



Original Research Article

An association of anti-thyroid peroxidase antibodies in clinical and subclinical hypothyroidism

Jusmita Dutta¹, Shweta Jain^{2,*}, Ashish Jain², Swapnil Jain³, Swati Jain⁴¹Dept. of Biochemistry, L.N. Medical College And Research Centre, Bhopal, Madhya Pradesh, India²Dept. of Biochemistry, People's College of Medical Science and Research Centre, Bhopal, Bhopal, Madhya Pradesh, India³PDA, Bhopal, Madhya Pradesh, India⁴Dept. of Community Medicine, RKDF Medical College Hospital and Research Centre, Bhopal, Madhya Pradesh, India

ARTICLE INFO

Article history:

Received 28-02-2019

Accepted 26-04-2019

Available online 21-09-2019

Keywords:

Anti-TPO

Clinical hypothyroidism

Subclinical hypothyroidism

TSH

ABSTRACT

Introduction: Among general people anti-thyroid antibodies and thyroid profile (TSH, T4, and T3) relation is not established. In several years presence of Anti-TPO antibodies may lead to progress of clinical thyroid disease. Therefore, to rule out underlying autoimmune method screening for Anti-TPO antibodies is required. Aim of the present study was to find an association of Anti-TPO antibodies in clinical and subclinical hypothyroidism.

Materials and Methods: A cross sectional study was conducted in department of medicine and biochemistry involving 74 cases and 74 controls during the period of December 2017 to December 2018. Patients history, clinical examination and various investigations including FT3, FT4, TSH, Anti-TPO, FBG and lipid profile were done.

Results: Out of 74 cases 30 cases had clinical hypothyroidism and 44 cases have subclinical hypothyroidism. 63.5% cases of hypothyroidism were Anti-TPO positive. 70.3% cases of Clinical hypothyroidism were Anti-TPO positive and subclinical hypothyroidism had 51.5% Anti-TPO positive. TSH had a significant positive correlation with Anti-TPO antibodies.

© 2019 Published by Innovative Publication.

1. Introduction

Thyroid diseases are, debatably, one of the commonest endocrine disorders in world. India too is not an exemption. Various studies results on thyroid disease, showed that more than 42 million people suffer from thyroid disorders in India.¹

Clinical Hypothyroidism is defined as high serum TSH concentration with decrease in free Thyroxine (FT4) and free Tri-iodothyronine (FT3) concentrations associated with clinical sign and symptoms. Subclinical hypothyroidism (ScHt) is defined as high Serum TSH concentration with normal serum free Tri-iodothyronine (FT₃) and free Thyroxine (FT₄) concentrations, allied with few or negligible sign and symptoms of hypothyroidism.² The prevalence is 9.4% for subclinical hypothyroidism.³

Clinical presentation may differ from mild and asymptomatic to severe and overt disease, it may depend on patient's age, gender, physical condition, and the pace at which hypothyroidism develops cold intolerance, puffiness, coarse skin, weight gain, decrease in physical and mental activity, periorbital puffiness, pitting oedema are the usual signs and symptoms which may present in clinical hypothyroidism and seldom in subclinical hypothyroidism. Hypothyroidism may be caused by defect in thyroid (Primary), pituitary (Secondary), hypothalamus (Tertiary). Generally, it is primary hypothyroidism caused by iodine deficiency, autoimmunity and other reasons.

The autoimmune thyroid diseases are associated with number of auto antibodies, which are categorized as either primary or secondary antibodies.^{4,5} Primary antibodies are directly pathogenic and often directed against cell membrane receptors, whereas secondary antibodies do not appear to be concerned in pathogenesis but can act as

* Corresponding author.

E-mail address: dr.sweta.ind@gmail.com (S. Jain).

useful diagnostic tool for the presence of autoimmune thyroid disease.⁶ One of the major secondary antibodies associated with autoimmune thyroid disease are Anti thyroid Peroxidase Antibodies. The most common autoimmune thyroid disease is Hashimoto thyroiditis which is characterized by gradual thyroid failure with or without goitre development.⁷ Nearly all Hashimoto thyroiditis patients have high serum concentrations of antibodies against one or more thyroid antigens that are produced by lymphocytic infiltrate in the thyroid gland or, to a small extent, by regional lymph nodes or bone marrow.

Among general people anti-thyroid antibodies and thyroid profile (TSH, T4, and T3) relation is not established. In several years presence of Anti-TPO antibodies may lead to progress of clinical thyroid disease.⁸ Determining anti-thyroid antibodies and thyroid profile relation could make out such group of patients who have unstable thyroid profile. Therefore, to rule out underlying autoimmune method screening for Anti-TPO antibodies is required.⁹ Diagnosis of autoimmune thyroid disorders is important for treatment modifications for autoimmunity.

2. Materials and Methods

Present research was done in the Department of Medicine And Biochemistry, following the approval of Research advisory committee and Institutional Ethics Committee People's College of Medical Science and Research Centre, Bhopal.

2.1. Study design

A hospital based cross sectional study was conducted. Sample size was of 148 subjects with 74 cases and 74 controls.

2.2. Source of data

The blood samples collected from healthy and hypothyroid persons under the supervision of physician who attend their routine checkup in the department of medicine during the period April to December 2018 at People's hospital Bhopal.

2.3. Sample size

148 subjects with 74 cases and 74 controls.

2.4. Inclusion criteria

1. Willing to participate in study.
2. Patients in the age group of 18 to 60 years of age.
3. Patients with clinical and biochemical evidence of hypothyroidism.

2.5. Exclusion criteria

1. Persons not willing to participate in study.

2. Females in gestational or postpartum period
3. Thyroid destruction (from radioactive iodine or surgery)
4. Medications causing thyroid dysfunction like amiodarone, lithium, anti thyroid drugs

2.6. Study variable

1. Age
2. Gender
3. Education

2.7. Sampling collection

After overnight fasting for 8-12 hours, approx. 3-5 ml blood sample for thyroid function test and lipid profile in plain vials, for glucose in fluoride vial samples was centrifuged at 3000rpm for 10 minutes. Serum is separated and immediately stored in freezer at -20°C till further analysis.

Estimation of FT3, FT4 and TSH was done with elecsys kit based on electro-chemiluminescence method. Anti-TPO was estimated with biodetect Thyroid peroxidase ELISA kit. TC, HDL, VLDL and LDL, FBG were also estimated. Normal value of FT3, FT4, TSH were 1.5-4.2pg/ml, 0.8-2ng/dl and 0.4-5.5 μ IU/ml. Anti-TPO normal value was <40IU/ml.

2.8. Statistical analysis

In present study SPSS Version 20.0 software has been used for analysis. Underlying statistical tests has been used to derive the Results: Descriptive analysis is done using percentage distribution among the subject under study and Variables are presented using Mean and SD. Significance (P value) is assessed at 5% level significance.

3. Results

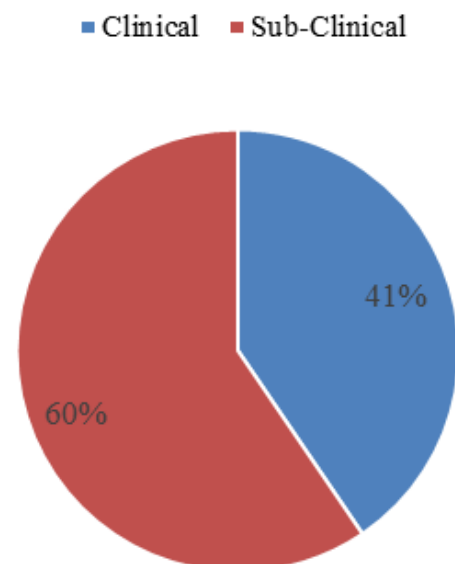
The Results of 148 subjects were analyzed. Age and sex matched healthy control subjects were chosen to compare the results. The percentage of case to control was 1:1. Data was analyzed and it was found that 59% cases have subclinical hypothyroidism and 41% cases have clinical hypothyroidism (Graph 1).

Table 1: Percentage of Anti-TPO positive in Hypothyroidism

	N	Percentage of Anti -TPO +ve
Cases	74	63.5%
Subclinical Hypothyroidism	44	70.3%
Clinical Hypothyroidism	30	51.5%

Subclinical hypothyroidism showed more anti-TPO positivity than clinical hypothyroidism.

Distribution of Hypothyroidism

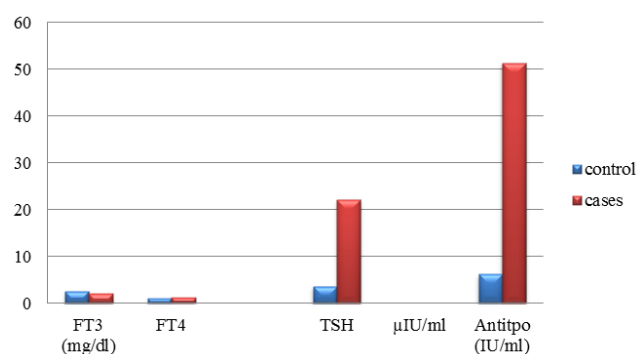


Graph 1:

Table 2: Gender Distribution of Hypothyroidism Patients

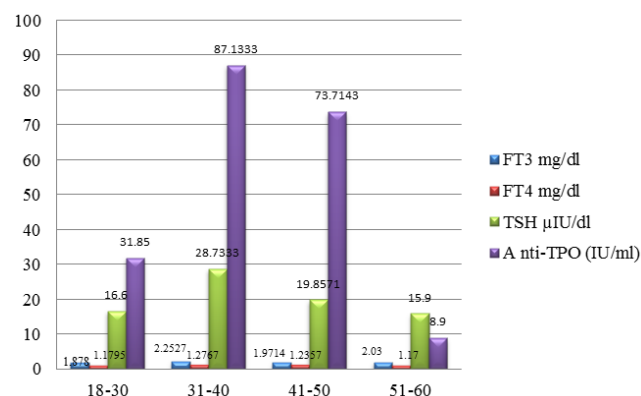
	Female	Male
Hypothyroidism	48	26
Clinical Hypothyroidism	20	10
Subclinical Hypothyroidism	28	16

In clinical hypothyroid from 30 cases 20 were female and 10 were male and from 44 cases of subclinical hypothyroidism 28 were female and 16 were male.

**Graph 2:** Distribution of Serum thyroid profile among controls and Hypothyroid patients

The mean value of FBG, TC, LDL, FT3, and FT4 was found to be higher in women as compared to men. But there was important variation seen in TSH and Anti-TPO between men and women of cases and controls (Graph 2). It was seen higher in women. In hypothyroid patients there was

noteworthy difference observed in TC, FT3, TSH, and Anti-TPO. But no significant change observed in FBG, HDL, LDL, and FT4.

**Graph 3:** Age wise changes in thyroid profile in hypothyroid patients

With age the mean value of FBG, TC, LD increases and it was seen that mean of Anti-TPO and TSH is considerably more in the age group 31-40 years. Mean value of TSH and Anti-TPO is higher in clinical hypothyroid patients than subclinical hypothyroid patients (Graph 3).

Table 3: Anti-TPO antibody and TSH correlation

	r	p
Hypothyroidism	.371	0.001
Clinical hypothyroidism	0.412	0.001
Subclinical hypothyroidism	0.476	0.003

Anti-TPO shows significant relation with TSH in cases but not in control. There was no important relation seen with other biomarkers in case. Correlation of Anti-TPO with TSH comes out as $r = .371$.

Correlation of anti-TPO with other biomarkers in clinical and subclinical hypothyroidism was positive. Correlation of Anti-TPO in subclinical hypothyroidism is $r = .476$ is significant. Correlation of Anti-TPO with clinical hypothyroidism is positive.

4. Discussion

Immune mediated alterations in the thyroid gland results in functional alterations in the thyroid hormone status leading to autoimmune thyroid disease. Transient phases of hypothyroidism occur in sub-acute thyroiditis, which commonly occurs due to destruction of the thyroid follicular cells. Sub clinical hypothyroidism is common than overt hypothyroidism with autoimmunity as the aetiology. The major indicators of hypothyroidism are weight gain, constipation, dryness of skin, cold intolerance and weakness tiredness.¹⁰ Long term undiagnosed hypothyroidism

results in dyslipidemia, diabetes, cardiovascular abnormalities and sexual dysfunction. Abnormalities of lipoprotein metabolism also found in hypothyroidism. The initiation of autoimmunity can be due to genetic factors, environmental factors, stress, infection, iodine deficiency.¹¹⁻¹³

In our study group total number of cases of hypothyroidism were 74 out of which 30(40.5)% clinical hypothyroidism and 44(59.5)% subclinical hypothyroidism. Similar results were shown by Jeena et al (2013) that the most common thyroid disorder was subclinical hypothyroidism in the study population followed by overt hypothyroidism.¹⁴ Study done by Khadka Shrestha Srijana et al (2017) also showed Subclinical hypothyroidism (56.3%) were most common than overt hyperthyroidism (18.0%), overt hypothyroidism (16.9%) and subclinical hyperthyroidism (8.8%).¹⁵

In present study 47 patients out of 74 patients were Anti-TPO positive that is 60.5%. Similar results were shown by Jeena et al (2013) that 60% of the total hypothyroid cases are Anti-TPO positive. From 44 cases of clinical hypothyroidism 70.3% cases were Anti TPO positive. 30 cases of subclinical hypothyroidism shows 51.5% Anti - TPO positivity. Similarly, Mohanty et al have showed 45 out of the 61 subjects (73.78%) had elevated anti-TPO in subclinical hypothyroid patients.¹⁶ Similarly Jay shankar CA et al (2015) 33 were anti-TPO positive and 17 were negative from the 50 subjects with either clinical or subclinical hypothyroidism. 80% were positive for anti-TPO among the clinical hypothyroidism Whereas patients with ScHt showed (50%) anti-TPO positivity.¹⁷

In this study, an elevated TPO antibody titre was in majority of hypothyroid patients. Cases with negative results for Anti-TPO antibodies, reason could be that few of them still showing cytological evidence of autoimmune destruction. The active autoimmune process continuing in the thyroid gland leads to the high antibody titres Also, 5 of our euthyroid subjects had positive antibody titres. This may be due to well-compensated thyroid function at present with a future risk of dysfunction in them. But, this high prevalence (25%) of positive antibody titre in euthyroid subjects reflects a referral bias, as this sampling was done on subjects attending the hospital.

In this study it was seen that mean value of TSH and Anti TPO was considerably elevated in patients as compared to normal controls ($p < 0.001$). It is also observed that FT4 mean is more in cases than in controls but FT3 mean is more in controls than in cases. Past few studies showed contradiction for FT4.

These findings confirm that our study group is having higher TSH and Anti TPO level. Previous studies Hollowel J G et al (1994) showed that the Anti-TPO Antibody is common in euthyroid subjects.¹⁸ In the NHANES III study, 14.6% and 8.0% of euthyroid females and males had Anti-TPO antibodies.

Our study shows that mean value of FBG is more in female in hypothyroid patients as well as controls. Same results were shown by Kim MK, Kwon HS (2010).¹⁹ Total cholesterol mean is more in men in cases while more in women in controls. HDL mean value is more in female than in male in controls and hypothyroid patients both. LDL mean value is higher in women than in men in controls and hypothyroid patients both. Our results are similar to the finding showed by Dhakal et al (2017) found higher prevalence of anti-TPO antibodies in women's than males (74% versus 26%).²⁰ Our study showed significant association with Anti-TPO so Anti TPO also increases with increase in blood sugar level. On the contrary, Ganesan Subramanyam, Josephine Latha Pushparaj et al (2014) No significant correlation exist between fasting blood sugar level and anti TPO level.²¹

Unnikrishnan et al (2013) studied in eight places of India and found higher prevalence of Anti-TPO antibodies in women than in men.²² United States national health and nutrition examination survey done by Hollowell et al (1994) showed that prevalence of both anti-TPO and anti-TG is higher in women.

In our study, it was found that Anti-TPO mean value was more in 31-40 years age group. Same results revealed by Swain et al (2005)²³ also showed that (95%) of the patients with autoimmune thyroid disease were women, mainly in the age group of 30-50 while Jay Shankar et al (2015) found autoimmune thyroid disease mainly in the age group of 26 to 40 years. Similar results shown by Saha et al (2005) showed that age group of 36-45 years was more common (40.5 %) for hypothyroidism with obvious female preponderance.²⁴

Our study shows TSH mean value is more in clinical hypothyroidism than subclinical hypothyroidism in and Anti-TPO is higher in clinical hypothyroid patients than subclinical hypothyroid patient's similar results shown by Ghoriasian et al,²⁵ Swain et al.

Present studies shows that Anti-TPO and TSH correlation comes out to be $r = 0.371$ in hypothyroid patients. In the past it showed positive relation between thyroid function test and anti-TPO antibody levels. Vaseghani et al found from their research that anti-TPO antibody titre correspond to TSH titres.²⁶ An important association between TSH or T4 concentration and increased anti-TPO antibody was shown by Ghoraisian et al. Recent study of Thomas cyriae et al (2015) showed the same results.²⁷ Studies done in Greece showed that Anti-TPO positive in subclinical hypothyroidism²⁸ while other showed no significant difference.²⁹

Weight and hypothyroidism In our study it was found that percentage of Clinical hypothyroidism patients were more obese than subclinical hypothyroidism and the same results were seen in the previous study by Verma A et al (2013) that, obesity was more common (46% vs. 34%) in overt than in subclinical hypothyroidism.³⁰

Limitations of present study are referral bias and small size of sample. It is a cross-sectional analysis of results. A larger sample with clinical and FNAC correlation is required to validate this test in our population.

5. Conclusion

Primary hypothyroidism is one of the most common endocrine disorders come across and managed and treated by physician. Unfortunately, the hypothyroidism symptoms are quite unclear and otherwise highly prevalent in the population. Therefore, to rule out the diagnosis of hypothyroidism physicians have to rely on biochemical testing. Therefore, present Study concludes as Anti-TPO comes out to be positive in cases of hypothyroid patients and autoimmunity is the main cause of hypothyroidism no a days. Subclinical hypothyroid cases are diagnosed only when they undergo routine thyroid profile testing with Anti-TPO.

6. Acknowledgement

I Heartly thankful to the almighty God and all the well wishers who guided and helped in my research work.

7. Source of Funding

None.

8. Conflicts of Interest

None

References

1. Available from: <http://www.ias.ac.in/currsci/oct252000/n%20kochochupillai..>
2. Werner and Ingbar's The Thyroid: A fundamental and clinical text. Philadelphia: Lippincott Williams and Wilkins . ; 2000,. p. 1001–7. 8th ed.
3. Menon U, Sundaram K, Unnikrishnan AG, Jaykumar RV, Nair V, Kumar H. High prevalence of undetected thyroid disorders in an iodine sufficient adult south Indian population. *J Indian Med Assoc.* 2009;107:72.
4. Ghoraishian SM, Moghaddam SHH, Afkhami M. Seyed Hossein Hekmati Moghaddam and Mohammad Afkhami. Relationship between Anti-Thyroid Peroxidase Antibody and Thyroid Function Tests. *W J Med Sci.* 2006;1(1):44–4712.
5. Pedersen I, Laurberg P, Knudsen N, Jørgensen T, Perrild H, et al. A population study of the association between thyroid auto antibodies in serum and abnormalities in thyroid function and structure. *Clin Endocrinol (Oxf).* 2005;62(6):713–20.
6. Prummel MF, Strieder T, Wiersinga WM. The environment and autoimmune thyroid diseases. *hypothyroidism Thyroid.* 2002;150(5):69–75. *Eur J Endocrinol.*
7. Caturegli P, A DR, Rose NR. Hashimoto thyroiditis: clinical and diagnostic criteria. *Autoimmun Rev.* 2014;13(4-5):391–7.
8. Hutfless SM, Matos P, Talor MV, Caturegli P, Rose R. Significance of prediagnostic thyroid antibodies in women with autoimmune thyroid disease. *The Journal of Clinical Endocrinology & Metabolism.* 2011;96(9):1466–71.
9. McLeod DSA, Cooper DS. The incidence and prevalence of thyroid autoimmunity. *Endocr J.* 2012;42(2):252–65.
10. Georgiou E, Ntalles K, Ch P, Anousis ST. Clinical manifestations and use of microcomputer in the differential diagnosis between hypothyroid and obese women. *Arch Hell Med.* 1987;4:30–3.
11. Nagataki S, Yamashita S, Tamai H. Immunogenetics of autoimmune endocrine disease in autoimmune diseases of endocrine system. R V, editor. Boca Raton: CRC Press ; 1990,.
12. Braverman LE. Iodine induced thyroid disease. *Acta Med Austriaca.* 1990;17:29–33. Suppl 1 S29–33.
13. B DB, Mclean E, Andersson M, Rogers L, L. Iodine deficiency in 2007: Global progress since. *Food Nutr Bull.* 2003;29:195–202.
14. Jeena EJ, Malathi M, Sudeep K. A hospital-based study of anti-TPO titer in patients with thyroid disease. *Muller J Med Sci Res.* 2013;4:74–7.
15. Shrestha SK, Karmacharya K, Giri M, Bajracharya MR, Jha S. Relationship of thyroid peroxidase antibody test with abnormal thyroid function tests. *J Pathol Nep.* 2017;7:1172–5. Available from: [10.3126/jpn.v7i2.17996](https://doi.org/10.3126/jpn.v7i2.17996).
16. Jayashankar CA, Avinash S, Shashidharan B, Vijaya S, Shruthi KR, et al. The prevalence of anti-thyroid peroxidase antibodies in subclinical and clinical hypothyroid patients. *Int J Res Med Sci.* 2015;3(12):3564–6.
17. Mohanty AW, Reddy GC, Kusumanjali G, Sabapathy AS, Pragna R. Diagnostic strategies for Subclinical hypothyroidism. *IJCB.* 2006;23(3):279–82.
18. Hollowell JG, Staehling NW, Flanders WD, Hannon WH, Gunter EW, et al. *J Clin Endocrinol Metab.* 1988;67:489–99. National Health and Nutrition Examination Survey. Available from: [10.1210/jc.87.2.489](https://doi.org/10.1210/jc.87.2.489).
19. Kim MK, Kwon HS, Baek KH. Effects of thyroid hormone on A1C and glycated albumin levels in nondiabetic subjects with overt hypothyroidism. *Diabetes Care.* 2010;33(12):2546–8.
20. Dhakal S, Nagila A, Koirala R, Bhatta MP, Regmi S, et al. Correlation of anti-thyroid peroxidase antibodies (anti-TPO) with thyroid hormones in local population of Western Nepal. *Int J Adv Res.* 2017;5(4):692–9.
21. Subramanyam G. Pushparaj prevalence of anti-tpo antibody in type 1 diabetes and thyroid dysfunction in TPO antibody positive diabetics ICS. *J Evol Med Dent Sci*;1:668–676. Available from: [10.14260/jemds/104](https://doi.org/10.14260/jemds/104).
22. Unnikrishnan AG, Kalra S, Sahay RK, Bantwal G, John M, et al. Prevalence of hypothyroidism in adults: An epidemiological study in eight cities of India. *Indian J Endocr Metab.* 2013;17:647–52.
23. Swain T, Swain B, Mohanty K. Autoimmune Thyroid Disorders - An Update. *Indian Journal of Clinical Biochemistry.* 2005;20(1):9–17.
24. Saha HR, Sarkar BC, Khan SA, Sana NK, Choudhury S. A comparative study of thyroid hormone and lipid status in diabetic and non diabetic adults. *Open Access Sci Rep.* 2012;1(9):2–5.
25. Ghoraishian SM, Moghaddam SHH, Afkhami-Ardekani M. Relationship between anti-thyroid peroxidase antibody and thyroid function test. *Iran J Immunol.* 2006;3:146–9.
26. Vasheghani M, Jalali R, Dabbaghmanesh MD, Sadeghalvad AS, Omrani GR. Thyroid autoimmunity role in the evolution of endemic goitre in rural area. *Arch Iran Med.* 2011;14:164–6.
27. Cyriac T. Prevalence of hypothyroidism and its association with anti-thyroid peroxidase antibody among adult sea food consuming population attending a tertiary health care centre in Kerala. *Int J Biomed Adv Res.* 2015;6(09):648–55.
28. Zois C, Stavrou I, Kalogera C, Svarna E, Dimoliatis I, et al. High prevalence of autoimmune thyroiditis in schoolchildren after elimination of iodine deficiency in North western Greece. *Thyroid.* 2003;13:485–9.
29. Zimmermann MB, Moretti D, Chaouki N, Torresani T. Introduction of iodized salt to severely iodine deficient children does not provoke thyroid autoimmunity: A one year prospective trial in northern Morocco. *Thyroid.* 2003;13:199–203.
30. Verma A, Jayaraman M, Kumar HK, Modi KD. Hypothyroidism and obesity? Cause or effect. *Saudi Med J.* 2008;29:1135–8.

Author biography**Jusmita Dutta** Professor and Head**Shweta Jain** Post Graduate Student**Ashish Jain** Assistant Professor**Swapnil Jain** Associate Professor**Swati Jain** Assistant Professor

Cite this article: Dutta J, Jain S, Jain A, Jain S, Jain S. An association of anti-thyroid peroxidase antibodies in clinical and subclinical hypothyroidism. *Int J Clin Biochem Res* 2019;6(3):415-420.