ERYTHROCYTE INDICES IN PATIENTS OF THYROID DYSFUNCTION – A CROSS-SECTIONAL STUDY IN A TERTIARY CARE HOSPITAL

Biochemistry

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ABSTRACT

Haematological abnormalities particularly anaemia are frequently associated with thyroid disorders. A study was conducted on 300 patients (within age of 20-60 year) of clinically suspected thyroid disorders to estimate and to compare different erythrocyte indices. Thyroid disorders were detected in 121 participants (40.4%) with 26% hypothyroidism and 14.4% hyperthyroidism. The frequency of subclinical and clinical hypothyroidism was 13.3% and 12.7% respectively. Anaemia was detected in 10.5% participants with normal thyroid profile and in 14.5% cases (17/121) with abnormal thyroid profile. Normocytic normochromic anaemia was mostly found. Present study shows hypothyroidism should be ruled out while investigating causes of anaemia. Early recognition and treatment of hypothyroidism is needed to prevent the risk of developing anaemia.

KEYWORDS

Anaemia, Erythrocyte indices, Hypothyroidism.

Introduction

Thyroid hormones have long been recognized for their importance in regulating normal development, cellular proliferation and differentiation, metabolism and physiological functions of virtually all tissues. They regulate haematopoiesis in bone marrow through proliferation of erythroid progenitor cells, transcription of erythropoietin and haemoglobin production. They are also prerequisite for normal B cell production in bone marrow[1,2].

Thyroid diseases are, arguably, among the commonest endocrine disorders world wide and India too, is no exception. Common thyroid disorders are hypothyroidism, hyperthyroidism and goitre. Hypothyroidism (subclinical and clinical) is the most prevalent type worldwide[3,4]. From different studies, it is observed that erythrocyte abnormalities are frequently associated with thyroid disorders. However, they are rarely investigated and related to the thyroid disorders [5,6,7]. Several forms of anaemia have been reported with thyroid disorder which may be normocytic normochromic, microcytic and/or macrocytic hypochromic [8]. Macrocytic anaemia may be unresponsive to vitamin B_{12} or folate. It has been suggested that thyroid hormone deficiency may lead to bone marrow repression and/or decrease in erythropoietin production due to the reduction of O_2 requirements or low levels of micronutrients like iron, folic acid and vitamin B_{12} etc.[9,10] Alterations in other haematological parameters such as leucopenia and neutropenia are common in hypothyroidism due to hypoplasia of myeloid cell lines [11]. Hyperthyroidism is rarely associated with anaemia [12].

Because of high prevalence of both thyroid dysfunction and anaemia in Indian population and a very few studies have been conducted on these problems in the sub-Himalayan region, a study was conducted to estimate and to compare different erythrocyte indices among the patients of different types of thyroid disorders with special emphasis on hypothyroidism both clinical and subclinical.

Materials and Methods

This was an institutional facility-based, non-interventional, descriptive, epidemiological study with cross-sectional design. The study was conducted on the patients attending Department of Biochemistry and Department of Pathology of North Bengal Medical College & Hospital (NBMCH), Sushrutnagar, Darjeeling during January, 2017 to June, 2017 after getting approval from Ethics Committee of NBMCH.

Inclusion Criteria:

Patients (male and female, aged 20-60 years) attending Department of Biochemistry, first time for thyroid function tests (TFT) and voluntarily given consent to participate.

Exclusion Criteria

a) Patients suffering from any thyroid disease, on anti-thyroid therapy or on any thyroid function modifying drugs.
b) Chronic diseases (nephropathy, hemoglobinopathies etc.) or any drugs causing anaemia.
c) Known anaemic or malnourished patients.
d) On prolong drug therapy such as anti-tubercular, chemotherapeutics, anti-psychotic, hormone (oral contraceptive pills), vitamins and/or iron supplementation.
e) Pregnancy or irregular menstrual cycle.
f) Acute illness.
g) Suffering from malignancy or on anti-cancer therapy.
h) Drug abuse or habitual alcoholic.
i) Chronic diseases (kidney disease, liver failure, diabetes mellitus).

Total 300 participants were chosen by systematic random sampling method and interviewed with a pre designed, pretested questionnaire. Five ml venous blood was collected from each participant after taking written consent, following standard protocol of blood collection under aseptic precaution and kept in EDTA and plain vial. Serum was separated by centrifugation for estimating fT4 and TSH by enzyme linked immunosorbent assay (ELISA) method at Biochemistry Department. EDTA blood was used to estimate Hb%, PCV, MCV, MCH, MCHC, RDW, RBC count by Blood Counter (Sysmex XS-800) at Pathology Department.

Reference range of serum TSH and fT4 were considered to be 0.50-5.50 µIU/ml and 0.7-2.0 ng/dl, respectively. Subclinical Hypothyroid (SCH) was defined as serum TSH level 5.50-10 µIU/ml with normal fT4.[13] Clinical hypothyroid was considered when TSH ≥10 µIU/ml and fT4 <0.7 ng/dl and primary hyperthyroid was granted when TSH <0.50 µIU/ml and fT4 >2.0 ng/dl.[14]

Anaemia was considered when Hb <11 g/dl (venous blood). Microcytic, normocytic, or macrocytic anaemia was classified as per MCV values such as <80 femtolitre (fl), 80-96 fl and ≥96 fl, respectively. The reference range for MCH was taken 27-33 picogram (pg); MCHC 33-36g/dl; PCV 35-50%; RDW-CV (Red cell distribution width- coefficient of variation) 11.5-14.5%. MCHC <33 g/dl indicated hypochromia.[19,20,21] Elevated RDW-CV is known as anisocytosis.

Statistical analyses were done as per requirement by using suitable statistical software (SPSS ver. 20). Shapiro- Wilk's test was done for normality testing and found that all data were skewed, except PCV. So the non-parametric tests were mandate for the statistical analyses.

Results and Analysis
Table 1: Descriptive statistics of different attributes

Parameters	Mean	Median	SD	SE of Mean
Age (years)	37.07	35	11.179	0.645
TSH (µIU/ml)	4.78	2.4	6.343	0.366
fT4 (ng/dl)	1.796	1.5	1.042	0.060
Hb (%)	12.09	12.25	1.668	0.096
PCV (%)	37.77	38.00	4.314	0.249
MCV (fl)	82.98	84.45	9.521	0.550
MCHC (g/dl)	32.06	32.00	1.511	0.087
MCH (pg/cell)	26.66	27.20	3.515	0.203
RDW-CV (%)	14.70	14.20	2.027	0.117
RBC count (trillion/l)	4.61	4.60	0.638	0.037

Thyroid disorders were detected in 121 participants (40.4%), and hypothyroidism was found to be more prevalent (78/121, 26%) than hyperthyroidism (43/121 or 14.4%). The frequency of subclinical and clinical hypothyroidism were 13.3% (40/121) and 12.7% (38/121), respectively. Most of the patients were within age of 30-40 year. Anaemia was detected in 10.5% (18/179 participants) with normal thyroid profile and 14.5% (17/121 cases) with abnormal thyroid profile. Normocytic normochromic anaemia was mostly found.

Table- 2: Relationship between fT4 groups and gender

Gender		fT4 (ng/dl)		
		<0.7	0.7-2.0	>2.0
Male	Count	5	22	10
	% within Gender	13.5%	59.5%	27.0%
Female	Count	10	188	65
	% within Gender	3.8%	71.5%	24.7%
Total	Count	15	210	75
	% of Total	5.0%	70.0%	25.0%

Pearson's Chi-square significance = 0.032. This meant that male and

females were dependently associated within the fT4 groups. If the distribution of this data was due entirely to chance, then there was only 3.2% chance of finding a discrepancy between the observed and expected distributions. Most of the participants (70%) were belonged to fT4 =0.7-2.0 ng/dl.

Table- 3: Relationship between TSH and MCV, MCHC groups

TSH Groups (µIU/ml)		MCV (fl)			MCHC (g/dl)		
		<80	80- 96	>96	<33	33- 36	>36
<0.5	Count	21	21	1	26	15	2
	% within TSH group	48.8	48.8	2.3	60.5	34.9	4.7
0.5- 5.5	Count	62	112	5	146	33	0
	% within TSH group	34.6	62.6	2.8	81.6	18.4	0.0
5.5- 10	Count	10	24	6	36	4	0
	% within TSH group	25.0	60.0	15.0	90.0	10.0	0.0
>10	Count	11	25	2	30	8	0
	% within TSH group	28.9	65.8	5.3	78.9	21.1	0.0
Chi-square test; significance		0.013			0.001		

When only female participants were considered significance level in TSH vs. MCV was found to be 0.001. MCV was dependently associated with TSH. The propensity of hypochromicity was found to be increased, with the increment of TSH.

Table- 4: Relationship between fT4 group and PCV, MCV, MCHC groups

fT4 groups(ng/dl)		PCV group(%)			MCV(fl)			MCHC(g/dl)			RBC count (trillion cells/L)		
		< 35	35-50	>50	<80	80-96	≥96	<33	33-36	≥36	<4	4-5.5	≥5.5
<0.7	Count	5	9	1	4	9	2	11	4	0	5	8	2
	% within fT4 group	33.3	60.0	6.7	26.7	60.0	13.3	73.3	26.7	0.0	33.3	53.3	13.3
0.7-2.0	Count	57	153	0	64	137	9	174	36	0	41	159	10
	% within fT4 group	27.1	72.9	0.0	30.5	65.2	4.3	82.9	17.1	0.0	19.5	75.7	4.8
≥2.0	Count	21	54	0	36	36	3	53	20	2	7	61	7
	% within fT4 group	28.0	72.0	0.0	48.0	48.0	4.0	70.7	26.7	2.7	9.3	81.3	9.3
Total	Count	83	216	1	104	182	14	238	60	2	53	228	19
	% within fT4 group	27.7	72.0	0.3	34.7	60.7	4.7	79.3	20.0	0.7	17.7	76.0	6.3
Chi-square test; significance		0.001			0.034			0.041			0.044		

In most of the cases (70%), fT4 and PCV were within reference range. Lower MCHC was due to anaemia of the participants, showing dependence on fT4.

Within the reference range of RBC count, a sharp gradual ascending trend was observed with the increment of fT4 but the downgrade trend

was observed when RBC count was lower than the reference range.

In hypothyroid, hyperthyroid and control groups, it was revealed that MCV and MCHC had strong statistical relationships (p-values 0.01 and 0.001, respectively).

Table 5: Spearman's Correlation between different attributes

		TSH	fT4	Hb	PCV	MCV	MCHC	MCH	RDW-CV	RBC count
TSH	rho	1.000	-0.524	0.036	0.038	0.211	-0.059	0.185	0.036	-0.150
	Sig.(2-tailed)		<0.001	0.538	0.514	<0.001	0.309	0.001	0.538	0.009
fT4	rho	-0.524	1.000	-0.048	-0.032	-0.091	0.053	-0.105	0.037	0.067
	Sig.(2-tailed)	<0.001		0.407	0.580	0.115	0.364	0.071	0.518	0.247
Hb	rho	0.036	-0.048	1.000	0.904	0.399	0.476	0.492	-0.399	0.346
	Sig.(2-tailed)	0.538	0.407		<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
PCV	rho	0.038	-0.032	0.904	1.000	0.374	0.217	0.382	-0.403	0.415
	Sig.(2-tailed)	0.514	0.580	<0.001		<0.001	<0.001	<0.001	<0.001	<0.001
MCV	rho	0.211	-0.091	0.399	0.374	1.000	0.122	0.884	-0.416	-0.499
	Sig.(2-tailed)	<0.001	0.115	<0.001	<0.001		0.035	<0.001	<0.001	<0.001
MCHC	rho	-0.059	0.053	0.476	0.217	0.122	1.000	0.339	-0.228	0.092
	Sig.(2-tailed)	0.309	0.364	<0.001	<0.001	0.035		<0.001	<0.001	0.112

MCH	rho	0.185	-0.105	0.492	0.382	0.884	0.339	1.000	-0.384	-0.426
	Sig.(2-tailed)	0.001	0.071	<0.001	<0.001	<0.001	<0.001		<0.001	<0.001
RDW-CV	rho	0.036	0.037	-0.399	-0.403	-0.416	-0.228	-0.384	1.000	0.069
	Sig.(2-tailed)	0.538	0.518	<0.001	<0.001	<0.001	<0.001	<0.001		0.234
RBC count	rho	-0.150	0.067	0.346	0.415	-0.499	0.092	-0.426	0.069	1.000
	Sig.(2-tailed)	0.009	0.247	<0.001	<0.001	<0.001	0.112	<0.001	0.234	

TSH was in positive significant correlation with MCV, MCH and negative significant correlation with RBC count. Hence, it could be concluded that in megaloblastic anaemia TSH might be found to be elevated; in other words, hypothyroidism might be a risk factor for developing megaloblastic anaemia. So, in patients with anaemia of unknown origin, especially those with increased MCV, ypothyroidism should be considered in the differential diagnosis.

Discussion

Thyroid disorders are frequently associated with Haematological abnormalities, among which anaemia is most prevalent [5]. Females are more prone to the develop thyroid disorders and/or anaemia. Anaemia in hypothyroidism leads to physiological compensation for the diminished need of tissues for oxygen.

The low plasma erythropoietin is found in hypothyroid-anaemia. MCV increases rapidly in association with evolving hypothyroidism. On replacement therapy with thyroxine, MCV was found to be reduced progressively. The cause of the increase in size of the red cells and of the minor degree of anisocytosis in uncomplicated hypothyroidism is unknown. In a study by Geetha J and Srikrishna R in 2012, revealed that RDW and MCV in patients with hypothyroidism and hyperthyroidism in comparison to euthyroid individuals have statistically significant difference but other RBC parameters like Hb and PCV did not show any significant difference in comparison with control group [18]. Kawa MP and et al in 2010 reported that MCH and MCHC were lower but PCV and MCV were increased in both hypothyroidism and hyperthyroidism in comparison with control group [19]. Lima et al. concluded that thyroid evaluation should be performed for all pancytopenia regardless of presence of symptoms [20]. Dorgalaleh et al. showed that Hb%, PCV, RDW were statistically different in hypothyroid, hyperthyroid and control groups but not for MCV [21].

The present study showed increased MCV, MCH and decreased RBC count in untreated hypothyroid as compared to euthyroid. Hypothyroidism should always therefore be considered as a possible cause of unexpected and unexplained anaemia.

Conclusion

Thyroid hormones (T_3 and T_4) have a significant influence on erythropoiesis. In our study, it was found that haematological parameters like RBC count were decreased, whereas MCV was increased in untreated hypothyroidism. Abnormal levels of thyroid hormones might substantially influence the size variability of circulating RBC. While investigating causes of anaemia, it is important to rule out hypothyroidism. It is suggested that prompt and early recognition and treatment of hypothyroidism is needed to prevent risk of developing anaemia.

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