

Thyroid status and its correlation with variations in metabolic parameters leading to other diseased condition

Navreet Kaur^{1*}, Sukhraj Kaur², Jaspinder Kaur³, Mridula Mahajan⁴

^{1, 2, 3, 4}Department of Biochemistry, Government Medical College, Amritsar- 143001, Punjab, INDIA.

Email: drnavreetsidhu@gmail.com

Abstract

Objective: Thyroid dysfunction, Diabetes Mellitus (DM) and cardiovascular diseases (CVD) are co-existent disorders, but there is less information on the correlation of DM and CVD with different stages of thyroid dysfunction. **Design and Methods:** We assessed the thyroid profile (T3, T4 and TSH) of 56 individuals with symptoms of DM and CVD (divided as having normal thyroid function (group1), subclinical (group 3,5) and clinical (group 2,4) hyper and hypothyroidism respectively, based on the levels of TSH). The association of thyroid hormones was investigated with carbohydrate metabolism (levels of fasting serum glucose, insulin, C-peptide, HOMA-IR) and lipid metabolism [total cholesterol, triglycerides (TG's), High density lipoprotein (HDL), Low density lipoprotein (LDL) and Very low density lipoprotein (VLDL)] in different groups. **Results:** As compared to group 1, there was significant decrease in %S ($p<0.001$, $p<0.05$) and significant increase in Insulin resistance (IR) ($p<0.05$) in Group 3 and Group 4 respectively with comparable IR in group 2 and group 4. Levels of triglycerides (TG's) were increased from (154.5 ± 18.31 mg/dl) to (196.0 ± 11.0 mg/dl) and (194.5 ± 12.21 mg/dl) and TG's showed significant positive correlation with IR ($r=+0.775$ $p<0.01$, $r=+0.643$ $p<0.001$) in group 4 and group 5 respectively. Also in group 1, 4 and 5 cholesterol showed significant positive correlation with LDL ($p<0.001$) and VLDL ($p<0.05$). **Conclusions:** Individuals in subclinical stages of thyroid dysfunction are at more risk for IR and its related disorders.

Keywords: Thyroid dysfunction, diabetes mellitus, cardiovascular diseases, insulin resistance

*Address for Correspondence

Dr. Navreet Kaur, Department of Biochemistry, Government Medical College, Amritsar- 143001, Punjab, INDIA.

Email: drnavreetsidhu@gmail.com

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INTRODUCTION

Thyroid dysfunction can be defined as the malfunctioning of thyroid that can result in the over functioning and insufficient functioning of the thyroid. Broadly thyroid dysfunction can be classified as: 1) Clinical or overt hypothyroid 2) sub clinical hypothyroid 3) clinical or overt hyperthyroid 4) sub clinical hyperthyroid. It has been reported that thyroid hormones effect the metabolism of carbohydrate and lipoprotein, leading to diabetes mellitus (DM) and cardiovascular diseases (CVD), the disorders which have been shown to mutually influence each other^{1,2}. On the other hand DM affects

thyroid function to a variable extends³. Of all the classes of thyroid dysfunction subclinical hypothyroidism far more common⁴. Although studies have shown that clinical hypo and hyperthyroidism is accompanied by variety of abnormalities in plasma lipids and glucose homeostasis^{3,5}, but presently there is no clear information regarding relation of subclinical thyroid dysfunction with metabolic parameter in Indian population. As patients with subclinical hypo and hyperthyroidism sometimes have subtle or no symptoms. Unchecked status of clinical condition for longer period may cause adverse clinical conditions keeping in view the prevalence of thyroid dysfunction in individuals with DM and cardiac disorders the present study was planned to find a correlation if any amongst these abnormalities.

MATERIAL AND METHODS

The present study was conducted in the Department of Biochemistry, Govt. Medical College, Amritsar, India. The individuals who reported for the investigation of thyroid profile i.e. T₃, T₄ and TSH and having symptoms of cardiac disorder (hypertension, tachycardia) and diabetes mellitus were selected for the present study. An

informed consent was taken from every patient. The project was approved by ethical committee of the institute. On the basis of the levels of TSH, according to Indian thyroid Society^{6,7} 56 individuals under study were divided into 5 groups

Group 1 (n= 21.0%): Normal TSH (0.44- 3.46 μ IU/ml)

Group 2 (n= 14.0%): Clinical Hyperthyroidism (TSH < 0.1 μ IU/ml)

Group 3 (n= 17.0%): Subclinical hyperthyroidism (TSH range 0.1- 0.44 μ IU/ml)

Group 4 (n= 19.5%): Clinical Hypothyroidism (TSH > 10 μ IU/ml)

Group 5 (28.5%): Subclinical Hypothyroidism (TSH range 3.46- 10 μ IU/ml)

Fasting blood samples collected were allowed to clot and serum was separated for various investigations. In vitro quantitative determination of hormones- T_3 ⁸, T_4 ⁹ and TSH¹⁰ was carried out by using direct solid phase enzyme immunoassay based ERBA thyrokit, on Erba Mannheim LISA scan. Blood glucose and its homeostatic parameters (Insulin, C- peptide) and lipid profile were measured on autoanalyzer- (Erba XL 300). Blood glucose was estimated by oxidase- peroxidase method as described by Trinder P (1969)¹¹. Insulin and C- peptide was estimated by method based on direct solid phase enzyme immunoassay as described by Boehm TM 1979¹² and Kuzuya H 1977¹³ respectively; and the test was

performed by using commercially available kits from Dia- metra, (Italy). Total cholesterol was estimated by enzymatic method as described by Charles CA (1974)¹⁴ using kits from Biosystems, (SA Costa Brava, 30- Barcelona Spain). HDL-C was estimated by method described by Brustein [1970]¹⁵ using kit from Transasia Biomedicals Ltd. Serum Triglycerides (TG) were estimated by Trinder's method as described by Mc Gowan (1983)¹⁶ using kits from Transasia Biomedicals (Daman). Serum LDL was determined by using Friedwald's and Fredrickson's formula¹⁷ i.e.

$LDL = \text{Total Cholesterol} - (\text{HDL} + \text{VLDL})$.

VLDL was estimated by using: $VLDL = \text{Triglycerides} / 5$ based on the average ratio of TG to Cholesterol in VLDL. Insulin resistance (IR), Insulin Sensitivity (%S) and β -cell function (%B) were estimated by the HOMA-IR (Matthews 1985)¹⁸.

Statistical Analysis

The data collected was analysed using computer software SPSS (version 16.0). Student's t- test was applied to study the variation between two groups. Comparison of means of different groups was done using one way ANOVA (SPSS 16.0). Pearson's coefficient of correlation was calculated to study the significance of correlation between different parameters. Level of significance used was $p < 0.05$.

Table 1: Levels of T3, T4 and TSH in the groups involved in the study

Groups	T3 (ng/ml)	T4 (μ g/ml)	TSH (μ IU/ml)
Group 1 Normal	0.94 \pm 0.50	10.13 \pm 2.01	1.40 \pm 0.53
Group 2 Hyper	2.19 \pm 0.21	16.46 \pm 3.68**	0.06 \pm 0.39***
Group 3 Sub hyper	0.89 \pm 0.27	9.23 \pm 1.50	0.23 \pm 0.09***
Group 4 Hypo	0.01 \pm 0.02	2.23 \pm 0.99	13.27 \pm 1.23**
Group 5 Sub hypo	0.9 \pm 0.35	10.32 \pm 2.93	5.19 \pm 0.22***

* $p < 0.05$ ** $p < 0.01$ *** $p < 0.001$

Group 2 – T3 with T4 $r = +0.839$ $p < 0.05$

Group 4 – T3 with insulin $r = +0.715$ $p < 0.05$, T3 with C-peptide $r = +0.778$ $p < 0.05$, T4 with IR $r = +0.746$

$p < 0.05$, TSH with insulin $r = +0.717$ $p < 0.05$, TSH with C-peptide $r = +0.737$ $p < 0.05$

Group 5 – T4 with C-peptide $r = -0.591$ $p < 0.01$

Table 2: Levels of Glucose, Insulin and C-peptide in the study groups

Groups	Glucose (mg/dl)	Insulin (μ IU/ml)	C-peptide (ng/ml)
Group 1 Normal	101.03 \pm 13.93	8.01 \pm 0.81	4.50 \pm 0.04
Group 2 Hyper	80.33 \pm 10.52	5.90 \pm 0.80	2.23 \pm 0.87*
Group 3 Sub hyper	101.26 \pm 27.78	30.30 \pm 1.70**	2.49 \pm 0.20
Group 4	128.88 \pm 9.17	5.3 \pm 0.59	3.69 \pm 0.48

Hypo			
Group 5	152.25 ± 12.21	14.24 ± 2.35	7.22 ± 1.02
Sub hypo			
*p<0.05 **p<0.01 ***p<0.001			
Group 5 – Insulin with VLDL r=+0.613 p<0.01, Insulin with TG r=+0.613 p<0.01			

Table 3: Lipids and lipoproteins of the groups involved in the study

Groups	Total Cholesterol (mg/dl)	Triglycerides (mg/dl)	HDL- Cholesterol (mg/dl)	LDL (mg/dl)	VLDL (mg/dl)
Group 1 Normal	196.70±5.84	154.5±18.31	44.5±3.26	121.35±10.50	30.9±1.16
Group 2 Hyper	183.33±31.11	126.33±30.51	45.83±5.19	112.23±28.29	25.26±1.21
Group 3 Sub hyper	179.0±6.68	152.8±10.25	41.4±1.67	107.04±5.79	30.56±2.05
Group 4 Hypo	226.0±17.8	196.0±11.0	52.23±5.84	134.56±15.46	39.2±2.20
Group 5 Sub hypo	203.06±51.5	194.5±12.21	47.68±6.15	116.47±21.84	38.9±2.44

Group1- Cholesterol with LDL r= +0.995 p<0.001, Cholesterol with VLDL r=+0.874 p<0.05, Cholesterol with TG r=+0.838 p<0.001

Group2 - TG with LDL r=+0.833 p<0.05, TG with VLDL r=+1.0 p<0.001

Group 3 – Cholesterol with LDL r=+0.959 p<0.01, VLDL with TG r=+1.0 p<0.001

Group 4 – Cholesterol with LDL r=+0.967 p<0.001, Cholesterol with VLDL r=+0.666 p<0.05, Cholesterol with TG r=0.666 p<0.05

Group 5- Cholesterol with LDL r=+0.859 p<0.001, Cholesterol with VLDL r=+0.541 p<0.05, Cholesterol with TG r=+0.541 p<0.05

Table 4: HOMA-IR of the groups involved in the study

Groups	% B	% S	IR
Group 1 Normal	180.65±18.51	104.47±8.06	2.01±0.28
Group 2 Hyper	185.11±7.78	86.98±6.17	1.95±0.14
Group 3 Sub hyper	282.8±2.93	35.28±2.31**	3.72±0.86*
Group 4 Hypo	108.23±18.04	106.7±6.70	1.34±0.82
Group 5 Sub hypo	111.03±12.68	68.98±7.53*	3.63±0.61*

*p<0.05 **p<0.01 ***p<0.001

Group 2 – T4 with %S r= - 0.820 p<0.01, TSH with IR r=+0.906 p<0.05

Group 4 - %S with IR r= - 0.869 p<0.001, VLDL with IR r=+0.775 p<0.01, TG with IR r=+0.775 p<0.01, IR with T4 r=+0.746 p<0.05

Group 3 – IR with Insulin r=+0.962 p<0.001, %S with IR r= - 0.916 p<0.05

Group 5 - %B with VLDL r=+0.662 p<0.001, %B with TG r=+0.662 p<0.001, IR with VLDL r=+0.643 p<0.001, IR with TG r=+0.643 p<0.001

RESULTS

In group 1 individuals a significant correlation was observed between insulin and c- peptide, cholesterol was positively correlated with LDL, VLDL and TG (p < 0.002) (table 2, 5, 4). % of beta cell function was positively correlated with IR and % sensitivity of insulin in these individuals (thus indicating normal function of the thyroid gland in this group and its impact on various metabolisms). When compared with normal reference range all the biochemical parameters examined in this study group were normal except an increase in C-peptide levels, TG and IR. In group 2 individuals the levels of T4

were positively correlated with T3 and sensitivity of insulin thereby decreasing resistance to the insulin action (r = 0.906, p < 0.013). In these patients cholesterol had a positive correlation with LDL, VLDL and triglycerides (r = 0.099, r = 0.87, r = 0.87, p < 0.001, p < 0.02, p < 0.02 respectively). Thus indicates that an increase in the levels of T4 has an effect on the levels of cholesterol (Table 3). The levels of TSH in group 3 individuals were significantly below te normal levels as compared to group 1but have normal T3 and T4 levels. No significant correlation was observed between these hormonal levels. % S and IR found to be significantly decreased and

increased respectively and there is negative correlation of insulin with % S and positive with IR ($r = -0.852$ and $r = +0.962$ respectively). In these patients cholesterol also had a positive correlation with LDL ($r = +0.959$, $p < 0.01$). VLDL and triglycerides are also positively correlated with each other ($p < 0.01$). HDL-C levels reduced significantly ($p < 0.05$) as compared to group 1 (Table 3). The levels of insulin were significantly more in this group as compared to group 1 and group 2 patients, however it showed a negative (non-significant) correlation with T3, T4, TSH. Although the individuals having subclinical hyperthyroidism had insulin resistance, yet insulin did not show any significant correlation with c-peptide and it was negatively correlated with % S and had a direct relation with IR ($r = -0.852$, $p < 0.06$ and $r = +0.962$, $p < 0.001$). This group with subclinical hyperthyroid individuals showed significant ($p < 0.05$) increase in IR and significant ($p < 0.01$) decrease in %S. Beta cell function had a positive correlation with insulin ($r = 0.151$, $p < 0.14$) and a negative correlation with lipid profile. In group 4 individuals, TSH was significantly increased as compared to group 1, group 2 and group 3 individuals and having T3, T4 levels below the normal range. The levels of T3 and TSH had a significant positive correlation with insulin and c-peptide ($r = +0.715$, 0.778 and $p < 0.03$, $p < 0.01$ respectively) (Table 1). Thus indicating that more levels of TSH leads to more synthesis and secretion of insulin as indicated by the levels of insulin and c-peptide. The levels of T3 did not showed any significant correlation with IR. On the contrary T4 was positively correlated with IR ($r = -0.746$, $p < 0.02$). The levels of total cholesterol were more (non significantly) in these individuals as compared to group 1, group 2 and group 3 individuals. The increased levels of cholesterol had a significant positive correlation with LDL, VLDL and triglycerides ($r = 0.967$, 0.666 , 0.666 and $p < 0.001$, $p < 0.05$, $p < 0.05$ respectively) (Table 3). The levels of lipid profile (total cholesterol, HDL, LDL, VLDL and TG's) were maximum in this group as compared to other groups. Beta cell function was decreased as compared to group 1 and group 2 with no change in sensitivity to insulin and IR. TSH levels in group 5 were significantly more than group 1 individuals but less than individuals in group 4. Levels of insulin and c-peptide depict that synthesis and secretion of insulin was there but the variation was not significant. Insulin showed a positive correlation with VLDL and Triglycerides ($r = +0.613$, $p < 0.012$) (Table 2) and C-peptide showed a negative correlation with T4 ($r = -0.591$, $p < 0.016$) (table 1). Total lipid profile although was less than group 4 individuals, yet did not show any significant variation as compared to group 1. Moreover

IR was significantly more prevalent in this group as indicated by decreased sensitivity to insulin.

DISCUSSION

It has been shown by previous studies that clinical hypothyroidism is closely associated with CVD and DM, but less work has been done on clinical hyperthyroidism and the relationship of subclinical stages of thyroid dysfunction with variations in metabolic parameters. Our results demonstrate that the individuals who reported with symptoms of Diabetes Mellitus, hypertension and tachycardia but having normal thyroid hormone levels showed normal levels of glucose and insulin except the C-peptide levels which were above the normal range. C-peptide has been suggested to be a mediator of atherosclerosis in diabetes, so the increased C-peptide levels may be a marker of initiation of atherosclerotic events in these patients¹⁹. With all the other parameters of lipid profile being normal the increase of TG in these individuals (group 1) suggest that may be the thyroid hormones play no role in changing the TG levels. On the other hand individuals with increased T3, T4 levels and decreased TSH levels (group 2) showed normal glucose homeostasis and normal lipid profile. The IR is comparable to group 1, although it showed two fold increase compared to standard reference values. This shows that IR may not be majorly influenced by thyroid hormones in clinical hyperthyroidism. Interestingly the subclinical hyperthyroid individuals (group 3) showed normal glucose, C-peptide and lipid profile levels but a significant increase in insulin and insulin resistance with significant decrease in insulin sensitivity. However the IR observed in this group does not seem to be clinically relevant in terms of significant hyperglycaemia, possibly due to compensatory decrease in hepatic glucose output as a result of hyperinsulinemia²⁰. In hypothyroid state (group 4) there is an increase in glucose, C-peptide, TG's, LDL, VLDL (non significantly) as compared to group 1, with C-peptide levels 2-fold higher than the normal reference range and the HOMA-index is found to be normal. The increase in glucose and lipid profile in this group may be due to altered oxidative and non-oxidative glucose metabolism²¹. Another interesting thing revealed in this study is that the individuals with subclinical hypothyroidism (group 4) have increased levels of glucose, insulin, C-peptide, TG's, VLDL and a significant rise in IR with significant decrease in insulin sensitivity as compared to group 1. Increased TG's and VLDL in this group (altered lipid metabolism) contribute to insulin resistance²². In intergroup comparisons, the levels of insulin and insulin resistance were higher in subclinical stages of both hypo as well as hyperthyroidism whereas it was comparable in normal thyroid and clinical stages of hypo and hyperthyroidism. It suggests that the individuals

in subclinical stages of thyroid disorders are at high risk of insulin resistance associated disorders such as DM and CVD and the increased IR in these stages may be due to abnormal hemostatic status and derailed immune system²³. In group 1 even though the thyroid profile was within normal limits yet these individuals exhibited a degree of insulin resistance; whereas in group3 and group5 having subclinical thyroid dysfunction were having significant IR when compared to other groups. This shows that IR is more in subclinical stages (with normal T3,T4) and this IR may cause T3, T4 imbalance which leads to clinical stages of thyroid dysfunction. Our study shows that TSH is negatively correlated with glucose and insulin in subclinical stages of hypo and hyperthyroidism and positively correlated with glucose and insulin in clinical stages of hypo and hyperthyroidism. From the above study it can be hypothesized that insulin resistance (hyperinsulinemia) leads to hormonal imbalance which is depicted as thyroid dysfunction and TSH may play a major role in affecting IR in subclinical stages of thyroid dysfunction. However a large group study can explore further the various underlying mechanisms.

CONCLUSION

Even subtle increase or decrease in the TSH levels (subclinical stages) can lead to IR. Therefore it can be suggested that the patients with mild thyroid failure should be treated early so that they may not acquire IR and its related disorders. Thus averting their progress to clinical hypo and hyperthyroidism.

REFERENCES

1. Feely J, Isles TE. Screening for thyroid dysfunction in diabetes: British Medical Journal. 1978; 1: 1678.
2. Gray RS, Irvins WJ, Clarke BF. Screening for thyroid dysfunction in diabetes: British Medical Journal. 1979; 2: 1439.
3. Hage M, Zantout MS, Azar ST. Thyroid disorders and diabetes mellitus: Journal of thyroid research. 2011; 2011: article ID 439463. <http://dx.doi.org/10.4061/2011/439463>.
4. Patricia Wu. Thyroid disease and diabetes: Clinical Diabetes. 2000; 18.
5. Rizor CV, Elisaf MS, Liberopoulor EN. Effects of thyroid dysfunction on lipid profile: The open cardiovascular medicine journal. 2011; 5: 76- 84.
6. Agarwal NK, Unnikrishnan AG, Singh SK. Subclinical hypothyroidism: An Update. Indianthyroidsociety.com
7. Donangelo I, Brawnstein GD. Update on subclinical hyperthyroidism: American Family Physician. 2011; 83: 933- 938.
8. Ingbar SH, Beaverman LE. Active form of the thyroid hormone: Annual Review of Medicine. 1975; 26: 443-449.
9. Attwood EC, Seddon RM, Probert DE. The T₄/ TBG ratio and the investigation of thyroid function: Clinical Biochemistry. 1978; 11: 218.
10. Bristow AF, Sutcliffe N, Ayling C, Bangham DR. Evaluation of candidate materials for replacement of International Reference preparation of Human thyroid stimulating hormone (TSH) for immunoassay. In hormone drugs, proceeding of FDA- USP workshop on drug and reference standards for insulins, somatropins and thyroid axis hormones. Bethesda MD. The US: pharmacopoeial convention, Inc. Rockville MD 524- 533.
11. Trinder P. Determination of blood glucose using an oxidase- peroxidase system with a non- carcinogenic chromogen: Journal of Clinical Pathology. 1969; 22: 158- 561.
12. Boehm TM, Lebovitz HE. Statistical analysis of glucose and insulin responses to intravenous tolbutamide. Evaluation of hypoglycemic and hyperglycemic and hyperinsulinemic states: Diabetes care. 1979; 2: 479-490.
13. Kuzuya H, Blix PM, Horwitz DL, Donald F Steiner, Arthur H Rubenstein. Determination of free and total insulin and c- peptide in insulin treated diabetes: Diabetes. 1977; 26: 22.
14. Charles CA, Lusysp, Cicelys. Enzymatic determination of total serum cholesterol: Clinical Chemistry. 1974; 20: 470- 475.
15. Brustein M, Scholnick HR, Morfin R. Rapid method for isolation of lipoprotein from human serum by precipitation with polyanions: Journal of lipid research. 1970; 11: 483- 495
16. Gowan Mc, Michael W, Joseph D. A peroxidase coupled method for the colorimetric determination of serum triglycerides: Clinical Chemistry. 1983; 29: 538-542.
17. William TF, Robert IL, Donald SF. Estimation of concentration of low density lipoprotein cholesterol in plasma without the use of preparative ultracentrifuge: Clinical Chemistry. 1972; 18: 499-502.
18. Matthews DR, Hosker JP, Rudenski AS. Homeostasis model assessment: Insulin resistance and β - cell function from fasting plasma glucose and insulin concentration in man: Diabetologia. 1985; 28: 412- 419.
19. Dusicavasic , Daniel W. C- peptide: A new mediator of atherosclerosis in diabetes mellitus: Journal meidators of Inflammation. <http://dx.doi.org/10.115/2012/858592>
20. Eirini Maratou, Dimitrios J Hadjidakis, Melpomeni Peppas, Maria Alevizaki, Katerina Tsegka, Vaia Lambadiari, Panayota Mitrou, Eleni Boutati, Anastasios Kollias, Theofanis Economopoulos, Sotirios A Raptis, George Dimitriadis. Studies of insulin resistance in patients with clinical and subclinical hypothyroidism: European Journal of Endocrinology. 2009; 160: 785- 790.
21. Potenza M, Via MA , Yanagisawa RT. Excess thyroid hormone and carbohydrate metabolism: Endocrine Practice. 2009; 15: 254-262.
22. Kraegen EW, Cooney GJ, Ye J, Thompson AL. Triglycerides, Fatty acids and Insulin resistance- hyperinsulinemia: Experimental and Clinical Endocrinology and Diabetes. 2001; 109: S 516-526.
23. Franchini M. Hemostatic changes in thyroid diseases: Haemostasis and thrombosis: Hematology. 2006; 11: 203- 208.

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