



STUDY OF HEMATOLOGICAL CHANGES AND PATTERN OF ANEMIA IN HYPOTHYROID PATIENTS

Physiology

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ABSTRACT

Background & Objectives: Thyroid function disorders are among the most common endocrine diseases. Females are affected more than males, especially during the reproductive age. Hypothyroidism is the most prevalent type with a reported frequency of 2–5% worldwide. Thyroid hormones are essential for virtually all body tissues. So deficiency may affect hematopoiesis in bone marrow and eventually hematopoietic cells. Hematological abnormalities have frequently been reported in thyroid disorders. Anemia is frequently observed in patients with primary hypothyroidism. This study aimed to evaluate the hematological changes and pattern of anemia in primary hypothyroid patients. Methods: This is a cross sectional study conducted on newly diagnosed 100 patients with primary hypothyroidism, between 18–60 years. They were categorized as 35 patients of subclinical and 65 patients of overt hypothyroidism based on TSH level. Patients' who fulfill the inclusion exclusion criteria were evaluated for thyroid function tests (T3, T4 and TSH) and hematological parameters (CBC and PBF). Results: Our analysis revealed an overall prevalence rate of anemia was 56% in patients with hypothyroidism which is higher than the WHO reported data of prevalence of anemia throughout the world. Our results showed that prevalence of normocytic normochromic anemia was significantly higher, microcytic anemia had the second rank, while macrocytic anemia had the lowest prevalence rate. On subgroup analysis, there was no statistical difference between subclinical hypothyroid and overt hypothyroid, in term of hematological parameters and type of anemia. Conclusion: Without proper diagnosis and effective treatment of the underlying thyroid disease, it is often difficult to achieve a complete correction of the anemia. The high prevalence of anemia in patients with hypothyroidism suggests screening for hypothyroidism during the differential diagnosis of cases presenting with anemia.

KEYWORDS

Hypothyroidism, subclinical, overt, anemia, hematological, thyroid, blood.

INTRODUCTION

Thyroid function disorders are among the most common endocrine diseases. Females are affected more than males, especially during the reproductive age. The prevalence of thyroid disorders differs from one society to another. Hypothyroidism is the most prevalent type with a reported frequency of 2–5% worldwide. Also, the prevalence of subclinical hypothyroidism is approximately 4–8.5% and it can increase up to 20% in women aged 60 years or older. Prevalence of overt hypothyroidism varies between 0.1–2 %. Alternatively, hyperthyroidism is less prevalent with a frequency ranging between 0.5 and 2% in women.¹

Thyroid hormones are essential for the normal development, differentiation, metabolic balance, and physiological function of virtually all tissues.² Thyroid hormones may affect Hematopoiesis in bone marrow and eventually hematopoietic cells.³ Hematological abnormalities have frequently been reported in thyroid disorders. In 1979, Fein showed that Graves' disease is associated with anemia.⁴ During the latter part of the nineteenth century Charcot and Kocher described the association of anemia with primary hypothyroidism and with hypothyroidism following thyroidectomy.^{5,6} Horton observed a decrease number of red blood cells (RBCs) in the peripheral blood of patients after thyroidectomy and anemia has been defined in 20–60% of the patients with hypothyroidism.⁸ Although early reports showed that Graves' disease is also associated with hematological disorders,^{4,7,9} anemia is not frequently observed in patients with hyperthyroidism, whereas erythrocytosis is common.^{9,10} Furthermore, it has been found that all hematological parameters return to normal when a euthyroid state is achieved.^{7,11} Although some other studies reported contradictory results.

As far as white blood cells and thrombocytes are concerned, a slightly depressed total leucocyte count, neutropenia, and thrombocytopenia have been observed in hypothyroid patients.⁹ Furthermore, elevated, normal, or slightly depressed total leucocyte counts have been found in hyperthyroid patients, with only a relative decrease in the number of neutrophils and a relative increase in the number of eosinophils and mononuclear cells (MNCs). Nevertheless, hyperplasia of all myeloid cell lines in hyperthyroidism and their hypoplasia in hypothyroidism were reported by Axelrod.¹²

These observations confirmed the association between thyroid gland dysfunction and hematopoiesis. Previously published studies suggested that there is an essential relationship between the hypothyroid state and low levels of iron, vitamin B12, and folic acid in the human body.^{8,18} Furthermore, it has been postulated that the influence of thyroid hormones on hematopoiesis involves an increased production of erythropoietin or hematopoietic factors by non erythroid cells.^{19,20} However, a growing number of studies have demonstrated a direct role of THs in normal human and animal erythropoiesis.^{3,21}

During the past century there has been an extensive accumulation of literature dealing with the relationship between the function of the thyroid gland and the status of the blood and bone marrow. Much of it is fragmentary and contradictory. We have attempted to gather the available facts and augment these with original observations in order to provide a clearly defined point of departure for future studies.

Anemia frequently occurs during the course of clinical hypothyroidism, and may present a challenging diagnostic problem. It is important to recognize that anemia may develop before other manifestations of hypothyroidism, or it may develop late in the course of illness. Several of the symptoms of hypothyroidism and anemia are similar, and the diagnosis of the former condition is often obscured when the latter is present. Without proper diagnosis and effective treatment of the underlying thyroid disease, it is often difficult to achieve a complete correction of the anemia. This study aimed to evaluate effects of hypothyroidism on hematological parameters.

OBJECTIVES

To study the hematological changes and type of anemia in hypothyroid patients.

MATERIALS & METHODS

This is a cross sectional and hospital-based study was carried out in the department of pathology and department of medicine, S.P. Medical College and attached group of hospitals, Bikaner (Rajasthan). The study was conducted on patients with primary hypothyroidism. The duration of study was one year, from January 2018 to December 2018.

Study Population: 100 patients (between 18–60 years) with

hypothyroidism were selected for the study after fulfilling inclusion criteria. They were categorized as 35 patients of subclinical hypothyroid and 65 patients of overt hypothyroidism based on TSH levels for each group.

Inclusion criteria: Patients with primary hypothyroidism (subclinical and overt). All patients in the study group were selected without any bias for age, sex and thyroid status.

Exclusion criteria: Patients with anemia due to other reasons including hemolytic anemia, malignancy, acute blood losses from gastrointestinal (peptic ulcer, hemorrhoids) or genitourinary system; presence of any co-morbid disease like renal failure, coronary heart disease, diabetes mellitus, tuberculosis; any liver disorder and any endocrine disorder other than thyroid, hemoglobinopathies, bleeding diathesis; patients with any autoimmune disorder (rheumatoid arthritis, celiac disease) cigarette smoking; patients with bone marrow suppression, or blood transfusion in last 3 months; patients who are under the treatment that affects blood parameter such as iron supplements or vitamins or who had received anemia treatment previously; pregnant and breast feeding mothers.

Data Collection: This study was conducted after ethical approval by the Rajasthan University of health and sciences, Jaipur. An informed consent was obtained from all participants. A questionnaire was specifically designed to obtain patients' information, which helps in either including or excluding certain individuals in or from the study respectively. Clinical data were taken from patients' medical record and investigations.

Study variables and methods of measurement: The patients who satisfy inclusion criteria were evaluated for thyroid function tests (T3, T4 and TSH) and hematological parameters (CBC and PBF).

[A]. Collection of blood sample: Venous blood was collected under aseptic condition after cleaning the area around the vein with 70% alcohol. Two early morning blood samples (2ml each) were obtained from each participant in two different vials. EDTA anticoagulant containing vial was used for hematology evaluation. Plain vial was used for T3/T4/TSH measurements. A Peripheral blood smear was also prepared immediately.

[B]. Thyroid hormone measurements: Whole blood sample collected in plain vial centrifuged at 3000 rpm for 20 min, and then separated serum samples were stored frozen at -20°C until use. The quantitative measurement of thyroid hormones T3, T4 and TSH was measured by radioimmunoassay methods using commercial ELISA kits. Normal reference range provided by kit provider is:-

TSH: 0.270-4.20 $\mu\text{IU/mL}$; total T4: 66-181 nmol/L; total T3: 1.30-3.10 nmol/L; free T3: 0.22-6.78 pmol/L; free T4: 10.3-35 pmol/L.

Subclinical hypothyroidism is defined as a condition without typical symptoms of hypothyroidism, elevated TSH ($>5 \mu\text{IU/mL}$), and normal circulating thyroid hormone. Overt hypothyroidism is defined as a clinical syndrome of hypothyroidism associated with elevated TSH and decreased serum level of T4 or T3.

[C]. Determination of hematological profile: Samples collected in EDTA vials were used for hematological evaluation. Sysmex automated hematology analyzer was used to analyze the sample. Hematology analyzers are based on electric impedance counting or optical light scatter counting techniques. Anemia is considered when hemoglobin levels $<12 \text{ g/dL}$ in women and $<13 \text{ g/dL}$ in men (According to World Health Organization).

[D]. Peripheral blood smear: Peripheral blood smears were prepared, stained with field's and Leishman's stain and examined to verify the values obtained from Sysmex automated hematology analyzer.

Data analysis & Statistical technique: The data collected in this study was analyzed using SPSS computer program. The mean and standard deviations of the blood parameters were calculated and Statistical Student's t-test was used for comparison ($p \text{ value} \leq 0.05$ is considered significant).

Results:

Demographic data and analysis of CBC values of the sub groups of hypothyroid patients participating in this study are shown in Table 1.

This is observational, cross sectional study included 100 patients of primary hypothyroidism, aged between 18 –60 years who were categorized as 35 patients of subclinical hypothyroid and 65 patients of overt hypothyroidism based on TSH levels for each group. The mean ages were 38 years and 40 years $\pm$ SD, respectively.

The male participants in this study were 20% of study population and females were 80%. Male: Female ratio was 1: 5. Subclinical hyperthyroid group included 7 males and 28 females while overt hypothyroid group included 13 males and 52 females mainly in the ages between 18-60 years.

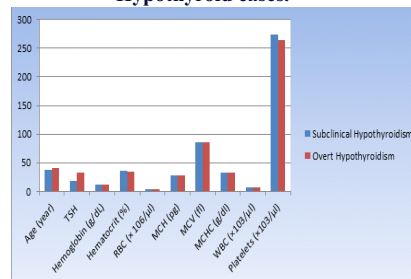
In our study; anemia is considered when hemoglobin levels $<12 \text{ g/dL}$ in women and $<13 \text{ g/dL}$ in men (According to World Health Organization). Our analysis revealed an overall prevalence rate of anemia is 56% in patients with hypothyroidism. Based on the data of the World Health Organization (WHO), anemia has an estimated prevalence of 24.8% throughout the world. The pooled rate in our study is higher than that reported by WHO. Anemia was determined in 18% of those with subclinical hypothyroidism and 38% of those with overt hypothyroidism. There was no statistical difference in terms of anemia frequency between subclinical and overt hypothyroid groups ($p=0.407$).

There was no statistical difference between RBC indices, hematocrit, hemoglobin, RBC, WBC and platelet counts of the patients (Table 1).

Table 1: Demographic and hematological parameters of hypothyroid patients

	Subclinical Hypothyroidism	Overt Hypothyroidism	P value
Number	35	65	-
Gender (male/female)	7/28	13/52	-
Age (year)	38 $\pm$ 11.28	40 $\pm$ 11.30	0.4
TSH (0.27-4.2 $\mu\text{IU/mL}$)	17.85 $\pm$ 11.52	32.18 $\pm$ 19.24	0.000
Hemoglobin (g/dL)	11.5 $\pm$ 2.03	11.15 $\pm$ 1.99	0.407
Hematocrit (%)	35.42 $\pm$ 4.71	34.6 $\pm$ 4.846	0.417
RBC ($\times 10^6/\mu\text{L}$)	4.1 $\pm$ 0.36	4.0 $\pm$ 0.446	0.258
MCH (pg)	27.8 $\pm$ 4.43	27.9 $\pm$ 4.247	0.912
MCV (fl)	85.70 $\pm$ 9.12	86.50 $\pm$ 9.732	0.690
MCHC (g/dl)	32.3 $\pm$ 2.23	32.1 $\pm$ 2.04	0.652
WBC ($\times 10^3/\mu\text{L}$)	7.53 $\pm$ 1.94	7.06 $\pm$ 1.70	0.211
Platelets ($\times 10^3/\mu\text{L}$)	273.3 $\pm$ 67.82	264.4 $\pm$ 72.95	0.554

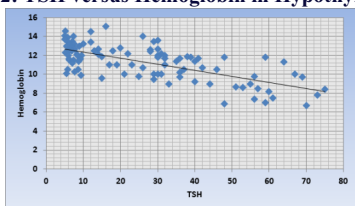
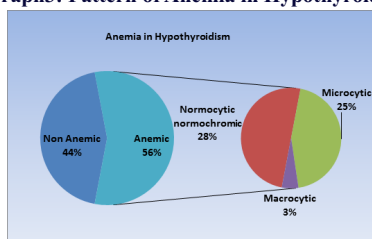
Graph 1. Comparison of mean of various parameters in Hypothyroid cases.



Pattern of anemia in patients with hypothyroid are shown at Table 2. Study of type of anemia shows that anemia of chronic disease was the most common anemia 50%. Microcytic hypochromic anemia (iron deficiency anemia) was second most common 45% and frequency of macrocytic anemia is found in 3%. In the subgroup analysis of the hypothyroid patients, the most frequently seen anemia type was the anemia of chronic disease in patients with overt and subclinical hypothyroidism. The ratio depending on this type of anemia was determined to be 53% in patients with clinical hypothyroidism, and 44% in patients with subclinical hypothyroidism. There was microcytic anemia in overt and subclinical hypothyroid patients respectively 39%, 56%. There was macrocytic anemia in overt and subclinical hypothyroid patients respectively 3%, 0%. There was no statistical difference between subclinical hypothyroid and overt hypothyroid, in term of type of anemia (Table 2).

Table 2: Type of anemia in primary hypothyroid patients.

Type of anemia	Subclinical Hypothyroidism	Overt Hypothyroidism	Total
Normocytic Normochromic (MCV 80-100 fl) (MCHC 30-36 %)	8 (44%)	20(53%)	28 (50%)
Microcytic Hypochromic (MCV <80 fl) (MCHC < 30)	10 (56%)	15 (39%)	25 (45%)
Macrocytic (MCV > 100 fl)	0	3 (8%)	3 (5%)
Total	18	38	56

Graph 2: TSH versus Hemoglobin in Hypothyroid cases**Graph3: Pattern of Anemia in Hypothyroidism**

DISCUSSION

Hypothyroidism constitutes a global health concern among clinical society. The disease varies in symptoms and signs according to the age of patients and ranges from asymptomatic to life-threatening illness mostly due to multiple organ dysfunctions as thyroid hormones are required for several metabolic activities²². Based on thyroxine and thyroid-stimulating hormone (TSH) level, hypothyroidism can be categorized into two pathologic subtypes. Subclinical hypothyroidism is characterized by low TSH and normal thyroxine production in addition to no or few clinical symptoms; therefore, the diagnosis of subclinical hypothyroidism is problematic. Meanwhile, overt hypothyroidism is associated with a decrease in both TSH and thyroxine levels with apparent clinical manifestations²³. Different etiologies are responsible for hypothyroidism affection. Hashimoto's hypothyroidism followed by idiopathic hypothyroidism were the common causes of hypothyroidism reported by Hunter, et al.²⁴.

Thyroid hormones provide an essential component in the normal process of erythropoiesis. Therefore, dysfunction of thyroid hormones results in anemia in those patients. The most common type of anemia in hypothyroidism patients is normocytic normochromic; however different types of anemia were observed such as macrocytic and microcytic anemia. The prevalence of anemia was found to be higher in overt type rather than subclinical type²⁵. Multiple etiologies lie behind the occurrence of anemia in many patients, including, nutritional deficiencies and chronic diseases. However, the underlying cause in approximately one-third of cases remains unexplained.²⁶ Noteworthy, anemia has been documented to occur in a large portion of patients with hypothyroidism, ranging from 23% to 60%.²⁵ It has been reported that anemia is the first presenting sign in patients with hypothyroidism.²⁷ Even though a considerable number of patients present with recurring anemia or persistent anemia that is unresponsive to treatment, most of these patients get better with levothyroxine, where a significant increase in hemoglobin level is noticeable.²⁸ That being said, with the lack of systematic reviews to study the association between anemia and hypothyroidism, no conclusions can be drawn out about the indication of screening for thyroid function disturbance in patients presenting with anemia.

Therefore, we conducted this investigation in order to assess the current literature and to determine the association between anemia and

hypothyroidism.

Our analysis revealed an overall prevalence rate of anemia was 56% in patients with hypothyroidism. The study of Rabet-Bensalah et al.²⁹ estimated the lowest reported prevalence rate of anemia in patients with hypothyroidism. On the other hand, the study of Anand et al.²⁵ reported the highest prevalence rate in the literature of 62.14%.

Based on the data of the World Health Organization (WHO), anemia has an estimated prevalence of 24.8% throughout the world.³⁰ The pooled rate in our study is higher than that reported by WHO, therefore, we can conclude that hypothyroidism can be considered as a risk factor for anemia, overall. However, the current evidence is insufficient and of poor quality. So, more studies are warranted to further elaborate such association.

Unfortunately, we encountered a significantly considerable heterogeneity in the analysis. The wide variation in laboratory definition of anemia among analyzed studies could be one of the factors attributing to the resulting substantial heterogeneity. This is also one of the limitations encountered in the literature, leading to a wide variation in prevalence rates of anemia among hypothyroidism patients. The majority of studies used the same laboratory definition of anemia (Hemoglobin below 12 g/dL for females and 13 g/dL for males).^{25,29} However, Chu, et al.³¹ used the criteria of Hemoglobin level below the third percentile as the defining criteria of anemia among children and adolescents. On the other hand, other authors used a Hemoglobin level below 14 gm% for males and 12 gm% for females³² and Hemoglobin below 11 regardless of gender as the criteria to define anemia.

In hypothyroidism, a drop in baseline metabolic rate with a reduction in baseline cellular oxygen consumption may result in a reduced secretion of erythropoietin with subsequent reduction in Hemoglobin concentration and, eventually, causing anemias either normocytic normochromic, microcytic, or macrocytic anemia, depending on presenting comorbidities.^{8,33}

In further subgroup analysis, our results showed that prevalence of normocytic normochromic anemia among patients with hypothyroidism was significantly higher than the other two types of anemias. On the other hand, microcytic anemia had the second rank, while macrocytic anemia had the lowest prevalence rate. This goes in line with what has been reported by other studies in literature.²⁵ They included hypothyroid patients who were untreated and also non-pregnant, so that they can control for such confounding factors, and they found that normocytic anemia was the most common type of anemia presenting in those patients. This can occur directly or indirectly through inhibition of erythroid colony development as a result of the lack of thyroid hormones and diminution of erythropoietin hormone level, with further reduction in oxygen transfer to tissues.³⁴ Some authors have reported that macrocytic anemia was the most prevalent type among hypothyroid patients, while others revealed that microcytic anemia was the most presenting type. Noteworthy, it is important to thoroughly investigate the cause of anemia of either type and properly manage it accordingly. Cinemre et al.³⁵ pointed out the importance of addressing hypothyroidism (subclinical type) in patients presenting with iron-deficiency anemia- unresponsive to treatment. They found that in hypothyroidism patients who were treated with oral iron supplementations that hemoglobin levels had a mean decrease of 0.4 gm/dl only, while those on iron plus levothyroxine a mean reduction in hemoglobin level of 1.9 gm/dl was noted.

From the perspective of the type of hypothyroidism, relatively similar prevalence rates of anemia in our study were noted between subclinical hypothyroidism and overt hypothyroidism. The rates in each type of hypothyroidism are comparably similar to that reported by WHO as regards the worldwide population (24.80%).³⁰ This points out that the frequency of anemia in subclinical and overt hypothyroidism is relatively similar to that of the general population. Moreover, this finding reveals that the risk of anemia in hypothyroidism still requires further investigation, an increase in prevalence rates of anemia could have been confounded by other co-existing comorbidities. Herein, we declare that the prevalence of anemia in hypothyroidism, overall, is relatively higher than the general population as declared by the WHO; however, the prevalence of anemia among each type of hypothyroidism is not higher but is relatively similar to that of the

general population. Moreover, we point out that current literature provides insufficient evidence regarding the risk of anemia in patients with subclinical and overt hypothyroidism.

In this study we have encountered several limitations. First, the selection of untreated subclinical cases, who merely have symptoms of hypothyroidism. Second, the lack of a standardized laboratory definition of anemia in previous reported studies, so this may be the factor for variability of prevalence rates of anemia. Third, the exclusion of previously treated cases in terms of anemia treatment or thyroxine medications which significantly impact the interpretation of findings. Finally, thyroid hormone levels were measured once only at baseline; patients who had subclinical hypothyroidism and developed overt hypothyroidism over time, or those reverted to euthyroidism may have been misclassified.

CONCLUSIONS

The high prevalence of anemia in patients with hypothyroidism suggests screening for hypothyroidism during the differential diagnosis of cases presenting with anemia. Normocytic anemia is the most common type in hypothyroidism. We strongly recommend that physicians should investigate for hypothyroidism when patients present with anemia that is either of unknown etiology or unresponsive to treatment. However, current evidence is insufficient and more studies are warranted to assess the association between anemia and hypothyroidism.

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Conflicts of Interest: None.

REFERENCES

1. Refaat B. Prevalence and characteristics of anemia associated with thyroid disorders in non-pregnant Saudi women during the childbearing age: a cross-sectional study. *Biomed J*. 2015; 38(4):307.
2. Franchini M, Montagnana M, Manzato F, Vescovi PP. Thyroid dysfunction and hemostasis: An issue still unresolved. *Semin Thromb Hemost* 2009; 35:288-94.
3. Golde DW, Bersch N, Chopra IJ, Cline MJ. Thyroid hormones stimulate erythropoiesis in vitro. *Br J Haematol*. 1977; 37(2):173-7.
4. Fein HG, Rivlin RS. Anemia in thyroid diseases. *Med Clin North Am* 1975; 59(5):1133-45.
5. Charcot M: Myxœdème, cachexie pachydermique ou état cretinoïde. *Gaz Hop Paris* 54:73, 1881.
6. Kocher T: Ueber Kropfexstirpation und Ihre Folgen. *Arch Klin Chir* 29:254, 1883.
7. J. A. Perlman and P. M. Sternthal. Effect of 131I on the anemia of hyperthyroidism, *Journal of Chronic Diseases*, vol. 36, no. 5, pp. 405–412, 1983.
8. Horton L, Coburn R, England J and Himsworth R. The Haemtolgy of Hhypothyroidism. *Quarterly Journal of Medicine*, 1976 vol. 45, no. 177, pp. 101-123.
9. Lima CS, Zantut Wittmann DE, Castro V, Tambascia MA, Lorand-Metze I, Saad ST, et al. Pancytopenia in untreated patients with Graves' disease. *Thyroid* 2006; 16(4):403-9.
10. Iddah M, Macharia B, Ng'wena A, Keter A, Ofulla A. Thyroid hormones and hematological indices levels in thyroid disorders patients at Moi teaching and referral hospital, Western Kenya. *ISRN endocrinol*. 2013.
11. Bashir H, Bhat MH, Farooq R, Majid S, Shoib S and Hamid R, (2012). Comparison of hematological parameters in untreated and treated subclinical hypothyroidism and primary hypothyroidism patients. [online]. *Med J Islam Repub Iran*. ;26(4):172-8.
12. Axelrod AR, Berman L: The bone marrow in hyperthyroidism and hypothyroidism. *Blood* 6:436, 1951.
13. Mackenzie GM. Anemia in hypothyroidism. *JAMA*. 1926; 86(7):462-64.
14. M.M. Kunde, M.F. Green, G.Burns: Blood changes in experimental hypo- and hyperthyroidism (rabbit) *American Journal of Physiology* 1932: 99(2)469-476.
15. Bomford R: Anaemia in myxoedema: and the role of the thyroid gland in erythropoiesis. *Quart. J. Med.* 7: 495, 1938.
16. Tudhope, G. R., and G. M. Wilson: Anemia in hypothyroidism: Incidence, pathogenesis and response to treatment. *Quart J Med* 29: 573, 1960.
17. Larsson SO: Anemia and iron metabolism in hypothyroidism. *Acta Medica Scand inavica* 157:349, 1967.
18. Hines JD, Halsted CH, Griggs RC, Harris JW: Megaloblastic anemia secondary to folate deficiency associated with hypothyroidism. *Ann Intern Med* 68:792, 1968.
19. N. Dainiak, D. Sutter, and S. Kreczko. L-triiodothyronine augments erythropoietic growth factor release from peripheral blood and bone marrow leukocytes, *Blood*, vol. 68, no. 6, pp. 1289–1297, 1986.
20. J. Fandrey, H. Pagel, S. Frede, M. Wolff, and W. Jelkmann, "Thyroid hormones enhance hypoxia-induced erythropoietin production in vitro," *Experimental Hematology*, vol. 22, no. 3, pp. 272–277, 1994.
21. L. A. Malgor, C. C. Blanc, E. Klainer et al., "Direct effects of thyroid hormones on bone marrow erythroid cells of rats," *Blood*, vol. 45, no. 5, pp. 671–679, 1975.
22. Surks MI and Hollowell JG. "Age-specific distribution of serum thyrotropin and antithyroid antibodies in the US population: implications for the prevalence of subclinical hypothyroidism". *The Journal of Clinical Endocrinology and Metabolism* 92.12(2007): 4575-4582.
23. Canaris GJ., et al. "The Colorado thyroid disease prevalence study". *Archives of Internal Medicine* 160.4 (2000): 526-534.
24. Virtanen M. "Manifestations of congenital hypothyroidism during the 1st week of life". *European Journal of Pediatrics* 147.3 (1988): 270-274.
25. Anand R., et al. "A study of prevalence and pattern of anemia in primary hypothyroidism". *International Journal of Medical Science and Public Health* 7.2 (2018): 153-160.
26. Patel KV. "Epidemiology of anemia in older adults". *Seminars in Hematology* 45.4 (2008): 210-217.
27. Christ-Crain M., et al. "Effect of restoration of euthyroidism on peripheral blood cells and erythropoietin in women with subclinical hypothyroidism". *Hormones* 2.4 (2003): 237-242.
28. Ravanbod M., et al. "Treatment of Iron-deficiency Anemia in Patients with Subclinical Hypothyroidism". *The American Journal of Medicine* 126.5 (2013): 420-424.
29. M'Rabet-Bensalah K., et al. "Thyroid dysfunction and anaemia in a large population-based study". *Clinical Endocrinology* 84.4 (2016): 627-631.
30. Benoist B., et al. "Worldwide Prevalence of Anemia: Who Global Database on Anemia". World Health Organization: Geneva, Switzerland (2008).
31. Chu JY., et al. "Anemia in children and adolescents with hypothyroidism". *Clinical Pediatrics* 20.11 (1981): 696-699.
32. Allen Dawson M and W Yarbrow J. "Anemia in Hypothyroidism". *Southern Medical Journal* 63.8 (1970): 966-970.
33. Nightingale S., et al. "The haematology of hyperthyroidism". *QJM: An International Journal of Medicine* 47.185 (1978): 35-47.
34. Kühr T., et al. "Genetically determined target organ susceptibility in the pathogenesis of spontaneous autoimmune thyroiditis: aberrant expression of MHC-class II antigens and the possible role of virus". *Journal of Autoimmunity* 7.1 (1994): 13-25.
35. Cinemre H, Bilir C, Gokosmanoglu F, Bahcebasi T. Hematologic effects of levothyroxine in iron-deficient subclinical hypothyroid patients: A randomized, double-blind, controlled study. *J Clin Endocrinol Metab* 2009; 94:151-6.