with hypothyroidism and treated with levothyroxine for one year or less. Group 3 includes 30 healthy Euthyroid individuals of both sexes (12 males and 13 females). The study excluded patients who met the following criteria: patients with diabetic mellitus, hypertension, thalassemia, kidney and liver disease, and iron supplements.

2.3. Blood sampling

We aspirated 5–6 ml of peripheral whole blood from each control and patient group using a plastic disposable syringe. We immediately dispensed this blood: we transferred 6 ml of blood into a gel tube, allowed it to clot for 10 minutes at 37 °C in water bath, and centrifuged it at 3000 rpm for 10 minutes. After obtaining the clear serum, we stored it at -20 C until we used it for hormonal and immunological parameter assays. We discarded the hemolyzed samples (Bernadette and Dianne, Bernadette Mazurek Melnyk and Dianne Morrison-Beedy, 2012).

2.4. Methods

The hormones triiodothyronine (T3), thyroxine (T4), and thyroid stimulating hormone (TSH) were tested using immunoassays to measure the amount of TFT in human serum and plasma. Elecsys and Cobas e immunoassay analyzers are the intended platforms for the electrochemiluminescence immunoassay "ECLIA".

We made the assay using an automated instrument (Cobase e411) and a TFT kit from Cobase, Germany. Therefore, this instrument automatically controls the assay temperature and steps. Normal ranges: for T3 0.8-2.0 (ng/ml), normal ranges for TSH 0.270-4.2 (µIU/ml), normal ranges for T4 5.1-14.1 (µg/dl). Determination of CRP An in vitro test quantitatively determines the CRP concentration in serum using photometric systems and the BS-430 Chemistry Analyzer Mindray automated kit from Mindary, China. Normal ranges for CRP are 0–5 (pg/mL). Determination of Hepcidin by using the ELISA technique, according to the manufacturer's instructions for the Abbexa kit, United Kingdom. This ELISA kit uses the Sandwich-ELISA principle. Normal ranges for Hepcidin are 6.5–11.3 pg/mL.

3. Results and Discussion

3.1. Classification of the studied groups

The present study enrolled 90 people, selected the subjects, and divided them into three groups. The first group (Group 1) included 30 patients with a history of newly diagnosed hypothyroidism; the second group (Group 2) involved 30 hypothyroid patients on thyroxin treatment. These two groups of patients were compared with 30 healthy controls (Group 3) (**Figure 1**).

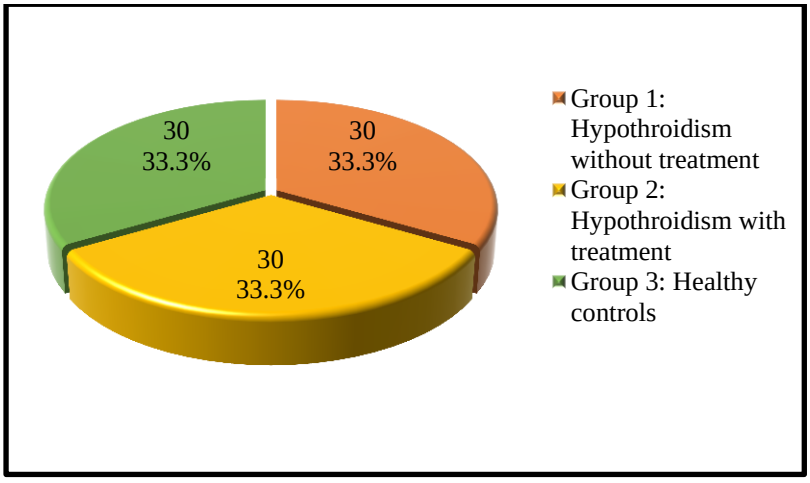

Figure 1. Classification of the studied groups.

3.2. Comparison of age and gender between the studied groups

The mean patients' age was expressed as mean ± standard deviation, and gender distributions were expressed as frequency and percentage. According to the results, there were no significant

differences in the mean patient's age (p=0.419) or gender distribution (p=0.638) between the three studied groups, as demonstrated in **Table 1** and **Figure 2**.

Table 1. Demographic features of the studied groups.

Parameter (Mean ± SD)	Hypothyroidism without treatment (Group 1)	Hypothyroidism with treatment (Group 2)	Healthy controls (Group 3)	*p* value
Age (years)	39.40 ± 8.50	38.30 ± 8.07	36.67 ± 7.49	0.419 *F* NS
Gender Male Female	9 (30 %) 21 (70 %)	9 (30 %) 21 (70 %)	12 (40 %) 18 (60 %)	0.638 X^2 NS

SD: Standard deviation; NS: Not significant (p > 0.05); *F* : ANOVA test; X^2: Chi square

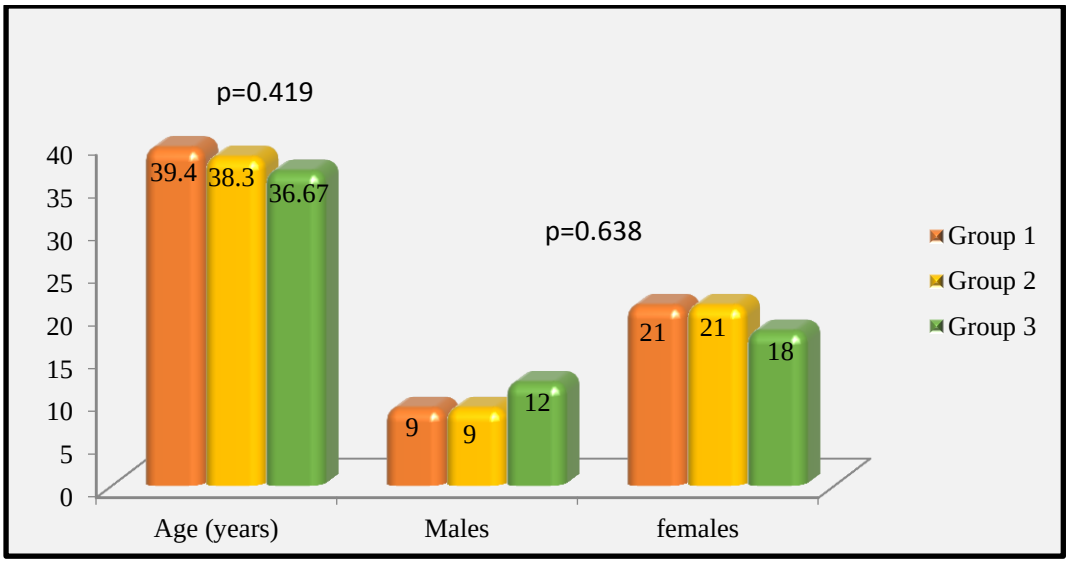

Figure 2. Comparison of demographic features between the studied groups.

There were no clear differences in the age groups, but there were differences in gender. According to this study [7], the rate of hypothyroidism is frequently higher in females than in males.

According to a study conducted on a group of people in Europe, they were diagnosed with hypothyroidism. According to the sensitivity analysis, the prevalence of hypothyroidism tends to be higher in female patients [8]. Hypothyroidism, a disorder in the thyroid gland that does not make enough thyroid hormone, is the second most common endocrine disorder in women [9].

3.3. Comparison of thyroid function tests between the studied groups

The comparison of thyroid function tests between the studied groups is illustrated in **Table 2** and **Figure 3**. According to the results, there were significantly lower T3 and T4 levels and higher TSH levels in hypothyroid patients before the treatment ($p < 0.001$).

Table 2. Comparison of thyroid function tests between the studied groups.

Thyroid function tests	Hypothyroidism without treatment (Group 1)	Hypothyroidism with treatment (Group 2)	Healthy controls (Group 3)	*p* value
T3 (ng/ml)	0.47 ± 0.28	1.35 ± 0.35	1.42 ± 0.34	< 0.001*F* S
T4 (µg/dl)	3.16 ± 1.64	8.06 ± 1.63	8.71 ± 2.07	< 0.001*F* S
TSH(µIU/ml)	19.72 ± 3.52	1.87 ± 0.93	1.85 ± 0.88	< 0.001*F* S

T3: Triiodothyronine; T4: Thyroxin; TSH: Thyroid stimulating Hormone; S: Significant (p≤0.05); *F*: ANOVA

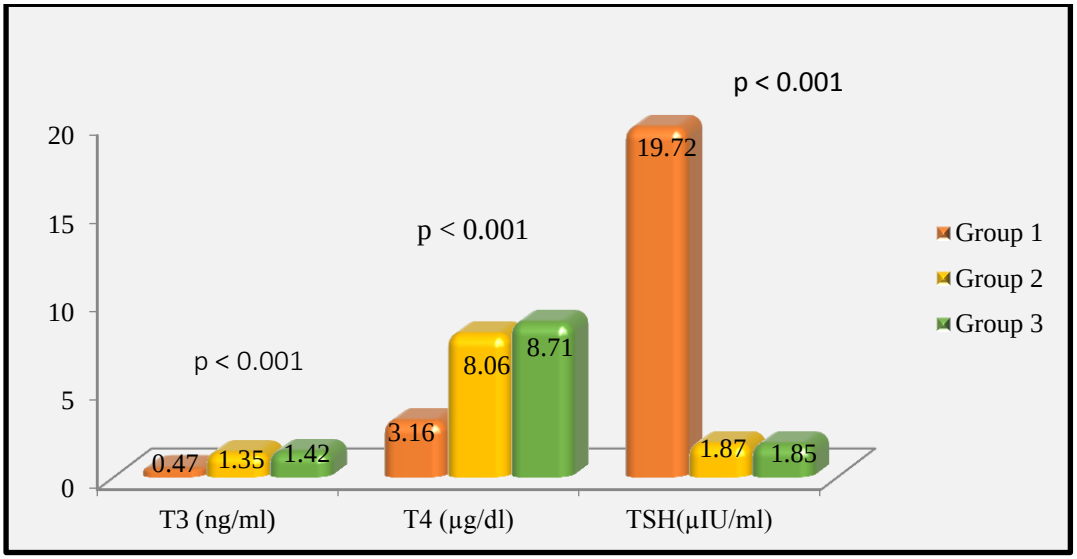

Figure 3. Comparison of demographic features between the studied groups.

A post hoc test of ANOVA showed significant differences between group 1 patients and group 2 and group 3 patients ($p< 0.001$) regarding all thyroid function tests; on the contrary, there were no significant differences in T3, T4, and TSH levels between group 2 and group 3 patients, with p values equal to 0.341, 0.729, and 0.1, as presented in **Table 3**.

Table 3. Post hoc test of ANOVA of thyroid function tests for multiple comparisons between groups.

	Post hoc test for multiple groups comparisons		*p* value
T3	Group 1	Group 2	< 0.001
	Group 1	Group 3	< 0.001
	Group 2	Group 3	0.341
T4	Group 1	Group 2	< 0.001
	Group 1	Group 3	< 0.001
	Group 2	Group 3	0.729
TSH	Group 1	Group 2	< 0.001
	Group 1	Group 3	< 0.001
	Group 2	Group 3	1.00

T3: Triiodothyronine; T4: Thyroxin; TSH: Thyroid stimulating hormone.

3.4. Comparison of serum hepcidin and CRP levels between the studied groups

The highest serum hepcidin and CRP levels (11.77 ± 0.98 and 14.61 ± 2.49, respectively) were found in group 1 patients; on the contrary, the lowest levels (7.61 ± 1.56 and 3.19 ± 0.29, respectively) were present in group 2 patients. The hepcidin and CRP levels in healthy subjects

were 8.45 ± 0.53 and 3.51 ± 0.91, respectively. Accordingly, there was a significant difference in hepcidin levels between the three studied groups, as presented in **Table 4** and **Figure 4.**

Table 4. Comparison of serum hepcidin level between the studied groups.

Parameters	Hypothyroidism without treatment (Group 1)	Hypothyroidism with treatment (Group 2)	Controls Group 3	*p* value
Serum hepcidin	11.77 ± 0.98	7.61 ± 1.56	8.45 ± 0.53	< 0.001*F* S
Serum CRP	14.61 ± 2.49	3.19 ± 0.29	3.51 ± 0.91	< 0.001*F* S

S: Significant (p ≤ 0.05); *F*: ANOVA; CRP: C-reactive protein.

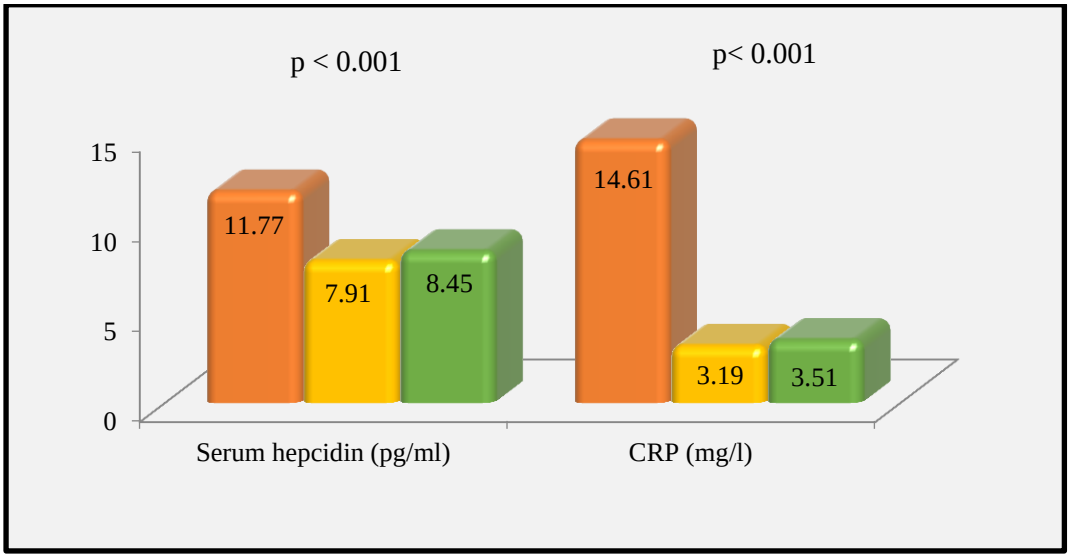

Figure 4. Comparison of serum hepcidin & CRP levels between the studied groups.

Post hoc tests of hepcidin and CRP (**Table 5**) demonstrated a significant difference ($p < 0.05$) in all parameters between groups 1 and 2 and between groups 1 and 3. There were also significant differences regarding hepcidin (p=0.573) and CRP (p=0.988).

Table 5. Post hoc test of ANOVA of iron status, hepcidin and CRP levels for multiple comparisons between groups.

Post hoc test for multiple groups comparisons			*p* value
Hepcidin	Group 1	Group 2	< 0.001
	Group 1	Group 3	< 0.001
	Group 2	Group 3	0.573
CRP	Group 1	Group 2	< 0.001
	Group 1	Group 3	< 0.001
	Group 2	Group 3	0.988

Hypothyroidism is one of the thyroid disorders that causes elevated levels of highly sensitive C-reactive protein. The results of group 1 showed an increase in CRP because they had hypothyroidism, but compared with groups 2, 3, and 4, there was no significant increase in CRP values in healthy subjects or treated patients [15]. One of the reasons for the increase in CRP in the body is the presence of inflammation, and it is also mentioned that hypothyroidism due to autoimmune disease can lead to an increase in the rate of inflammation in the body and thus lead

to an increase in CRP. Primary autoimmune hypothyroidism is an inflammatory disease characterized by elevated cytokine levels that decrease with LT4 therapy [16].

Additionally, studies suggest that individuals with thyroiditis or Hashimato disease experience a significant increase in CRP levels, a protein that increases when the body experiences inflammation [17]. The rise CRP is response to the rise of IL6, and thus leads to a rise in CRP, where the response of IL6 is to the presence of inflammation in the body, where it rise leads to a positive response in CRP six hours after the rise of IL6 [18]. As for the hepcidin hormone, it is a response to the rise IL-6, where the rise the cytokine IL-6 leads to a positive response in the hepcidin hormone, therefore the rise of this hormone leads to a reduction in the proportion of iron in the body, and since it responds to IL-6, this means that there is a positive response from this hormone to inflammation in the body [19].

3.5. Correlations between thyroids function tests and serum hepcidin and CRP levels

3.5.1. Correlations between thyroid function tests with serum hepcidin and CRP levels in untreated hypothyroid patients

The correlations between thyroid function tests and iron hepcidin levels in untreated hypothyroid patients are presented in **Table 6**. According to the results, there were:

Significant positive correlation between T3 and hepcidin levels (r=0.382 & *p*=0.037) (**Figure 5**).

Table 6. Correlation between thyroid function tests with serum iron status, hepcidin and CRP levels in untreated hypothyroid patients

Parameters		T3	T4	TSH
Hepcidin	r	0.382*	0.077	-0.234
	p	0.037	0.686	0.213
CRP (mg/l)	r	0.134	-0.172	-0.224
	p	0.480	0.362	0.235

Pearson's correlation coefficient; T3: Triiodothyronine ; T4: Thyroxin ;TSH: Thyroid stimulating hormone; CRP: C-reactive protein.

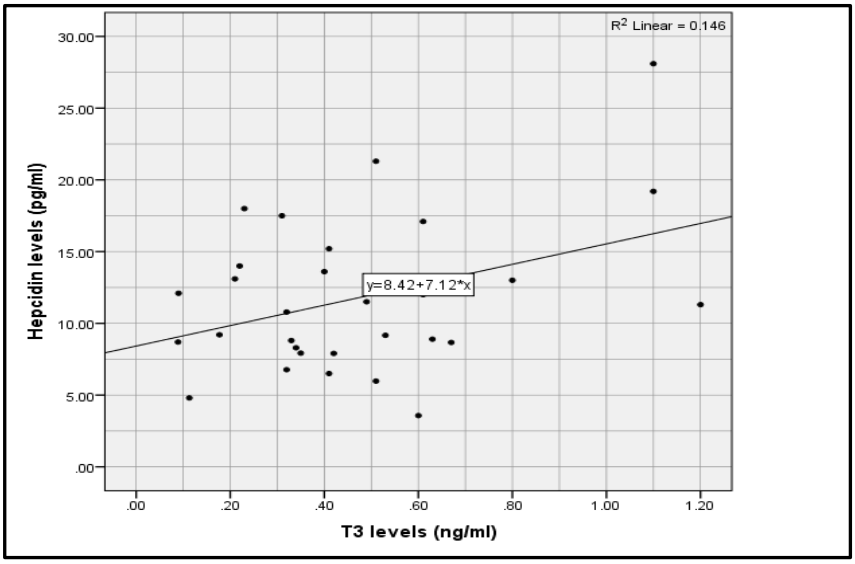

Figure 5. Correlation between T3 and hepcidin levels in untreated hypothyroid patients.

3.5.2.Correlations between thyroid function tests with hepcidin and CRP levels in hypothyroid patients treated with thyroxin

There were no significant correlations between thyroid tests and hepcidin levels, as demonstrated in **Table 7**.

Table 7. Correlation between thyroid function tests with serum hepcidin and CRP levels in hypothyroid patients treated with thyroxin**.**

Parameters		T3	T4	TSH
Hepcidin	r	-0.141	0.083	0.080
	p	0.457	0.662	0.674
CRP (mg/l)	r	0.036	0.189	0.323
	p	0.851	0.316	0.082

Pearson's correlation coefficient; T3: Triiodothyroxin; T4: Thyroxin ;TSH: Thyroid stimulating hormone; CRP: C-Reactive protein

3.5.3. Correlations between thyroids function tests with serum hepcidin and CRP levels in healthy control group

There were also no significant correlations between T3, T4, TSH, hepcidin, and CRP levels in healthy subjects (group 3), as demonstrated in **Table 8.**

Table 8. Correlation between thyroid function tests with serum hepcidin levels in healthy control group.

Parameters		T3	T4	TSH
Hepcidin	r	-0.243	-0.043	0.112
	p	0.196	0.821	0.554
CRP (mg/l)	r	0.053	0.250	0.055
	p	0.779	0.183	0.775

Pearson's correlation coefficient; T3: Triiodothyroxin; T4: Thyroxin ;TSH: Thyroid stimulating hormone; CRP: C-Reactive protein

3.5.4. Correlations between hepcidin with CRP levels in untreated hypothyroid patients (group 1)

There was positive significant correlation between serum hepcidin with serum CRP (r=0.683 & p < 0.001); however serum hepcidin, as illustrated in **Table 9** and **Figure 6.**

Table 9. Correlations between hepcidin and CRP levels in untreated hypothyroid patients.

Correlation between serum hepcidin with	Pearson's correlation coefficient	*p* value
CRP	0.683*	< 0.001

CRP: C-reactive protein

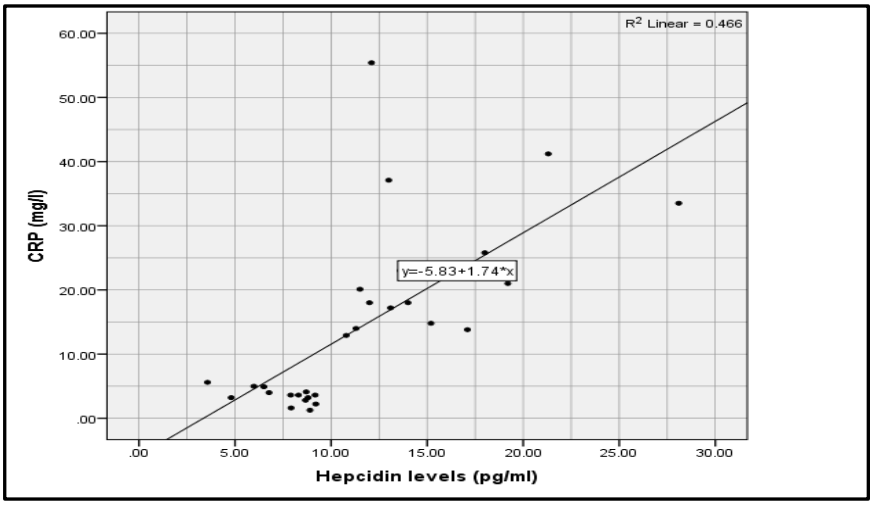

Figure 6. Correlations between hepcidin and CRP levels in untreated hypothyroid patients.

The aim of this study was to measure hepcidin in a prospective study of patients with newly diagnosed hypothyroidism in the context of thyroiditis and after successful treatment. The study consisted of conducting clinical evaluations and tests. The study was found that there is a positive correlation between hypothyroidism and hepcidin, where in group 1 patients who had a decrease in T4 and an increase in TSH, they had a rise in hepcidin rates for group 2 who were treated and group 3 healthy people, and it was observed.

Also, a decrease in the concentration of hepcidin during the transition from hypothyroidism to a healthy thyroid condition in patients is associated with the observed dynamics in the iron balance. This means that there is a positive correlation between T3 and T4, as well as between hepcidin, in subjects with inflammatory hypothyroidism [20]. Studies have shown that there is a link between the hormone hepcidin and interleukin-6. This is because interleukin-6 levels rise when the body's inflammation level rises, so the rise in levels of this cytokine can be linked to the hormone hepcidin. In addition, CRP rises with an increase in interleukin-6, which means that there is a possible relationship between CRP and Hepsidin hormone, and this is what appears in the results of the study in people with inflammatory hypothyroidism [21, 22].

Also Hepcidin, a hormone that regulates iron, is a promising diagnostic for distinguishing between iron shortage and inflammatory anemia. When there is inflammation, both the amounts of smaller, non-biologically active hepcidin isoforms and the concentration of hepcidin in the plasma increase. When the concentration of hepcidin increases as a result of inflammation, the increased concentration leads to a reduction in the level of iron in the body. This implies a correlation between the elevated CRP due to autoimmunity and thyroiditis and the hormone hepsidin, which regulates the body's iron levels [23, 24]. Also Elevated levels of the inflammatory markers CRP and IL-6 are associated with subclinical hypothyroidism. Therefore, individuals directly linked to the hepcidin hormone exhibit a positive correlation between elevated inflammatory markers and elevated hepcidin levels in subclinical hypothyroidism [25, 26].

In another study, a correlation was shown between patients with Hashimoto's thyroiditis and elevated IL6 and CRP, as we mentioned earlier, and the results of this research confirm a positive relationship between hypothyroidism and elevated CRP and IL6, which are also positively associated with the hormone that controls the level of iron. It's called hepcidin [27, 28]. Another study confirmed that people with hypothyroidism due to Hashimoto's disease had an increase in the inflammatory indicators CRP and IL6, and after treatment with levothyroxine, the rate of inflammation in the thyroid gland and the inflammatory indicators CRP and IL6 significantly decreased [29, 30].

4. Conclusions

The current study yielded the following conclusions: Women had a higher risk of hyperthyroidism than men. T3 and T4 decreased, whereas TSH increased in hypothyroid patients with clinical hypothyroidism and thyroidite auto-immune Hashimoto. T3 and T4 increased while TSH decreased in hypothyroid patients treated with the levothyroxine drug. Additionally, hypothyroid patients experienced an increase in CRP levels, which subsequently decreased after a year or less of levothyroxine treatment. Hypothyroid patients experienced an increase in hepcidin levels, which continued to decrease after treatment with the levothyroxine drug.

Studies have also shown that levothyroxine has a positive effect on hepcidin in treated patients. The results indicated an increase in hepcidin and C-reactive protein levels in patients with untreated hypothyroidism, which is caused by autoimmunity, such as Hashimoto disease, or

subclinical hypothyroidism. These conditions are the most common causes of hypothyroidism, and they are associated with high levels of inflammation, which are linked to high CRP and hepcidin. However, the results also showed a decrease in hepcidin and protein levels after treatment. After treatment, the drug levothyroxine reduced the rate of inflammation resulting from hypothyroidism.

Acknowledgment

Many thanks to editor-in-chief and members of the editorial committee in Ibn Al-Haitham Journal for Pure and Applied Sciences.

Conflict of Interest

There is no conflict of interest.

Funding

There is no funding for the article.

References

1. Chaker, L.; Razvi, S.; Bensenor, I.M.; Azizi, F;, Pearce, E.N.; Peeters, R.P. Hypothyroidism *Nature Reviews Disease Primers* **2022,** *8(1),* 30. https://doi.org/10.1038/s41572-022-00357-7.
2. Hughes, K.; Eastman, C. Thyroid disease: Long-term management of hyperthyroidism and hypothyroidism. *Australian Journal of General Practice* **2021,** *50(1-2),* 36–42. https://doi.org/10.31128/ajgp-09-20-5653.
3. Lei, Y.; Yang, J.; Li, H.; Zhong, H.; Wan, Q. Changes in glucose-lipid metabolism, insulin resistance, and inflammatory factors in patients with autoimmune thyroid disease. *Journal of Clinical Laboratory Analysis* **2019,** *33(7),* e22929. https://doi.org/10.1002/jcla.22929.
4. Kwiatek-Majkusiak, J.; Geremek, M.; Koziorowski, D.; Tomasiuk, R.; Szlufik, S.; Friedman, A. Serum levels of hepcidin and interleukin 6 in Parkinson's disease. *Acta Neurobiologiae Experimentalis* **2020,** *80(3),* 297-304.
5. Yu, Y.T.; Ho, C.T.; Hsu, H.S.; Li, C.I.; Davidson, L.E.; Liu, C.S.; Lin, W.Y. Subclinical hypothyroidism is associated with elevated high-sensitive C-reactive protein among adult Taiwanese. *Endocrine* **2013,** *44,* 716-722. https://doi.org/10.1007/s12020-013-9915-0.
6. Krysiak, R.; Okopien, B. The effect of levothyroxine and selenomethionine on lymphocyte and monocyte cytokine release in women with Hashimoto's thyroiditis. *The Journal of Clinical Endocrinology and Metabolism* **2011,** *96(7),* 2206-2215. https://doi.org/10.1210/jc.2010-2986.
7. Ahmed, M.J.; Mohammed, A.A.; Qasim, B.A. Lipid profile IN subclinical hypothyroidism: a two centers experience. *Duhok Medical Journal* **2019,** *13(1),* 56-65. https://doi.org/10.31386/dmj.2019.13.1.6.
8. Mendes, D.; Alves, C.; Silverio, N.; Marques, F.B. Prevalence of undiagnosed hypothyroidism in Europe: a systematic review and meta-analysis. *European Thyroid Journal* **2019,** *8(3),* 130-143. https://doi.org/10.1159/000499751.
9. Dunn, D.; Turner, C. Hypothyroidism in women. *Nursing for Women's Health* **2016,** *20(1),* 93-98. DOI: https://doi.org/10.1016/j.nwh.2015.12.002.
10. Livingston, E.H. Subclinical hypothyroidism. *Jama* **2019,** *322(2),* 180-180. https://doi.org/10.1001/jama.2019.9508.
11. Rosinha, P.; Dantas, R.; Alves, M.; Azevedo, T.; Inácio, I.; Esteves-Ferreira, S.; Guimarães, J. Subclinical Hypothyroidism in Pediatric Age: How Important Is Autoimmunity? *Cureus* **2022,** *14(8),* e28507. https://doi.org/10.7759%2Fcureus.28507.

12. Almandoz, J.P.; Gharib, H. Hypothyroidism: etiology, diagnosis, and management. *Medical Clinics* **2012,** *96(2),* 203-221. https://doi.org/10.1016/j.mcna.2012.01.005.
13. Hueston, W.J. Treatment of hypothyroidism. *American Family Physician* **2001**, *64(10),* 1717.
14. Hennessey, J.V.; Espaillat, R. Current evidence for the treatment of hypothyroidism with levothyroxine/levotriiodothyronine combination therapy versus levothyroxine monotherapy. *International Journal of Clinical Practice* **2018,** *72(2),* e13062. https://doi.org/10.1111/ijcp.13062.
15. Kazemi Tanha, M.; Nayebifar, S.; Ghasemi, E.; Nosrat Zehi, S. Investigating the synergistic effect of Nasturtium officinale extract and High-intensity interval training on fatty acid-binding protein 4 (FABP4) and high-sensitivity C-reactive protein (hs-CRP) in overweight subclinical hypothyroid patients: a rand. *Sport Physiology* **2023,** *14(56),* 175-198. https://doi.org/10.22089/spj.2022.13463.2214.
16. Tayde, P.S.; Bhagwat, N.M.; Sharma, P.; Sharma, B.; Dalwadi, P.P.; Sonawane, A.; Varthakavi, P.K. Hypothyroidism and depression: are cytokines the link?. *Indian Journal of Endocrinology and Metabolism* **2017,** *21(6),* 886. https://doi.org/10.4103%2Fijem.IJEM_265_17.
17. Erge, E.; Kiziltunc, C.; Balci, S.B.; Atak Tel, B.M.; Bilgin, S.; Duman, T.T.; Aktas, G. A Novel Inflammatory Marker for the Diagnosis of Hashimoto's Thyroiditis: Platelet-Count-to-Lymphocyte-Count Ratio. *Diseases* **2023,** *11(1),* 15. https://doi.org/10.3390/diseases11010015.
18. Ji, J.; Sun, C.L.; Cohen, H.J.; Synold, T.; Muss, H.; Sedrak, M.S. Inflammation and clinical decline after adjuvant chemotherapy in older adults with breast cancer: Results from the Hurria older patients prospective study. *Journal of Clinical Oncology* **2023,** *41(2),* 307-315. https://doi.org/10.1200/jco.22.01217.
19. Nemeth, E.; Ganz, T. Hepcidin and iron in health and disease. *Annual Review of Medicine*, **2023,** *74*, 261-277. https://doi.org/10.1146%2Fannurev-med-043021-032816.
20. Hernik, A.; Szczepanek-Parulska, E.; Filipowicz, D.; Abdolall, A.; Borowczyk, M.; Wrotkowska, E.; Ruchała, M. The hepcidin concentration decreases in hypothyroid patients with Hashimoto's thyroiditis following restoration of euthyroidism. *Scientific Reports* **2019,** *9(1),* 16222. https://doi.org/10.1038/s41598-019-52715-3.
21. Wrighting, D.M.; Andrews, N.C. Interleukin-6 induces hepcidin expression through STAT3. *Blood* **2006,** *108(9),* 3204-3209. https://doi.org/10.1182/blood-2006-06-027631.
22. Banzet, S.; Sanchez, H.; Chapot, R.; Bigard, X.; Vaulont, S.; Koulmann, N. Interleukin-6 contributes to hepcidin mRNA increase in response to exercise. *Cytokine* **2012,** *58(2),* 158-161. https://doi.org/10.1016/j.cyto.2012.01.006.
23. Oppen, K.; Brede, C.; Skadberg, Ã.Y.; Steinsvik, T.; Holter, J.C.; Michelsen, A.E.; Heggelund, L. Hepcidin analysis in pneumonia-comparison of immunoassay and LC-MS/MS. *Annals of Clinical Biochemistry* **2023,** *60(5),* 298–305. https://doi.org/10.1177/00045632231159529.
24. Gupta, G.; Sharma, P.; Kumar, P.; Itagappa, M. Study on subclinical hypothyroidism and its association with various inflammatory markers. *Journal of Clinical and Diagnostic Research: JCDR* **2015,** *9(11),* BC04. https://doi.org/10.7860%2FJCDR%2F2015%2F14640.6806.
25. Taddei, S.; Caraccio, N.; Virdis, A.; Dardano, A.; Versari, D.; Ghiadoni, L.; Monzani, F. Low-grade systemic inflammation causes endothelial dysfunction in patients with Hashimoto's thyroiditis. *The Journal of Clinical Endocrinology and Metabolism* **2006,** *91(12),* 5076-5082. https://doi.org/10.1210/jc.2006-1075.
26. Marchiori, R.C.; Pereira, L.A.; Naujorks, A.A.; Rovaris, D.L.; Meinerz, D.F.; Duarte, M. M.; Rocha, J.B. Improvement of blood inflammatory marker levels in patients with hypothyroidism under levothyroxine treatment. *BMC Endocrine Disorders* **2015,** *15(1),* 1-9. https://doi.org/10.1186/s12902-015-0032-3.
27. Ross, E.; Ganz, T. Hepcidin and iron in health and disease. *Annals of Clinical Biochemistry*, **2023,** *54*, 261-277. https://doi.org/10.1146%2Fannurev-med-043021-032816.

28. Koorts, A.M.; Viljoen, M. Acute phase proteins: Ferritin and ferritin isoforms. In: *Acute phase proteins-regulation and functions of acute phase proteins*. Veas, F. Edn. IntechOpen, **2011**. https://doi.org/10.5772/20586.
29. Gehrer, C.M.; Mitterstiller, A.M.; Grubwieser, P.; Meyron-Holtz, E.G.; Weiss, G.; Nairz, M. Advances in Ferritin Physiology and Possible Implications in Bacterial Infection. *Indian journal of Endocrinology and Metabolism* **2017,** *31(4),* 146. https://doi.org/10.3390/ijms24054659.
30. Oppen, K.; Ueland, T.; Siljan, W.W.; Skadberg, Ø.; Brede, C.; Lauritzen, T.; Aukrust, P.; Steinsvik, T.; Husebye, E.; Michelsen, A.E.; Holter, J.C.; Heggelund, L. Hepcidin and Ferritin Predict Microbial Etiology in Community-Acquired Pneumonia. *Open Forum Infectious Diseases* **2021**, *8(4),* ofab082. https://doi.org/10.1093/ofid/ofab082.