# Research Article Exploring the Patterns of Biomarker Variability in Thyroid Dysfunction: Clinical Insights from a Prevalence Perspective

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(Received: 29-09-2024 Revised: 15-12-2024 Accepted: 02-01-2025)

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#### ABSTRACT

**Introduction and Aim:** Thyroid disorders, prevalent globally, represent a significant public health issue in India, where approximately 42 million individuals are affected. This study investigates the biochemical, immunological, and inflammatory profiles associated with thyroid dysfunction in northwestern Uttar Pradesh, India.

**Methods:** A prospective, cross-sectional study was conducted with 225 participants from Teerthanker Mahaveer University and Teerthanker Mahaveer Hospital and Research Center, categorized into hypothyroid, hyperthyroid, and euthyroid groups. Data collection involved biochemical analysis of thyroid hormones (T3, T4, TSH), lipid profiles (Cholesterol, Triglyceride, HDL, LDL, & VLDL), glycemic indices (Fasting Blood Glucose & HbA1c), and inflammatory markers (CRP), alongside immunological assessment of anti-thyroid antibodies (Anti-TPO, Anti-TG).

**Results:** Thyroid function tests revealed significantly elevated TSH levels in hypothyroid subjects and T3 levels in hyperthyroid subjects. Hypothyroid individuals showed higher BMI, fasting blood sugar (FBS), and glycated hemoglobin (HbA1c) levels, while hyperthyroid individuals had elevated triglycerides and reduced HDL cholesterol levels. Elevated anti-TPO and anti-TG antibodies were noted across both thyroid dysfunction groups, indicating an autoimmune component. Inflammatory marker CRP levels were significantly higher in hypothyroid patients. Significant gender differences were observed, with a higher proportion of females in hypothyroid and hyperthyroid groups compared to euthyroid individuals.

**Conclusion:** These findings underscore the impact of thyroid disorders on metabolic and inflammatory pathways, emphasizing the need for tailored diagnostic and therapeutic strategies. The study highlights the complex interplay between thyroid dysfunction and various health markers, advocating for integrated approaches to enhance the management and understanding of thyroid disorders in the Indian context.

**Keywords:** Thyroid disorders, Autoimmune thyroiditis, Hyperthyroidism, Euthyroidism, Hypothyroidism, Inflammatory markers, Anti-TPO antibodies, Anti-TG antibodies.

### 1. INTRODUCTION

Thyroid disorders represent a significant public health concern globally, and India is no exception. With an estimated 42 million individuals affected, thyroid diseases are among the most prevalent endocrine disorders in the

country [1]. In a nation where communicable diseases have traditionally dominated the public health agenda, the rising incidence of noncommunicable diseases (NCDs), such as thyroid disorders, is now recognized as a growing concern [2]. Among endocrine disorders, thyroid dysfunctions rank second only to diabetes mellitus in terms of prevalence and impact [1]. The thyroid gland, a small butterfly-shaped organ located in the neck, plays a crucial role in regulating numerous physiological processes through the secretion of thyroid hormones, triiodothyronine (T3) and thyroxine (T4) [3]. These hormones are vital for maintaining metabolic homeostasis, influencing energy thermogenesis, expenditure. and overall metabolic rate. The balance of thyroid hormone production is delicate, and any disruptions can lead to significant health issues. Hyperthyroidism, characterized by excessive thyroid hormone production, accelerates metabolism and manifests in symptoms such as nervousness, weight loss, heat intolerance, insomnia. palpitations, and Conversely, hypothyroidism, marked by insufficient hormone production, results in a slowed metabolism, with symptoms including weight gain, lethargy, cold intolerance, and depression [4].

Recent studies have highlighted the prevalence of thyroid disorders in India, with a crosssectional, multicentre study involving over 5,000 adults across eight major cities reporting the prevalence of subclinical hypothyroidism at 8.02%, overt hypothyroidism at 10.95%, subclinical hyperthyroidism at 1.27%, and overt hyperthyroidism at 0.67% [5]. These findings underscore the significant burden of thyroid disorders in the Indian population. Given that thyroid hormones affect nearly every tissue in the body, disturbances in their levels can have widespread implications, influencing not only metabolic processes but also cardiovascular lipid health. metabolism, and glucose homeostasis [6].

Traditionally, thyroid disorders in India were classified under iodine-deficiency disorders (IDDs). However, a World Health Organization assessment in 2004 reclassified India as having optimal iodine nutrition. Despite this, the high incidence of thyroid dysfunctions, such as hypothyroidism and hyperthyroidism, suggests that factors beyond iodine deficiency, such as autoimmune thyroiditis, contribute significantly the burden of thyroid diseases [7]. to Autoimmune thyroiditis, including conditions like Hashimoto's thyroiditis and Graves' disease, is a major cause of thyroid dysfunction. These conditions are characterized by the presence of specific antibodies, such as AntiTPO and AntiTG, which target and impair thyroid function. Elevated levels of these antibodies are often associated with reduced production of thyroid hormones, leading to hypothyroidism [8]. Furthermore, thyroid disorders profoundly impact lipid metabolism, with hypothyroidism often linked to elevated cholesterol levels and an increased risk of cardiovascular disease. Conversely, hyperthyroidism is associated with lower cholesterol levels but can still pose cardiovascular risks due to its effects on heart rate and rhythm [9]. Additionally, the interplay thyroid function and between glucose metabolism is critical, as thyroid hormones influence glucose absorption, insulin secretion, and gluconeogenesis. Disruptions in thyroid hormone levels can lead to metabolic imbalances, affecting glycemic control and increasing the risk of diabetes [10].

This study delves deeper into the patterns of thyroid dysfunction in Uttar Pradesh, India, with a focus on the biochemical, immunological, and inflammatory irregularities associated with these conditions. By examining the levels of thyroidstimulating hormone (TSH), T3, T4, and associated markers, this research aims to enhance our understanding of thyroid disorders and inform more effective diagnostic and therapeutic strategies.

### 2. MATERIALS AND METHODS

**2.1 Study setting and design:** A prospective, cross-sectional study was conducted in northwestern Uttar Pradesh, district of Moradabad.

**2.2 Sample size and sampling technique:** The research was conducted at the Study Center of the College of Paramedical Sciences,

Teerthanker Mahaveer University, and Teerthanker Mahaveer Hospital and Research Center, Moradabad, Uttar Pradesh, India. The simple random sampling technique was used to select samples from Teerthankar Mahaveer University Hospital and Research Centre. The study criteria outlined inclusion and exclusion criteria. Inclusion criteria encompassed patients with thyroid dysfunction aged 18 years and above. Exclusion criteria included patients with diseases other than thyroid, those above 65 years, individuals with surgeries affecting thyroid function, and pregnant women.

2.3 Ethics, consent, and permissions: The study conducted at Teerthanker Mahaveer College of Paramedical Sciences in Moradabad, Uttar Pradesh, India, has received approval from the College Ethics Committee. The approval was granted during the Ethics Committee Meeting held on January 19, 2023, at the Committee Room of the College of Paramedical Sciences, Teerthanker Mahaveer University, Moradabad. The study protocol, outlined in Annexure I to V, was approved by both the College Ethics Committee members and Academic Board members. Before participation, all patients were provided with a patient information sheet, and informed consent in both Hindi and English was obtained from each patient. The study is registered with the College Ethics Committee under reference PM/ETHICAL/COPS/2023/022. 2.4 Data collection: From 2022 to 2023, a crosssectional study was conducted among the thyroid population in Moradabad District, India. The study included 225 participants from Teerthankar Mahaveer Hospital and Research Centre, Moradabad, categorized into 75 Hypothyroid, 75 Hyperthyroid, and 75 Euthyroid individuals, aged between 18-65 years. Participants with thyroid dysfunction were involved, and a prevalidated questionnaire, both closed and openended, was administered after obtaining informed consent. The questionnaire, validated by experts, was translated into Hindi and backtranslated to English to ensure tool reliability.

**2.5 Clinical Examination:** In our study, the clinical examination was a systematic process designed to gather comprehensive data from

participants. It began with the completion of a pre-validated questionnaire and the acquisition of informed consent. This process ensured that participants were fully aware of the study's aims and methods and voluntarily agreed to participate.

**2.6 Sample Collection and Processing**: Following consent, blood samples were collected using venipuncture into three types of vacutainers: red, grey, and purple. The red and grey vacutainers were used for serum and plasma separation, respectively, by centrifugation at 2500 rpm for 15 minutes. The purple vacutainers were used to collect whole blood for further analysis.

**2.7 Sample Processing**: The collected samples underwent various biochemical, thyroid, and inflammatory tests:

- 1. Biochemical Parameters:
- Plasma Glucose: Levels were measured using a semi-analyzer (ERBA) employing Trinder's method, which quantifies glucose through an enzymatic reaction involving glucose oxidase.
- **HbA1c**: Determined using the Turbilatex method, this test evaluates average blood glucose levels over the previous three months.
- Serum Total Cholesterol: Measured using the semi-analyzer (ERBA) based on Allain's method, assessing lipid levels to evaluate cardiovascular risk.
- Serum Triglycerides: Assessed using the semi-analyzer (ERBA) with lipase enzymatic breakdown, important for diagnosing and managing hyperlipidemia.
- Serum HDL-Cholesterol: Measured using a modified precipitation method, reflecting the cholesterol transport capacity of high-density lipoproteins.
- Serum LDL-Cholesterol and VLDL-Cholesterol: Calculated using the Friedewald method, providing insight into cardiovascular risk by measuring lowdensity and very low-density lipoproteins.
- Triiodothyronine-T3, Thyroxine-T4, and Thyroid Stimulating Hormone-TSH: All measured using ELISA (Quanti Microlisa

Method), these tests assess thyroid function and hormone levels.

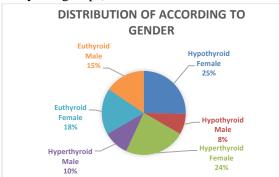
#### 2. Immunological Parameter:

- Anti-TG and Anti-TPO Autoantibodies: Detected using ELISA (Calbiotech Method), these markers are crucial for identifying autoimmune thyroid disorders.
- 3. Inflammatory Parameter:
- Plasma C-Reactive Protein (CRP): Assessed using the turbidimetric immunoassay method (Turbilyte-CRP kit), which measures systemic inflammation by evaluating CRP levels in serum or plasma.

**2.8 Statistical analysis**: Data were analyzed using SPSS software. The Kolmogorov-Smirnov test was used to assess data distribution normality. Descriptive statistics summarized demographic and clinical parameters, while inferential statistics, including paired t-tests, Pearson correlation, and ANOVA, were employed for analysis. Statistical significance was set at an alpha level of 5%, with a study power of 80%.

### 3. RESULTS

**3.1 Distribution of Cases by Gender:** In this study, out of the 225 participants, 151 were female (57 in hypothyroid, 53 in hyperthyroid, and 41 in euthyroid groups), and 74 were male (18 in hypothyroid, 22 in hyperthyroid, and 34 in euthyroid groups).



Graph 3.1: Distribution of cases of Hypothyroid, Hyperthyroid & Euthyroid according to Gender

A significant difference was observed in the gender distribution between hypothyroid and hyperthyroid groups compared to the euthyroid group, with a higher proportion of females in the former two groups ( $\chi^2 = 6.822$ , p = 0.0151).

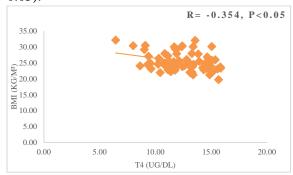
3.2 Thyroid Function Parameters: The mean T3 levels were significantly higher in both hypothyroid (2.00 ng/ml) and hyperthyroid (2.48 ng/ml) groups compared to euthyroid subjects with p-values < 0.00001. (1.05)ng/ml) between hypothyroid Differences and hyperthyroid groups were also significant (p <0.05). For T4 levels, hypothyroid subjects had lower mean levels (10.92 µg/dl) compared to euthyroid (8.12  $\mu$ g/dl) and hyperthyroid (12.58 µg/dl) groups, with all comparisons showing pvalues < 0.00001. TSH levels were markedly higher in the hypothyroid group (12.70 µIU/ml) compared to the euthyroid (2.61 µIU/ml) and hyperthyroid groups (0.09 µIU/ml), with pvalues < 0.00001.

3.3 Biochemical Parameters of BMI & Glycemic Index: Hypothyroid individuals had a significantly higher average BMI (26.27 kg/m<sup>2</sup>) compared to euthyroid (23.74 kg/m<sup>2</sup>) and hyperthyroid (25.25 kg/m<sup>2</sup>) individuals, with pvalues < 0.00001 and < 0.05, respectively. FBS and HbA1c levels were higher in both hypothyroid and hyperthyroid groups compared to the euthyroid group (FBS: 89.39 mg/dl in euthyroid vs. 121.45 mg/dl in hypothyroid; HbA1c: 5.39% in euthyroid vs. 8.05% in hypothyroid), with p-values < 0.00001. Differences in FBS between hyperthyroid and hypothyroid groups were not significant (p >0.05), whereas HbA1c levels were significantly different between these groups (p < 0.00001).

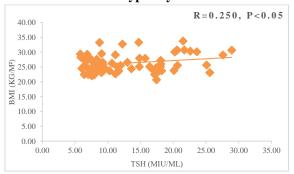
3.4 Lipid Profile Parameters: Hypothyroid individuals had higher cholesterol (179.95 mg/dl) compared to euthyroid (165.47 mg/dl), while hyperthyroid individuals also had elevated levels compared to euthyroid, though the differences between hypothyroid and hyperthyroid groups were not significant (p > p)0.05). Triglyceride levels were highest in the hyperthyroid group (132.47 mg/dl) compared to the euthyroid (123.73 mg/dl) and hypothyroid (112 mg/dl) groups, with significant differences (p < 0.05). HDL levels were lowest in hyperthyroid individuals (32.76 mg/dl) and highest in euthyroid individuals (47.79 mg/dl), with significant differences among groups (p <0.00001). LDL and VLDL levels were higher in hypothyroid individuals compared to euthyroid and hyperthyroid groups, with significant differences noted (p < 0.00001 for LDL and p < 0.05 for VLDL).

**3.5 Immunological Parameters:** Anti-TPO and anti-TG antibodies. Both hypothyroid and hyperthyroid groups had significantly higher levels of anti-TPO and anti-TG antibodies compared to euthyroid individuals. However, differences in anti-TPO and anti-TG levels between hypothyroid and hyperthyroid groups were not significant (p > 0.05).

**3.6 Inflammatory Parameters:** CRP levels were higher in hypothyroid individuals (8.08 mg/dl) compared to euthyroid (5.05 mg/dl) and hyperthyroid (5.07 mg/dl) groups. The differences in CRP levels between euthyroid and hyperthyroid groups were not significant (p > 0.05), but the differences between hypothyroid and the other two groups were significant (p < 0.05).



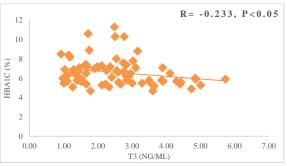
Graph 3.2: Correlation between serum T4 and BMI in hyperthyroidism



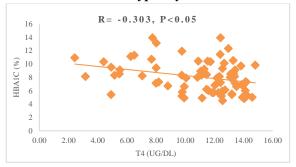
Graph 3.3: Correlation between serum TSH and BMI in hypothyroidism

**3.7 Correlation Between Thyroid Markers and BMI:** The correlation between thyroid markers and BMI. Significant negative correlations were observed between T4 levels and BMI (r = -0.354) in hypothyroid patients, while a positive correlation was noted in hyperthyroid patients (r = 0.250).

**3.8 Correlation Between Thyroid Markers and Glycemic Status:** A negative correlation between T3 levels and HbA1c (r = -0.233) in hyperthyroid individuals and between T4 levels and HbA1c (r = -0.303) in hypothyroid individuals.

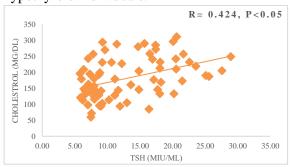


Graph 3.4: Correlation between serum T3 and HbA1c in hyperthyroidism

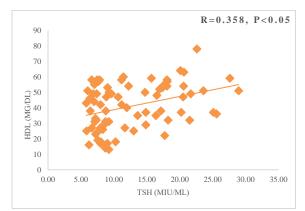


Graph 3.5: Correlation between serum T4 and HbA1c in hypothyroidism

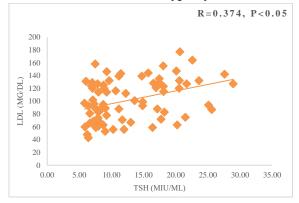
**3.9 Correlation Between Thyroid Markers and Lipid Profile:** The correlations between thyroid markers and lipid profiles. Significant positive correlations were observed between TSH levels and cholesterol (r = 0.424), HDL (r =0.358), and LDL (r = 0.374) in hypothyroid individuals. A negative correlation between T3 levels and VLDL (r = -0.237) in hyperthyroid and also between T4 levels and HDL (r = -0.242) in hypothyroid individuals.



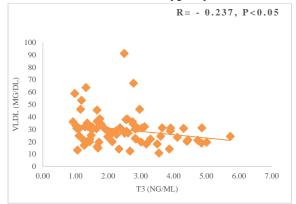
Graph 3.6: Correlation between serum TSH and Cholesterol in hypothyroidism



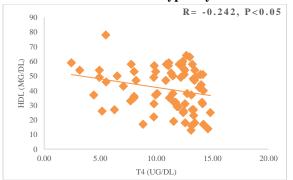
Graph 3.7: Correlation between serum TSH and HDL-Cholesterol in hypothyroidism



Graph 3.8: Correlation between serum TSH and LDL-Cholesterol in hypothyroidism

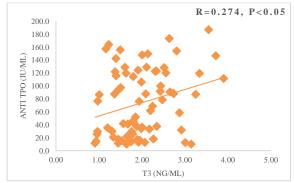


Graph 3.9: Correlation between serum T3 and VLDL-Cholesterol in hyperthyroidism

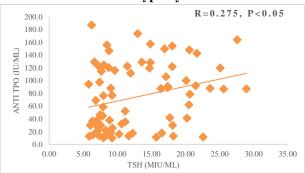


Graph 3.10: Correlation between serum T4 and HDL-Cholesterol in hypothyroidism

**3.10** Correlation Between Thyroid Markers and Immunological Parameters: A significant positive correlation between T3 levels and anti-TPO (r = 0.274) and TSH levels and anti-TPO (r = 0.275) in hypothyroid individuals. T4 levels also showed a significant positive correlation with anti-TG (r = 0.332) in hyperthyroid individuals.

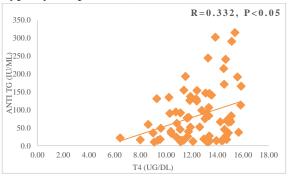


Graph 3.11: Correlation between serum T3 and Anti-TPO in hypothyroidism



Graph 3.12: Correlation between serum TSH and Anti-TPO in hypothyroidism

**3.11 Correlation Between Thyroid Markers and Inflammatory Parameters:** The correlation between thyroid markers and CRP levels. No significant correlations were found between T3, T4, and CRP levels in hypothyroid and hyperthyroid groups, although a modest positive correlation was noted for TSH levels in hypothyroid patients (r = 0.214).



Graph 3.13: Correlation between serum T4 and Anti-TG in hyperthyroidism

**3.12** Comparative Distribution of Thyroid and Related Parameters: Comparative analysis of T3, T4, TSH, BMI, FBS, HbA1c, cholesterol, triglycerides, HDL, LDL, VLDL, anti-TPO, anti-TG, and CRP levels across hypothyroid, hyperthyroid, and euthyroid groups. Statistically significant differences were found across all parameters, indicating distinct biochemical and immunological profiles among the three groups. Specifically:

- T3, T4, and TSH levels showed significant differences with hypothyroid individuals having the highest TSH and lowest T4 levels, while hyperthyroid individuals had the highest T3 levels.
- BMI and glycemic indices (FBS and HbA1c) were significantly higher in hypothyroid compared to euthyroid individuals.
- Lipid profile variations included higher cholesterol and LDL levels in hypothyroid individuals and higher triglycerides in hyperthyroid individuals.
- Anti-TPO and anti-TG antibodies were highest in hypothyroid individuals, indicating a strong autoimmune component.

### 4. **DISCUSSION**

4.1 Overview: This study aimed to evaluate the consequences of thyroid disorders on biochemical, immunological, and inflammatory markers, and to assess variations in these parameters across hypothyroid, hyperthyroid, and euthyroid groups. By categorizing 225 participants into these three groups-75 in each category-this research provides insights into how thyroid dysfunction influences various health markers. Our findings reveal significant differences in biochemical, immunological, and inflammatory parameters among the thyroid condition groups, with implications for understanding thyroid health and its broader impact.

#### 4.2 Biochemical Markers

**4.2.1 BMI and Glycemic Index:** Our study corroborates previous research indicating a significant association between obesity and hypothyroidism. The observed higher average BMI in hypothyroid individuals aligns with

Rong-hua Song et. al., [2019], who demonstrated a similar link. Additionally, we found a significant negative correlation between free T4 levels and BMI, consistent with [11], which suggests that lower T4 levels are associated with increased BMI. The elevated fasting blood sugar (FBS) levels observed in both hypothyroid and hyperthyroid groups corroborate [12], who reported similar findings. Our data also show significantly higher glycated hemoglobin (HbA1c) levels in hypothyroid individuals, in agreement with [13] and [14], highlighting the impact of thyroid dysfunction on glycemic control. The findings can be justified by the role of thyroid hormones in regulating metabolism and glucose homeostasis. In hypothyroidism, reduced hormone levels slow the basal metabolic rate (BMR), leading to decreased energy expenditure, weight gain, and fluid retention, which contribute to a higher BMI. Free T4, a precursor to T3, is negatively correlated with BMI, as lower levels slow metabolism, reduce thermogenesis, and impair fat burning, further promoting weight gain.

Additionally, thyroid dysfunction affects glucose metabolism. In hypothyroidism, insulin resistance is increased, leading to elevated fasting blood sugar (FBS) and higher HbA1c due to chronic hyperglycemia. Hyperthyroidism, conversely, enhances glucose production and reduces insulin sensitivity. These disruptions in glucose regulation highlight the impact of thyroid hormones on both energy balance and glycemic control.

4.2.2 Lipid Profile: Our results reveal markedly elevated cholesterol and triglyceride levels in hypothyroid individuals, consistent with previous studies such as [12]. The positive correlation between TSH levels and cholesterol, as well as LDL cholesterol, observed in our study, aligns with findings by [15] and [16]. In contrast, hyperthyroid individuals showed reduced HDL levels, in line with [17], indicating that thyroid dysfunction influences lipid metabolism differently depending on the thyroid condition. The elevated cholesterol and triglyceride levels in hypothyroid individuals can be attributed to reduced thyroid hormones, which slow LDL clearance and decrease lipid breakdown. The positive correlation between TSH and cholesterol, especially LDL, reinforces the connection between hypothyroidism and dyslipidemia. In contrast, hyperthyroidism lowers HDL levels due to faster lipid turnover and cholesterol clearance, indicating thyroid dysfunction affects lipid metabolism differently in hypo- and hyperthyroid conditions.

#### 4.3 Immunological Parameters

4.3.1 Anti-TPO AutoAb: Our findings of significantly elevated anti-thyroid peroxidase (Anti-TPO) antibodies in both hypothyroid and hyperthyroid individuals corroborate [18] and [19]. This suggests that elevated Anti-TPO levels are a common feature of thyroid dysfunction, reflecting underlying autoimmune an component. This consistent elevation of Anti-TPO antibodies across different thyroid dysfunctions may be due to an underlying autoimmune response affecting thyroid function. This suggests that thyroid dysfunction in both conditions involves an autoimmune mechanism targeting thyroid peroxidase, an enzyme crucial for thyroid hormone production.

4.3.2 Anti-TG AutoAb: Similarly, increased anti-thyroglobulin (Anti-TG) antibodies in both hypothyroid and hyperthyroid groups support previous findings by [20] and [21]. The positive correlation between T4 levels and anti-TG in hyperthyroidism, as observed in our study, indicates a potential interplay between thyroid hormone levels and autoimmune responses in hyperthyroid conditions. This finding may be due to an autoimmune response involving thyroglobulin, a protein essential for thyroid hormone synthesis. This suggests that thyroid hormone levels could influence the autoimmune with elevated Τ4 potentially response, exacerbating the immune attack on thyroglobulin in hyperthyroid conditions.

### 4.4 Inflammatory Parameters

**4.4.1 CRP and Hs-CRP:** Our research supports previous studies, such as those by [22], which observed elevated CRP levels in hypothyroid and hyperthyroid individuals. Notably, while hypothyroid individuals had significantly higher CRP levels compared to euthyroid controls, the

difference in hyperthyroid individuals did not achieve statistical significance, reflecting the variability observed in previous research like that of [23]. Our findings of significant variations in serum high-sensitivity C-reactive protein (Hs-CRP) levels across thyroid conditions align with [24], indicating that inflammatory responses differ with thyroid dysfunction status. The lack significant difference in hyperthyroid of individuals might be due to a less pronounced inflammatory response or a different underlying mechanism compared to hypothyroidism. This indicates that inflammatory responses associated with thyroid dysfunction may vary between hypo- and hyperthyroid states.

### 5. CONCLUSION

In conclusion, our study confirms significant associations between thyroid dysfunction and various metabolic and immunological parameters. Hypothyroid individuals exhibit higher BMI, elevated fasting blood sugar (FBS), and glycated hemoglobin (HbA1c) levels, consistent with reduced thyroid hormone levels slowing metabolism and impairing glucose control. Our findings also highlight markedly increased cholesterol and triglyceride levels in hypothyroid individuals, reflecting disrupted lipid metabolism. The elevated anti-thyroid peroxidase (Anti-TPO) and anti-thyroglobulin (Anti-TG) antibodies in both hypothyroid and hyperthyroid individuals suggest a common autoimmune component underlying thyroid dysfunction. Inflammatory markers, such as CRP and Hs-CRP, show significant variation across thyroid conditions, with hypothyroid individuals exhibiting higher levels, while hyperthyroid individuals show less pronounced changes. These results underscore the complex interplay between thyroid function, metabolic processes, and autoimmune responses, emphasizing the need for tailored management strategies for different thyroid disorders.

### 6. LIMITATIONS OF THE STUDY

We advocate for further investigations with larger sample sizes to validate our findings regarding the association between thyroid hormones and inflammatory markers. While our study offers insights, its cross-sectional nature warrants caution in interpretation. Future studies should explore associations between Mean Platelet Volume (MPV) and thyroid hormone levels in hypothyroidism, considering variables like sex and age. It's crucial to note that correlation doesn't imply causation, necessitating deeper research into underlying mechanisms and clinical implications.

### 7. ACKNOWLEDGEMENT

We acknowledge Teerthanker Mahaveer University Hospital and Research Center, College of Paramedical Sciences Department for data collection.

### 8. CONFLICT OF INTEREST

We declare that there is no conflict of interest in this research work.

## 9. FUNDING

There is no funding provided for this research.

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