Acta Biomed 2014; Vol. 85, N. 2: 127-134

Original article

Serum lipids, tHcy, hs-CRP, MDA and PON-1 levels in SCH and overt hypothyroidism: effect of treatment

Sarika Singh[1], *Purmina Dey Sarkar*[2]

[1]Nscb Medical College Jabalpur MP and [2]Medical Biochemistry MGM Medical College Indore MP, India

Summary. *Background.* Thyroid hormones are the most important factors involved in the regulation of the basal metabolic condition, as well as in the oxidative metabolism. Hypothyroidism is associated with increased cardiovascular morbidity, which cannot be fully explained by the atherogenic lipid profile observed in these patients. *Aim:* Study of serum lipids, tHcy, hs-CRP, MDA and PON-1 levels in SCH and overt Hypothyroidism: Before and After Therapy" *Methods:* 100 hypothyroid (56 SCH and 44 OH) and 100 controls, with a mean age range 20–65 years were enrolled for this study. Serum TSH, FT3 and FT4 levels were evaluated by Chemiluminescence Enzyme Immunoassay. T-CHO by CHOD – POD Method. HDL-C by Direct Detergent Method. TG by GPO-POD Method. LDL-C by Friedewald's Formula. tHcy by auto analyzers. hs-CRP by Latex–High Sensitivity. MDA by Kei Satoh Method. PON-1 by spectrophotometric method using P-Nitrophenyl Acetate. All tests were done before and after 8-12 week of treatment. *Result:* Pretreatment, in both SCH and OH groups, we found significantly higher serum TSH (P<0.001), TC, LDL-C, TG (P<0.01, P<0.001 and P<0.05) in SCH and (P<0.001) in OH, hs-CRP (P<0.05 and P<0.001) and MDA levels (P<0.001) than in controls. A significant decrease was seen in FT3 and FT4 (P<0.05 and P<0.001) and PON-1 activity (p<0.01 and p<0.001) in both groups than in controls, whereas HDL levels were lower and Homocysteine levels were found to be higher in only OH groups. L-thyroxine therapy significantly decreased serum TSH, TC, LDL-C, TG and t-Hcy (P<0.001) concentrations in both groups. *Conclusion:* Our study showed that both SCH and OH patients have elevated atherogenic and oxidative stress markers. Increased CRP and homocysteine might be a key molecule linking inflammation to oxidative stress in atherosclerosis.

Key words: Subclinical hypothyroidism, overet hypothyroidism, lipid, CRP, homocysteine, PON-1

Introduction

Hormones serve within the body as invaluable messengers, governors of development, and regulators of metabolism (1). Thyroid gland secretes thyroxine (T4) & triiodothyronine (T3) hormones which influence metabolic rate, increases synthesis & degradation of cholesterol & triglycerides (2). Thyroid hormones are associated to the oxidative and antioxidative status of the organism. Depression of metabolism due to hypothyroidism has been reported to decrease oxidant production and thus protects tissues against oxidant damage (3). It is estimated that more than 200 million people at minimum worldwide have thyroid disease (4). The prevalence of thyroid dysfunction in adults in the general population ranges from 1 to 10 % (5). Thyroid disease prevalence increases with age and is more common in women. The prevalence of hypothyroidism has varies from 0.9 to 17.5% (6,7). The prevalence of subclinical thyroid dysfunction is higher than that of overt thyroid dysfunction (6).

Dyslipidemia is a common finding in patients with thyroid disease, explained by the adverse effects of thyroid hormones in almost all steps of lipid metabo-

lism. Not only overt but also subclinical hypothyroidism, through different mechanisms, are associated with lipid alterations, mainly concerning total and LDL cholesterol and less often HDL cholesterol, triglycerides, lipoprotein (a), apolipoprotein A1, and apolipoprotein B. In addition to quantitative, qualitative alterations of lipids have been also reported, including atherogenic and oxidized LDL and HDL particles. In thyroid disease, dyslipidemia coexists with various metabolic abnormalities and induce insulin resistance and oxidative stress via a vice-vicious cycle. Hypothyroidism is the second leading cause of high cholesterol, after diet. When TH levels drop, the liver no longer functions properly and produces excess cholesterol, fatty acids, and triglycerides, which increase the risk of heart disease (8).

Hypothyroidism is associated with increased cardiovascular morbidity, which cannot be fully explained by the atherogenic lipid profile observed in these patients. Elevated C-reactive protein and plasma total homocysteine (tHcy) were recently identified as risk factors for cardiovascular disease (9). High-sensitive C-reactive protein (hs-CRP) levels, a marker of subclinical inflammation, have been identified as an independent predictor of atherosclerosis. Although it is well known that thyroid hormones play an important role in cardiovascular hemodynamics, the association between elevated thyroid hormones and low grade inflammation is still unclear (10). Elevated tHcy levels have been reported in overt hypothyroidism, and have been proposed as independent risk factors for cardiovascular disease. Whether SCH is a risk factor for premature cardiovascular disease is controversial (11).

Oxidative stress, characterized by an elevation in the steady-state concentration of reactive oxygen species, has been involved in a wide range of biological and pathological conditions. However, data on the oxidative status of hypothyroidism are limited and controversial (12). Thyroid hormones have well-known effects on mitochondrial oxygen consumption, but data about how hypothyroidism affects oxidative stress are controversial, and little is known about oxidative stress in subclinical hypothyroidism. Malondialdehyde (MDA) is a lipid peroxidation marker used to assess lipid peroxidation due to increased oxidative stress (13). Antioxidant effects of Paraoxonase, (HDL-associated enzyme) that inhibits LDL-C oxidation in human serum, have also been reported (14).

Table 1. Various biochemical parameters in subclinical hypothyroid patients before and after therapy compared to controls

		MEAN±S.D.			t-test		
		Subclinical Hypothyroid					
SN	Parameters	Control (A) (n = 100)	Before Therapy (B) (n = 56)	After 8-12 Weeks of Therapy (C) (n = 56)	A-B	A-C	B-C
1	FT^3 (pg/ml)	2.91±0.61	2.67±0.61	2.78±0.55	P≤0.05	NS	P≤0.01
2	FT^4 (ng/dl)	1.39±0.21	1.13±0.19	1.28±0.17	P≤0.001	P≤0.001	P≤0.001
3	TSH (μIU/ml)	2.22±0.78	13.06±4.02	3.48±1.56	P≤0.001	P≤0.001	P≤0.001
4	T-CHO (mg/dl)	184.97±22.50	202.13±37.64	193.86±30.30	P≤0.01	P≤0.05	P≤0.01
5	LDL-C (mg/dl)	113.38±17.98	129.46±34.69	121.96±28.84	P≤0.001	P≤0.05	P≤0.01
6	HDL-C (mg/dl)	50.81±6.07	48.53±7.76	49.36±7.18	NS	NS	P≤0.05
7	TG (mg/dl)	103.86±38.25	120.68±47.66	112.69±37.71	P≤0.05	NS	P≤0.01
8	Hcy (μmol/L)	8.60±1.64	9.14±1.80	9.10±1.68	NS	NS	NS
9	hs-CRP (mg/L)	1.92±0.50	2.37±1.35	2.30±1.15	P≤0.05	P≤0.05	NS
10	MDA (μmol/L)	0.99±0.10	1.60±0.37	1.25±0.35	P≤0.001	P≤0.001	P≤0.001
11	PON-1 (KU/L)	51.61±10.12	45.79±12.48	48.94±11.07	P≤0.01	NS	P≤0.001

Materials and methods

Design of study

Present thesis titled "Study of serum lipids, tHcy, hs-CRP, MDA and PON-1 levels in SCH and overt Hypothyroidism: Before and After Therapy," has been conducted in Department of Biochemistry, NSCB Medical College, Jabalpur, MP, India, from January 2010 to January 2013.

A. *Patient selection:* For this study newly diagnosed hypothyroid patients (According to WHO criteria) were selected with prior permission of ethical committee. Patients were taken from outpatient department. Written informed consent was taken from all patients.

B. *Sample size:* Newly detected 100 hypothyroid and 100 euthyroid controls were enrolled for this study, whose pre & post-treatment (irrespective of therapy) values were compared.

C. *Design of the study:*

Inclusion criteria:

Recently detected or poorly controlled hypothyroid patients.

Exclusion criteria:

- Patients with pre-existing renal and cardiac abnormality.
- Patients having Primary dyslipidemia.

C. *Age eligible for study:* 20 years to 65 years.

D. *Study was carried out under two groups:*

- Control vs. Patients
- Hypothyroid patients were divided in two groups according to TSH levels as:

Group I (Subclinical Hypothyroid): TSH level 6-20 μIU/ml

Group II (Overt Hypothyroid): TSH level more than 20 μIU/ml

Methods

- Serum TSH, FT3 and FT4 levels were evaluated by Chemiluminescence Enzyme Immunoassay (CLIA).
- Serum T-CHO levels were measured by CHOD – POD Method.
- Serum HDL-C levels were measured by Direct Detergent Method.
- Serum TG levels were measured by GPO-POD Method.

Table 2. Various biochemical parameters in overt hypothyroid patients before and after therapy compared to controls:

		MEAN±S.D.			t-test		
		Subclinical Hypothyroid					
SN	Parameters	Control (A) (n = 100)	Before Therapy (B) (n = 44)	After 8-12 Weeks of Therapy (C) (n = 44)	A-B	A-C	B-C
1	FT^3 (pg/ml)	2.91±0.61	2.05±0.46	2.36±0.41	P≤0.001	P≤0.001	P≤0.001
2	FT^4 (ng/dl)	1.39±0.21	0.67±0.25	1.15±0.27	P≤0.001	P≤0.001	P≤0.001
3	TSH (uIU/ml)	2.22±0.78	47.66±18.87	3.69±1.03	P≤0.001	P≤0.001	P≤0.001
4	T-CHO (mg/dl)	184.97±22.50	234.62±33.89	197.67±25.66	P≤0.001	P≤0.01	P≤0.001
5	LDL-C (mg/dl)	113.38±17.98	154.87±33.91	122.57±27.50	P≤0.001	P≤0.05	P≤0.001
6	HDL-C (mg/dl)	50.81±6.07	47.65±7.90	48.80±7.52	P≤0.05	NS	P≤0.05
7	TG (mg/dl)	103.86±38.25	160.44±44.67	131.48±31.13	P≤0.001	P≤0.001	P≤0.001
8	Hcy (μmol/L)	8.60±1.64	10.37±2.69	9.30±1.95	P≤0.001	P≤0.05	P≤0.001
9	hs-CRP (mg/L)	1.92±0.50	2.65±1.44	2.30±1.24	P≤0.01	P≤0.05	P≤0.001
10	MDA (μmol/L)	0.99±0.10	2.16±0.58	1.85±0.57	P≤0.001	P≤0.001	P≤0.001
11	PON-1 (KU/L)	51.61±10.12	39.88±14.12	44.51±13.48	P≤0.001	P≤0.01	P≤0.001

- Serum LDL-C levels were measured by Friedewald's Formula.
- Serum tHcy levels were measured by auto analyzers.
- Serum hs-CRP levels were measured by Latex – High Sensitivity.
- Serum MDA levels were measured by Kei Satoh Method.
- Serum PON-1 activity was estimated by spectrophotometric method using P-Nitrophenyl Acetate as the substrate.

Statistical methods

Results are expressed as mean ± SD range values. Unpaired 't' test is used for comparing different biochemical parameters between cases and controls and paired 't' test is used for comparison of parameters before and after treatments. Pearson's correlation coefficient was used to assess the relationship between different variables. All calculation were done by SPSS 17 software. A P value of <0.05 was considered slightly significant. A P value of < 0.001 was considered highly significant. All the parameters were compared with TSH levels.

Result

The present study was conducted in Department of Biochemistry, NSCB medical college, Jabalpur, MP, India, from January 2011 to Jan 2013. In the current study, out of 100 hypothyroid patients, 56 % were SCH and 44% were overt hypothyroid patients. In subclinical hypothyroid group 30% were males whereas 70% were females. In overt hypothyroid group 23% were males whereas 77% were females. In SCH group the mean age was 45.75 ± 12.56, in overt hypothyroid group the mean age was 44.27 ±12.51, whereas in control group the mean age was found to be 42.92 ±11.60.

Pretreatment, serum FT3 and FT4 levels were found to be decreased significantly (P<0.05 and P<0.001 respectively), whereas serum TSH was found to be increased significantly (P<0.001) in both SCH and overt hypothyroid patients as compared to controls.

A significant changes were seen in these levels by L-thyroxine therapy.

Serum T-cholesterol, LDL-C and TG levels were significantly higher (P<0.01) (P<0.001) (P<0.05 respectively) in SCH and (P<0.001 for all comparison) in OH patients, whereas no significant changes were seen in HDL-C in SCH patients while these levels

Table 3. Comparison Of Various Biochemical Parameters In Sch And Overt Hypothyroid Patients

		MEAN±S.D.		t-test
SN	Parameters	Subclinical Hypothyroid (A) (n = 56)	Overt Hypothyroid (B) (n = 44)	A-B
1	FT3 (pg/ml)	2.67±0.61	2.05±0.46	P≤0.001
2	FT4 (ng/dl)	1.13±0.19	0.67±0.25	P≤0.001
3	TSH (uIU/ml)	13.06±4.02	47.66±18.87	P≤0.001
4	T-CHO (mg/dl)	202.13±37.64	234.62±33.89	P≤0.001
5	LDL-C (mg/dl)	129.46±34.69	154.87±33.91	P≤0.001
6	HDL-C (mg/dl)	48.53±7.76	47.65±7.90	NS
7	TG (mg/dl)	120.68±47.66	160.44±44.67	P≤0.001
8	Hcy (µmol/L)	9.14±1.80	10.37±2.69	P≤0.01
9	CRP (mg/L)	2.37±1.35	2.65±1.44	NS
10	MDA (µmol/L)	1.60±0.37	2.16±0.58	P≤0.001
11	PON (KU/L)	45.79±12.48	39.88±14.12	P≤0.05

were found to be decreased significantly ($p<0.05$) in OH patients as compared to controls.

L-thyroxine therapy significantly decreased serum TSH, TC, LDL-C and TG ($P<0.001$) concentrations in both groups.

Serum CRP levels were found to be significantly higher in both SCH (($p<0.05$) and in OH cases ($p<0.01$), whereas Homocysteine levels were similar in SCH cases but the levels were significantly elevated ($p<0.001$) in OH cases when compared to control subjects.

After therapy no changes were seen in CRP levels in SCH cases, while we observed a significant decrease ($p<0.001$) in tHcy levels in both groups, after treatment.

Serum MDA levels were found to be significantly higher ($p<0.001$) in both cases. The activity of Paraoxonase was lower ($p<0.01$ and $p<0.001$ respectively) in both SCH and OH patients as compared to controls. L-thyroxine therapy significantly decreased ($p<0.001$) serum MDA and significantly increased ($p<0.001$) serum PON-1 activity.

In overt group, serum FT4 levels showed a significant negative correlation with T-CHO ($P\leq0.01$), LDL-C ($P\leq0.01$) and CRP ($P\leq0.05$) levels ($P\leq0.01$), whereas a positive correlation was observed between FT4 and HDL-C levels, on the other hand serum TSH showed a significant positive correlation with T-CHO ($P\leq0.001$), LDL-C, TG, Hcy, CRP and MDA levels ($P\leq0.01$ for all comparison).

Discussion

In this study we found that the prevalence of thyroid disease is higher in women and increasing with advancing age. Alka *et al.,* (2002) (15) also reported higher prevalence rates for hypothyroidism with advancing age especially in females. Otto Mayer *et al.,* 2006 (16) in his study found the overall prevalence of hypothyroidism was 11.5%. It was about 4 times more prevalent in females, than in males (23.4% vs 6.9 %, respectively). The higher prevalence of thyroid disease in women suggests that estrogen might be involved in the pathophysiology of thyroid dysfunction. Estradiol has an antagonistic effect on the hormones T3 and T4. The reason being, estradiol competes with T3 and T4 for binding sites on the receptor proteins (Vasudevan N *et al.,* (2002) (17)). Moreover, estradiol also limits the thermogenic action of T4 and promotes storage of fat.

The effect of subclinical hypothyroidism on serum lipid levels and cardiovascular disease remain surrounded by controversy. In our study in SCH cases we found that the mean levels of T-CHO, LDL-C, and TG were significantly increased ($P\leq0.01$), ($P\leq0.001$) and ($P\leq0.05$ respectively), on the other hand the mean level of serum HDL was non-significantly decreased when compared to healthy controls. A significant reduction of serum cholesterol, LDL-cholesterol and TG concentrations were established during thyroid hormone replacement therapy in our study. This is in agreement with the study of Caparevi Z *et al.,* (2003) (18) who also found increased serum cholesterol, LDL-cholesterol and lower HDL-cholesterol values in SCH cases. J. Kvetny *et al.,* (2004) (19) reported higher triglycerides and lower HDL cholesterol levels in SCH patients compared to euthyroid subject.

In overt hypothyroid cases mean levels of T-CHO, LDL-C, and TG were found to be significantly increased ($P\leq0.001$ for each comparison) and the mean level of serum HDL was found to be significantly decreased ($P\leq0.05$) when compared to healthy controls. These findings support other studies done by Duntas LH *et al.,* (2002) (20) and K. Suganthy *et al.,* (2011) (21). Thyroid receptors seem to mediate the effects of thyroid hormones on lipid metabolism, and more specifically alpha 1 receptors control the lipogenesis in white adipose tissue, and βreceptors regulate the activity of lipogenic and lipolytic enzymes in the liver (22,23). Thyroid hormones stimulate cholesterol synthesis by inducing 3-hydroxy-3-methyl-glutaryl coenzyme A reductase in the liver (13). Thyroid hormones affect lipoprotein lipase activity and thus, the hydrolysis of triglycerides into very-low density lipoprotein (VLDL) and chylomicrons into fatty acids and glycerol (22). In hypothyroidism, lipoprotein lipase activity in the adipose tissue has been found normal or decreased, in addition to decreased hepatic lipase activity resulting in normal or high levels of triglycerides (24-26). Thyroid hormones affect cholesteryl ester transfer protein and hepatic lipase activity, decreased in hypothyroidism, with consequent changes not only in total high-density lipoprotein (HDL) but also in HDL subfraction levels (23,2788,90). Experimental evidence suggests that thy-

roid hormones might also affect cholesterol-7alpha-hydroxylase in liver (28,29). Thyroid hormones, especially triiodothyronine (T3), induce low- density lipoprotein (LDL) receptor gene expression in the liver, enhancing LDL clearance and explaining the increased LDL levels observed in hypothyroidism (22).

In our study we found that pretreatment tHcy levels were similar in SCH patients and control subjects, whereas in overt hypothyroid cases, serum tHcy levels were significantly (P≤0.001) higher than those of the healthy subjects and after treatment these levels were found to be decreased significantly (P≤0.001). This is in agreement with the study of Ozmen B *et al.,* (2006) (30), Christ-Chrain *et al.,* (2003) (31) and Cakal B *et al.,* (2007) (32). The pathogenesis of elevated tHcy in hypothyroidism is generally attributed to the reduction of glomerular filtration rate, because of the well-established effect of hypothyroidism on the kidney (Kreisman *et al.,* 1999) (33). Thyroid status also has a profound influence on a variety of biochemical processes, some of which may have secondary effects on the tHcy metabolism. Thyroid hormones markedly affect riboflavin metabolism, mainly by stimulating flavokinase and thereby the synthesis of flavin mononucleotide and flavin adenine dinucleotide. Conceivably, these metabolic changes may affect tHcy metabolism because flavin mono-nucleotide and FAD serve as cofactors for enzymes involved in the metabolism of vitamin B6, cobalamin, and folate. Circulating tHcy concentrations in hypothyroidism can rise through reduced activity of the flavoprotein methylene tetrahydrofolate reductase (MTHFR), an enzyme involved in the catalysis of Hcy and its remethylation to methionine. Hypothyroid individuals can be defective in converting riboflavin to the co-enzyme FAD, and consequently, deficient in MTHFR activity (32).

In our study the mean CRP levels were found to be significantly higher in both SCH patients (P≤0.05) and in overt hypothyroid patients (P≤0.01) than that of control subjects. After treatment CRP levels were found to be decreased significantly (P≤0.001) in overt hypothyroid patients. Similar to our study, Ozcan *et al.,* (2005) (34) found significantly elevated CRP levels in the high-normal range in subclinical hypothyroidism. Furthermore, they showed that the elevated CRP levels were reduced after thyroxine replacement. In another study, Tuzcu *et al.,* 2005 (35) observed that CRP levels are significantly increased and positively correlated with serum TSH in subclinical hypothyroidism. It might be suggested that cellular hypothyroidism induces an energy lack leading to a chronic intracellular inflammatory process, affecting the endothelial cells causing development of atherosclerosis (36). So it could be speculated whether inflammatory mechanisms may be involved in detoriation of thyroid function and at the same time increase the risk of CVD. An increase in CRP was caused by thyroiditis causing SCH.

Resch *et al.,* (2002) (37) found that hypothyroidism was associated with enhanced oxidative stress and lipid peroxidation, and supposed that this might lead to the development and progression of atherosclerosis. In our study we found a significant increase (P≤0.001) in MDA levels in both SCH and overt hypothyroid cases, after treatment these levels were found to be decreased significantly in all cases. Similar results are showed by Erdamar H *et al.,* (2008) (38), Dumitriu L *et al.,* (1998) (39) and Baskol G et al., (2007) (40) they found that the mean MDA level was significantly higher in hypothyroid patients by comparison to the control group.

We found a significant decrease in PON-1 levels in SCH (P≤0.01) and in overt hypothyroidism cases (P≤0.001). These results are in agreement with the study of E. Cebeci *et al.,* (2011) (41) and Baskol G *et al.,* (2007) (40) and are disagreed with the study of Coria M J *et al.* (2009) (8), in his study PON-1 activity did not show differences between SCH, overt hypothyroidism and ET woman. The protective effect of thyroid hormone against oxidative stress can be explained by the function of antioxidants as a defense system. However, a chronic state of hypothyroidism is characterized by impairments in the redox potential. This leads to free radical chain reactions and to metabolic suppression of antioxidant capacity. Our results from this study support the suggestion that the hypothyroidism of patients with intellectual disability in some way is linked to the low levels of the major antioxidant molecules found in these patients. The depletion of antioxidants observed in hypothyroid individuals may reflect the increased free radical production in the electron transport chain in the mitochondrial inner membrane. The increase of free radicals is not compensated, as one would expect, by a decrease of antioxidants. A high oxidative state in hypothyroid people has metabolic and biochemical

characteristics such as increased mitochondrial enzyme activity. Thus, it is likely that patients' cells are damaged by prolonged oxidative stress that far exceeds the capacity of the patients' organs to synthesize antioxidant molecules or to synthesize them from extracellular sources (42).

Conclusion

Thyroid dysfunction has to be considered a highly prevalent condition, mainly in females, which could potentially contribute to the overall coronary risk. Both subclinical and overt hypothyroidism are associated with increased T-CHO, LDL-C, triglycerides, CRP, MDA and creatinine levels and accelerates the process of atherogenesis and elevates cardiovascular risk. We confirmed the observation of increased homocysteine levels in overt hypothyroid patients. In contrast it does not appear to contribute to the increased risk for atherosclerotic disease in patients with SCH. As hypothyroidism may be a treatable cause of hyperhomocysteinemia, and a fasting t-Hcy is associated with a significant increased relative risk of coronary artery disease, measurement of t-Hcy to screen this dynamic association of cardiovascular risk factors during hypothyroidism may be of interest. Present study suggests a high production of ROS and oxidative stress in patients with thyroid diseases, with enhanced lipid peroxidation and concomitant failure of antioxidant defense mechanisms.

In conclusion it has been shown in our study that patients with thyroid diseases are at high risk for developing cardiovascular disorders. They have elevated atherogenic and cardiac risk marker in addition to altered oxidative stress parameters. Increased CRP and homocysteine might be a key molecule linking inflammation to oxidative stress in atherosclerosis. The activity of paraoxonase may be used for the determination of therapeutical response and during the follow-up. Therefore Patients with thyroid diseases should be monitored for cardiac risk also with the supplementation with antioxidants from an early stage of the disease. Treatment with thyroid hormone replacement to restore euthyroidism reverses the risk ratio.

References

1. Jennifer A. Phillips. Thyroid hormone disorders. May 2001.
2. Carl A. Burtis, Edward R. Ashwood & David E. Bruns. Textbook of Clinical Biochemistry & Molecular Diagnostics Tietz fourth edition. 52: 2053-2093.
3. Mariela Janet Coria, Adriana Ines Pastran, Maria Sofia Gimenez. Serum oxidative stress parameters of women with hypothyroidism. Acta bio-medica. 2009; 80(2): 135-9.
4. Mary J. Shomon. Thyroid awareness month guided to thyroid disease. 2010.
5. Marc B. Stone and Robert B. Wallace. Medicare coverage of routine screening for thyroid dysfunction. ISBN-10: 0-309-08885-2.
6. Juan J. Diez. Hypothyroidism in patients older than 55 Years an analysis of the etiology and assessment of the effectiveness of therapy. The Journals of Gerontology Series A 2002; 57: M315-M320.
7. Lithell H, Boberg J, Hellsing K, et al., Serum lipoprotein and apolipoprotein concentrations and tissue lipoprotein lipase activity in overt and subclinical hypothyroidism: the effect of substitution therapy. Eur J Clin Invest. 1981; 11: 3-10.
8. Melpomeni Peppa, Grigoria Betsi, and George Dimitriadis. Lipid abnormalities and Cardiometabolic Risk in Patients with Overt and Subclinical Thyroid Disease. Journal of Lipids. 2011.
9. Xiang Gao, Odilia I. Bermudez, and Katherine L. Tucker. Plasma C-Reactive Protein and Homocysteine Concentrations Are Related to Frequent Fruit and Vegetable Intake in Hispanic and Non-Hispanic White Elders. J. Nutr. 2004; 134(4): 913-918.
10. Sibel Guldiken, Muzaffer Demir Ender Arıkan Armagan Tugrul. The Level of Serum High Sensitive C - reactive protein in Women with Hyperthyroidism. Turkish Journal of Endocrinology and Metabolism. 2005; 3: 85-88.
11. Serpil Turhan, Sevilay Sezer, Gonul Erden, Ali Guctekin, Fatma Ucar, Zeynep Ginis, Ozlem Ozturk, Sezin Bingol. Plasma homocysteine concentrations and serum lipid profile as atherosclerotic risk factors in subclinical hypothyroidism. Annuals of Saudi Medicine. 2008; 28(2): 96-101.
12. Verhelst J, Berwaerts J, Marescau B, Abs R, Neels H, Mahler C & De Deyn PP. Serum creatine, creatinine, and other guanidino compounds in patients with thyroid dysfunction. Metabolism 1997; 46: 1063–1067.
13. Torun AN, Kulaksizoglu S, Kulaksizoglu M, Pamuk BO, Isbilen E, Tutuncu NB. Serum total antioxidant status and lipid peroxidation marker malondialdehyde levels in overt and subclinical hypothyroidism. Clin Endocrinol. 2009; 70(3): 469-74.
14. Farbod Raiszadeh, Mehrdad Solati, Arash Etemadi, Fereidoun Azizi. Serum paraoxonase activity before and after treatment of thyrotoxicosis. 2004; 60 (1): 75–80.
15. Alka M. Kanaya, Fran Harris, Stefano Volpato, Eliseo J. Pérez-Stable, Tamara Harris, Douglas C. Bauer, Association Between Thyroid Dysfunction and Total Cholesterol Level in an Older Biracial Population.Arch Intern Med. 2002; 162: 773-779.
16. Otto Mayer, Jr, Jaroslav Simon, Jan Filipovsky, Marketa Plaskova, and Richard Pikner, Hypothyroidism in coronary heart

disease and its relation to selected risk factors. Vasc Health Risk Manag. 2006; 2(4): 499–506.
17. Vasudevan N, Ogawa S, Praff D. Estrogen and thyroid hormone interactions: Physiological stability by molecular specificity. Physiol Rev. 2002; 82: 923-944.
18. Caparevi Z, Bojkovi G, Stojanovi D, Ili V. Dyslipidemia and subclinical hypothyroidism. Med Pregl. 2003; 56(5-6): 276-80.
19. Kvetny J, Heldgaard PE, Bladbjerg EM, Gram J. Subclinical hypothyroidism is associated with a low-grade inflammation, increased triglyceride levels and predicts cardiovascular disease in males below 50 years. Clin Endocrinol 2004; 61: 232–238.
20. Duntas LH. Thyroid disease and lipids. Thyroid. 2002; 12(4): 287-93.
21. K. Suganthy, P. K. Mohanty, V. Shanmugapriya. A Study of Lipid Profile as a Predictor of Cardiovascular Risk in Women with Subclinical Hypothyroidism. Int J Nutr Pharmacol Neurol Dis 2011; 1: 19-26.
22. X. Zhu and S. Y. Cheng, "New insights into regulation of lipid metabolism by thyroid hormone," Current Opinion in Endocrinology, Diabetes and Obesity, 2010; 17(5): 408–413.
23. M. Heimberg, J. O. Olubadewo, and H. G. Wilcox, "Plasma lipoproteins and regulation of hepatic metabolism of fatty acids in altered thyroid states," Endocrine Reviews. 1985; 6(4): 590–607.
24. J. J. Abrams, S. M. Grundy, and H. Ginsberg, "Metabolism of plasma triglycerides in hypothyroidism and hyperthyroidism in man," Journal of Lipid Research. 1981; 22(2): 307–322.
25. K. S. L. Lam, M. K. Chan, and R. T. T. Yeung, "High-density lipoprotein cholesterol, hepatic lipase and lipoprotein lipase activities in thyroid dysfunction—effects of treatment," Quarterly Journal of Medicine. 1986; 59(229): 513–521.
26. R. M. Krauss, R. I. Levy, and D. S. Fredrickson, "Selective measurement of two lipase activities in post heparin plasma from normal subjects and patients with hyperlipoproteinemia," Journal of Clinical Investigation. 1974; 54(5): 1107–1124.
27. J. A. Berti, M. E. C. Amaral, A. C. Boschero et al., "Thyroid hormone increases plasma cholesteryl ester transfer protein activity and plasma high-density lipoprotein removal rate in transgenic mice," Metabolism, 2001; 50(50): 530–536.
28. G. Sauter, M. Weiss, and R. Hoermann, "Cholesterol 7β hydroxylase activity in hypothyroidism and hyperthyroidism in humans," Hormone and Metabolic Research, 1997; 29(4): 176–179.
29. V. A. B. Drover and L. B. Agellon, "Regulation of the human cholesterol 7βhydroxylase gene (CYP7A1) by thyroid hormone in transgenic mice," Endocrinology, 2004; 145(2): 574–581.
30. Ozmen B, Ozmen D, Parildar Z, Mutaf I, Turgan N, Bayindir O. Impact of renal function or folate status on altered plasma homocysteine levels in hypothyroidism. Endocr J. 2006; 53(1): 119-24.
31. Christ-crain M, Meier, Gugliel Metti M, Huberpr, Riesen W, Staub JJ et al., Elevated c-reactive protein and homocysteine values: cardiovascular risk factors in hypothyroidism? A cross-sectional and a double-blind, placebo - controlled trial. Atherosclerosis. 2003; 166: 379-386.
32. Cakal B, Cakal E, DemirbaβB, Ozkaya M, Karaahmetoβu S, Serter R, Aral Y. Homocysteine and fibrinogen changes with L-thyroxine in subclinical hypothyroid patients.J Korean Med Sci. 2007; 22(3): 431-5.
33. Kreisman SH & Hennessey JV. Consistent reversible elevations of serum creatinine levels in severe hypothyroidism. Archives of Internal Medicine. 1999; 159: 79–82.
34. Ozcan O, Cakir E, Yaman H, Akgul EO, Erturk K, Beyhan Z, Bilgi C, Erbil MK. The effects of thyroxine replacement on the levels of serum asymmetric dimethylarginine (ADMA) and other biochemical cardiovascular risk markers in patients with subclinical hypothyroidism. Clin Endocrinol. 2005; 63: 203–206.
35. Tuzcu A, Bahceci M, Gokalp D, Tuzun Y, Gunes K: Subcinical hypothyroidism may be associated with elevated high-sensitive C-reactive protein (low grade inflammation) and fasting hyperinsulinemia. Endocr J 2005; 52: 89–94.
36. Ibrahim M. Madani, El amin Ammar M., Mahmoud Niamat O., El Sanosi Mai M., EL Bagir Nabiela M. Effects of subclinical hypothyroidism on important serum lipids values of Sudanese women. Journal of Chinese Clinical Medicine. 2010; 5(7).
37. Resch, U., Helsel, G., Tatzber, F., and Sinzinger, H.: Antioxidant status in thyroid disfunction. Clin. Chem. Lab. Med., 2002; 40: 1132-1134.
38. Erdamar H, Demirci H, Yaman H, Erbil MK, Yakar T, Sancak B, Elbeg S, Biberoβu G, Yetkin I. The effect of hypothyroidism, hyperthyroidism, and their treatment on parameters of oxidative stress and antioxidant status. Clin Chem Lab Med. 2008; 46(7): 1004-10.
39. Dumitriu L, Bartoc R, Ursu H, Purice M, Ionescu V. Significance of high levels of serum malondialdehyde (MDA) and ceruloplasmin (CP) in hyper and hypothyroidism. Endocrinology. 1988; 26(1): 35-8.
40. Baskol G, Atmaca H, Tanriverdi F, Baskol M, Kocer D, Bayram F. Oxidative stress and enzymatic antioxidant status in patients with hypothyroidism before and after treatment. Exp Clin Endocrinol Diabetes. 2007; 115(8): 522-6.
41. E. Cebeci, F.A. Oner, M. Usta, et al., Evaluation of oxidative stress, the activities of paraoxonase and arylesterase in patients with subclinical hypothyroidism. Acta Bio-Medica. 2011; 82: 3.
42. Palanisamy Pasupathi and Raman Latha, Free Radical Activity and Antioxidant Defense Mechanisms in Patients with hypothyroidism Thyroid Science 2008; 3(12): 1-6.

Received: 23 December 2013
Accepted: 21 March 2014
Correspondence:
Purmina Dey Sarkar, MD
Prof and HOD of Medical Biochemistry MGM Medical College Indore MP. India
E-mail: sarudream.girl@gmail.com