

Diagnostic Role of Serum Ferritin in Patients with Hypothyroidism and Its Association with Thyroid Hormone LevelsAleema Banu S¹, Romy. W. Marshnil², Duraisingh V³¹Assistant Professor, Department of Physiology, Government Thoothukudi Medical College, Thoothukudi, Tamil Nadu, India²Associate Professor, Department of Biochemistry, Government Thoothukudi Medical College, Thoothukudi, Tamil Nadu, India³Assistant Professor, Department of Economics, Rani Anna Government College for Women, Tirunelveli, Tamil Nadu, India

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Abstract:

Introduction: Thyroid diseases are common worldwide and its prevalence in India is about 42 million. It is more common among women than in men, and also it increases with age. Among the thyroid diseases, Hypothyroidism is a multiorgan disorder that occurs due to decrease in thyroid hormone synthesis; which is associated with low circulating levels of thyroid hormones. Iron plays an important role in thyroid hormone synthesis along with other minerals. Ferritin, is an important indicator of body iron stores, and its level is altered in Hypothyroidism. Various studies reveal that most of the hypothyroid individuals do suffer from associated anaemia due to iron deficiency. Iron deficiency can be detected earlier using serum Ferritin, since it out-performs in its sensitivity & specificity than many other iron deficiency markers thereby preventing cognitive ailments.

Aim: This study was conducted to assess the diagnostic role of serum Ferritin in Hypothyroid individuals and to find out its association with Thyroid hormone levels.

Materials and Methods: This cross-sectional study included 100 subjects (50 subjects of Hypothyroids as cases and 50 subjects of Normothyroids as controls) with age group 20-50 years of both genders. Blood samples were collected under strict aseptic precautions, and serum Total T3, T4, TSH & serum Ferritin levels were assayed. Data was analysed statistically using suitable SPSS statistical package (version 21.0).

Results: There was a statistically significant decrease in serum Ferritin levels in hypothyroid individuals (40.6µg/l), when compared to Normothyroid individuals (82.9µg/l) with female preponderance. Also, there was a statistically significant positive correlation between serum Ferritin and Thyroid hormone levels (Total T4) in hypothyroid individuals. There was also a statistically significant association between serum Ferritin and Thyroid hormone levels (Total T3 and Total T4) with (p<0.05).

Conclusion: Serum Ferritin levels are significantly reduced in patients with Hypothyroidism. Thus, serum Ferritin can be used as an effective diagnostic marker in patients with Hypothyroidism, which can be used for diagnosis, monitoring and follow up of treatment in Hypothyroids, as well as for early detection of Iron deficiency anaemia, thereby preventing the development of dementia and cognitive decline in Hypothyroid individuals by early intervention.

Keywords: Serum Ferritin, TSH-Thyroid stimulating hormone, T3- triiodothyronine, T4 -thyroxine, Iron deficiency anaemia, Hypothyroidism.

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Introduction

In India, it is estimated that, about 42 million people suffer from thyroid disorders. Hypothyroidism is the commonest thyroid disorder, affecting one in ten adults in India and its prevalence is about 11% [1, 2]. Also, it is 10 times more common in women than in men, which increases with age [3].

It results mostly due to deficiency of thyroid hormones, and can also occur due to hypothalamic or pituitary disease and due to disorders that affect the

thyroid gland directly [4]. The key laboratory findings of early detection of Hypothyroidism are, decrease in Total T3 and Total T4 concentrations, which inturn leads to hypersecretion of pituitary TSH as well as an amplified increase in serum TSH levels [5].

Deficiency of iron leads to Iron deficiency anaemia, which is more common in Hypothyroidism [6]. Ferritin is an important indicator of body iron stores,

and its level is significantly altered in Hypothyroidism. This is because, thyroid hormones regulate Ferritin expression, and binding of thyroid hormones to Ferritin gene elements increases Ferritin synthesis [7].

The Thyroid Peroxidase (TPO) which is a membrane bound glycosylated hemoprotein, requires iron as a cofactor, and it plays a key role in thyroid hormone biosynthesis [8,9]. Iron deficiency in turn, can also trigger hypothyroidism. Symptoms like dementia, mental sluggishness are seen in both Iron deficiency anaemia and in Hypothyroidism. According to latest research, elderly people with Hypothyroidism may be at high risk of developing dementia. Studies revealed that, anaemia had been found to increase the risk of Parkinsons disease and dementia [10,11], and people with dementia were more likely to have Iron deficiency anaemia [12]. Also, many cases of Hypothyroidism in India remain undiagnosed and untreated. Thus, this study aims to assess the diagnostic role of serum Ferritin in detecting Iron deficiency anaemia earlier in hypothyroid individuals, and also to find the association between serum Ferritin and thyroid hormone levels in patients with Hypothyroidism.

Materials and Methods

Study design: This study was a cross-sectional observational study, which was conducted in a tertiary care medical college and hospital of South Tamilnadu. The study protocol was approved by the institutional ethical committee of the hospital (TIREC REF NO: 765/PHY/2015). After explaining the study procedure thoroughly, both oral and informed written consent was obtained from all the study participants. Emphasis was given that participation in this study was voluntary. The study involved a total of 100 subjects (50 subjects of newly diagnosed hypothyroids as cases and 50 subjects of normothyroids as controls) within age group 20-50 years and

of both genders. They were recruited from the non-communicable disease OPD, Medicine OPD and from the medicine wards and were included in the study after considering suitable inclusion and exclusion criteria.

Study Procedure: Under strict aseptic precautions, about 3 ml of venous blood was collected from the antecubital vein from both the study groups using 5ml disposable syringe in fasting condition. Serum was separated within half an hour by centrifugation and was stored at 2-8°C temperature till analysis was done. Serum TSH, Serum Total T3 and Serum total T4 were determined by Fully Automated Biochemical Analyzer EM 360 – 60519 and quality control was performed by ERBA norms. The normal values of Total T3 → 0.5-2ng/ml, Total T4 → 4.4-11.6µg/dl and TSH → 0.5-7µIU/ml. The diagnosis of Hypothyroidism was based on laboratory criteria of decreased serum Total T4 associated with increased TSH (> 10µIU /ml). The quality control used for determining Serum Ferritin was SPINREACT Ferritin control (Ref: 1107044). All these parameters were compared with age and sex matched healthy normothyroids. The normal values of serum Ferritin in Males → 20-200µg/dl and in Females → 20-110µg/dl.

Statistical Analysis: Statistical analysis was performed with Software Package for social sciences version 21.0 (SPSS 21.0).

Results were tabulated and compared between Normothyroids and Hypothyroids as mean ± SD, Pearson correlation analysis, 't' test - Paired sample test and One -Way ANOVA test. Statistically significant difference was considered when p value was less than 0.05 (p < 0.05).

Results

Table 1: Sex-wise Distribution of the Respondents (Hypothyroids)

Sex	No. of Respondents	Percentage
Male	12	24.00
Female	38	76.00*
Total	50	100.00

*Significant

Table 1- depicts that, out of 50 (N) Hypothyroid individuals included in our study, Hypothyroidism was much more common among females; Males: 24% and Females: 76%

Table 2: Comparison of Thyroid hormones and Serum Ferritin levels between Normal Thyroid and Hypothyroid individuals

Parameters	Normothyroid individuals	Hypothyroid individuals	p- value
T3	1.598 ±0.200	3490±11.420	0.000**
T4	9.214±2.284	6.060 ±2.568	0.000**
TSH	1.072 ±1.016	39.836 ±19.143	0.000**
Ferritin	85.734±7.395	65.433 ± 179.053	0.016*

** - Highly Significant (p < 0.001), * - Significant (p < 0.05)

Table 2- shows a comparison of thyroid hormones and Serum Ferritin levels between Normal Thyroid and Hypothyroid individuals.

It demonstrates a highly statistically significant difference of Thyroid hormones and Serum Ferritin levels between Normal Thyroid and Hypothyroid

individuals with ($p < 0.001$) and ($p < 0.05$) respectively. Serum Ferritin levels and serum Total T3, Total T4 levels were found to be decreased, while TSH levels were found to be increased in Hypothyroid individuals, when compared to Normothyroids.

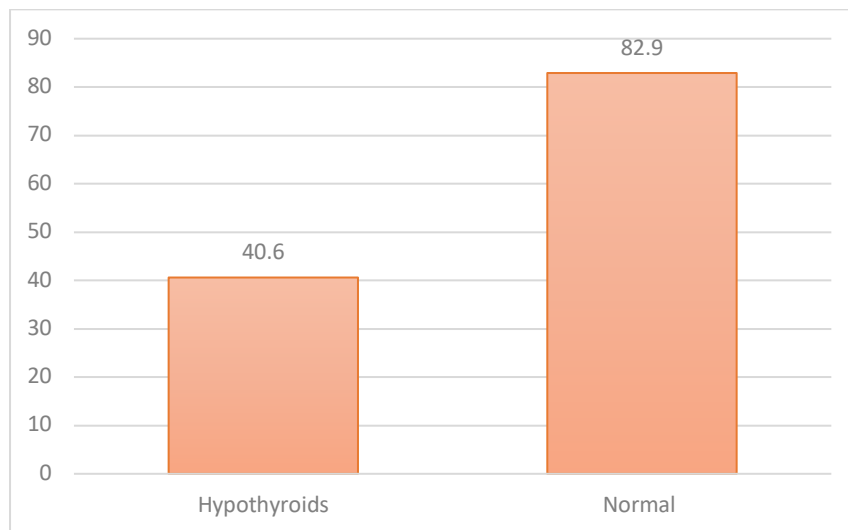


Figure 1: Comparison of mean Serum Ferritin levels between hypothyroid individuals and Normothyroid individuals

Figure 1- shows a graphical comparison of mean Serum Ferritin levels between hypothyroid individuals and Normothyroid individuals. The mean serum Ferritin levels were significantly lowered in Hypothyroid individuals (40.6 µg/dl), when compared with the Normothyroid individuals (82.9 µg/dl).

Table 3: Correlation between Serum Ferritin and Thyroid hormones in hypothyroid individuals

		Serum Total T3	Serum Total T4	Serum TSH
Serum Ferritin	Pearson correlation	0.093	0.358**	0.094
	Sig.(2-tailed)	0.52	0.01	0.51
	N	50	50	50

** - Correlation is significant at the 0.01 level (2-tailed)

Table 3- demonstrates the Correlation between Serum Ferritin and Thyroid hormone levels in hypothyroid individuals.

There was a positive correlation between Serum Ferritin levels and Total T3, Total T4 and TSH levels in

Hypothyroids ($r=0.093$, $r=0.358^{**}$, $r=0.094$) respectively.

There was also a highly statistically significant positive correlation between Serum Ferritin and Total T4 levels in hypothyroid individuals.

Table 4: Association between Serum Ferritin levels and Thyroid hormone levels in hypothyroid individuals

Variables	Serum Ferritin with T3, T4, TSH	t-value	Difference	p-value
Total T3	63.07±179.99	-2.478	49	.017*
Total T4	60.50±179.78	-2.380	49	.021*
TSH	26.72±179.90	-1.050	49	.299

** - Highly Significant ($p < 0.001$) * - Significant ($p < 0.05$)

Table 4- illustrates a paired 't' test, which shows the association between serum Ferritin and thyroid hormones in hypothyroid individuals. In Hypothyroids, the results showed statistically significant association between serum Ferritin with Total T3 and Total T4 ($p < 0.05$).

Table 5: Comparison of Serum Ferritin levels between hypothyroid individuals and Normothyroid individuals

Serum ferritin (in $\mu\text{g/l}$)	No. of Respondents	
	Normothyroids	Hypothyroids
Below 75	6 (12)	48* (96)
75 – 89.3	27 (54)	1 (2)
Above 89.3	17 (34)	1 (2)
Total	50 (100)	50 (100)

*significant

Table 5- illustrates that, in 96% (N=48) of Hypothyroid individuals, the serum Ferritin level was below $75\mu\text{g/l}$ and in 2% (N=1) was between 75-89.3 $\mu\text{g/l}$ and remaining 2% (N=1) had serum Ferritin levels above 89.3 $\mu\text{g/l}$; When compared to the Normothyroid individuals, they had 12% (N=6), 54% (N=27) and 34% (N=17) respectively.

Discussion

Thyroid gland is one of the largest endocrine gland in our body. It secretes two hormones, thyroxine (T4) and triiodothyronine (T3), which plays a critical role during cell differentiation in development and also helps to maintain the thermogenic and metabolic homeostasis in the adult [13]. Thyroid diseases have been found to be one of the most prevalent endocrinopathies across the world. Diseases of thyroid gland include those conditions associated with excessive release of thyroid hormones (hyperthyroidism) or those associated with deficiency of thyroid hormones (hypothyroidism) and mass lesions of the thyroid [14]. It has been determined from various studies that, in India about 42 million people suffer from thyroid disorders and, hypothyroidism is the most common, affecting about one in ten adults [1]. The normal functioning of the thyroid gland depends on certain trace elements like iron, selenium, iodine and zinc. So, their deficiencies also impairs thyroid function [15].

Thyroid peroxidase, a membrane bound glycosylated hemoprotein is required for the iodization of tyrosine residues of thyroglobulin and formation of T3 and T4 [16]. Hypothyroidism is associated with reduced erythrocyte mass, due to reduction in plasma volume and hence, it is undetectable by routine haemoglobin estimation. Ferritin is a combination of apoferritin with iron. In our body, majority of iron is stored in this form in reticuloendothelial cells, hepatic parenchymal cells and skeletal muscles. Hence, Ferritin is an important indicator of body iron stores. Previously conducted

studies reveal that, serum Ferritin level is altered in Hypothyroidism. Reduced iron or more specifically saying, reduced Ferritin level is found to be one of the most over- looked causes of decreased thyroid function [17]. In our study, mean serum Ferritin levels were significantly decreased in hypothyroids (40.6 $\mu\text{g/dl}$), when compared with normothyroids (82.9 $\mu\text{g/dl}$) with female predominance. Hence, alterations in serum Ferritin reflects thyroid function. The findings in our study were in accordance with another study done by Sahana KR, Kruthi BN et al. [18]. The results of our study were found to be similar to a study done by Ashuma Sachdeva, Veena Singh et al. which revealed that depletion of iron stores will cause reduction in serum FT3 & serum FT4 levels [19]. In our study, we have found a positive correlation between serum Ferritin and Total T3 ($r=0.093$), Total T4 ($r=0.358^{**}$) and TSH ($r=0.094$) in Hypothyroid individuals. Similarly in our study, 't' test - paired sample test revealed that, there was a statistically significant positive association between serum Ferritin and Total T3, Total T4 levels in Hypothyroids ($p < 0.05$). That is, low level of serum Ferritin in hypothyroidism is more associated with decreased thyroid hormones- Total (T3 and T4).

There is reduction in reactive oxygen species (ROS) production and antioxidant activity due to oxidative stress, which results in immunosuppression in Hypothyroids. Also, serum Ferritin plays an efficient role in iron sequestration, with some antioxidant properties [20, 21, 22, 23]. With increasing TSH concentration in the body, as seen in hypothyroidism, the levels of antioxidants including serum Ferritin decreases [24]. Our study findings disagree with the study findings reported by Farooq MS et al. who found negative correlation between serum Ferritin and serum TSH [25]. Our study results indicate that, there is significant reduction in serum Ferritin levels in Hypothyroids, when compared to Normothyroids. This finding may be

due to effect of TSH, at higher concentrations seen in Hypothyroidism, which induces inflammatory cytokines and decreases the concentration of antioxidants in the body.

In our study, comparison of serum Ferritin levels between Normothyroids and Hypothyroids also revealed that, in 96% (N=48) of Hypothyroids, the serum Ferritin levels were below 75µg/l and 2% (N=1) was between 75-89.3µg/l and in 2% (N=1) had serum Ferritin levels above 89.3µg/l, when compared to the Normothyroids.

Normothyroid group had 12% (N=6), 54% (N=27), 34% (N=17) respectively. Hence, the comparison states that, serum Ferritin levels has been found to be significantly decreased in newly diagnosed Hypothyroids than in Normothyroids. So, serum Ferritin measurement in hypothyroidism is very useful in evaluating thyroid hormone action on peripheral tissues.

So, the aim of our study is to assess whether serum Ferritin can be used as a diagnostic tool to detect iron deficiency anaemia earlier in Hypothyroidism, even though various laboratory values within iron panel are available. Though, bone marrow biopsy using iron stain is gold standard, a low serum Ferritin (12µg /L) remains highly specific for iron depletion [26]. Also, serum Ferritin is an important test to distinguish between iron deficiency anaemia and anaemia of chronic disease. Hypothyroidism and ascorbate deficiency are the only two conditions other than iron deficiency which have been found to lower serum Ferritin.

Literature reveals that in Hypothyroidism, there is reduced levels of hydrochloric acid that lead to poor iron absorption [27]. Also, Hypothyroidism results in lowered body temperature leading to production of fewer red blood cells from temperature sensitive bone marrow. Most of the symptoms of anaemia mimic Hypothyroidism. Examples are hair loss, fatigue, shortness of breath, palpitations, cognitive decline including memory impairment, attention deficit disorders, language issues and dementia [28, 29].

According to ICMR, the recommended daily allowance (RDA) of iron for adults in India is 28mg for men and 30mg for women. Studies reveal that iron deficiency anaemia develops slowly over several months due to the presence of serum Ferritin as iron stores. An average adult male has about 1000 mg of stored iron, which is enough for about 3 years and average adult female has about 300mg of stored iron, which is enough for six months [30].

Previous studies also stated that, many healthcare providers start iron supplements, when serum Ferritin level goes below 60 mg/dl, and it can take three to six months to restore serum iron levels and to normalize serum Ferritin levels with oral iron therapy. Serum Ferritin outperforms transferrin saturation, mean cell volume, and red cell zinc protoporphyrin levels in terms of sensitivity and specificity at any level [31]. As many cases of Hypothyroidism in India remain undiagnosed and untreated, it will lead to various complications. So, serum Ferritin can be used as a diagnostic tool to detect iron deficiency anaemia earlier in Hypothyroids, and by proper early intervention, various symptoms and side effects can be wisely prevented.

Conclusion

Serum Ferritin can be used as an effective diagnostic tool to detect Iron deficiency anaemia earlier in Hypothyroidism, and can be done as a routine investigation along with the thyroid function tests in suspected hypothyroid cases. In newly diagnosed hypothyroid cases, by providing proper earlier intervention with thyroid medications, as well as with iron supplements, we can prevent the risk of developing dementia, cognitive impairment and attention deficit disorders. Hence, we strongly recommend early diagnosis, proper intervention and simultaneous treatment of both iron deficiency anaemia as well as hypothyroidism, which will be greatly helpful in preventing hypothyroid individuals from developing these serious health ailments due to associated iron deficiency anaemia, as it is a preventable cause of illness.

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