Original Research Article

ROLE OF THYROID AUTOANTIBODIES IN PREDICTING PROGRESSION OF SUBCLINICAL HYPOTHYROIDISM: A PROSPECTIVE COHORT STUDY

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Received:08 Aug 2026 | Accepted: 14 Aug 2026 | Published: 19 Aug 2026

Abstract

Background: Subclinical hypothyroidism (SCH) is a common thyroid disorder characterized by elevated serum thyroid-stimulating hormone (TSH) levels with normal free thyroxine (FT4) concentrations. Although many patients remain stable, a proportion progress to overt hypothyroidism. Identification of reliable predictors of progression is essential for appropriate monitoring and management. Thyroid autoantibodies, particularly thyroid peroxidase antibodies (TPOAb) and thyroglobulin antibodies (TgAb), may serve as important markers of autoimmune thyroid dysfunction and disease progression.

Objectives: To evaluate the role of thyroid autoantibodies in predicting progression of subclinical hypothyroidism to overt hypothyroidism and to identify clinical and biochemical factors associated with disease progression.

Methods: A prospective cohort study was conducted among patients diagnosed with subclinical hypothyroidism. Baseline clinical characteristics, serum TSH, FT4, TPOAb, and TgAb levels were assessed at enrollment. Patients were followed prospectively, and progression to overt hypothyroidism was defined by elevated TSH levels with reduced FT4 concentrations during follow-up. The association between thyroid autoantibody status and progression was analyzed using appropriate statistical methods.

Results: Among the enrolled patients, a proportion progressed to overt hypothyroidism during follow-up. Progression was significantly higher among individuals with positive thyroid autoantibodies, particularly TPOAb positivity. Patients who progressed had higher baseline TSH levels and lower FT4 concentrations compared with those who remained stable. Multivariate analysis demonstrated that TPOAb positivity and higher baseline TSH levels were independent predictors of progression.

Conclusion: Thyroid autoantibody positivity, especially TPOAb, is an important predictor of progression from subclinical hypothyroidism to overt hypothyroidism. Assessment of thyroid autoantibodies along with baseline thyroid function tests may help identify high-risk patients and guide individualized follow-up and management strategies.

Keywords: Subclinical hypothyroidism; Thyroid autoantibodies; Thyroid peroxidase antibody; Thyroglobulin antibody; Overt hypothyroidism; Disease progression; Prospective cohort study.

Introduction

Thyroid disorders are among the most common endocrine disorders worldwide and represent an important public health concern due to their effects on metabolism, cardiovascular health, reproduction, and quality of life[1]. Hypothyroidism occurs due to inadequate thyroid hormone production and may present as overt or subclinical disease[2]. Subclinical hypothyroidism (SCH) is defined as an elevated serum thyroid-stimulating hormone (TSH) concentration with normal total thyroxine (T4) or

free thyroxine (FT4) levels. [3]SCH is frequently encountered in clinical practice, with an estimated prevalence of 4–10% in the general population. Its occurrence varies according to age, sex, ethnicity, iodine status, and the reference range used for TSH interpretation.[4] The prevalence increases with advancing age and is higher among females and individuals with autoimmune thyroid disease. Although many patients remain asymptomatic, approximately one-third may experience symptoms suggestive of thyroid hormone deficiency, including fatigue, dry skin, memory impairment, muscle cramps, cold intolerance, and voice changes. [5]Despite being considered a mild biochemical abnormality, SCH has been associated with several adverse metabolic, cardiovascular, and reproductive outcomes[6]. Previous studies have demonstrated associations between SCH and dyslipidemia, endothelial dysfunction, atherosclerosis, coronary artery disease, peripheral vascular disease, and myocardial infarction. [7,8] In women of reproductive age, SCH has also been linked with infertility and adverse pregnancy outcomes, including abortion and miscarriage. [9]]SCH represents an early stage of thyroid failure and may progress to overt hypothyroidism (OH), characterized by elevated TSH with reduced thyroid hormone levels. However, the natural history of SCH is variable; some patients experience spontaneous normalization of TSH, while others develop persistent thyroid dysfunction or progress to OH. Reported progression rates range from 3% to 18% annually depending on baseline TSH concentration, thyroid autoantibody status, age, sex, and other risk factors. [10,11]In a prospective study by Huber et al., involving women with SCH followed for 10 years, 57% remained with persistent thyroid dysfunction, 34% progressed to overt hypothyroidism, and 9% achieved normalization of thyroid function[12]. The Whickham survey further demonstrated that elevated TSH levels and thyroid antibody positivity were independent predictors of future thyroid dysfunction, with individuals having TSH levels >6 mIU/L showing a significantly increased risk of progression to overt hypothyroidism. [13,14]Several predictors of progression from SCH to OH have been identified, including higher baseline TSH, female sex, increasing age, low-normal FT4 levels, lithium therapy, previous radioiodine treatment, and neck radiation exposure. Among these factors, thyroid autoantibodies, particularly thyroid peroxidase antibodies (TPOAb), have emerged as important predictors due to their association with autoimmune-mediated thyroid destruction[15,16].Autoimmune thyroiditis, especially Hashimoto's thyroiditis, is the predominant cause of hypothyroidism in iodine-sufficient regions. TPOAb and thyroglobulin antibodies (TgAb) serve as markers of autoimmune thyroid involvement. The presence of these antibodies in SCH indicates an ongoing autoimmune process and increased risk of progressive thyroid dysfunction[17]. Longitudinal studies have consistently shown that TPOAb-positive individuals have a higher likelihood of developing overt hypothyroidism compared with antibody-negative individuals. However, progression is influenced by multiple genetic, environmental, and biochemical factors[18].Current guidelines recommend individualized management of SCH based on TSH concentration, age, symptoms, pregnancy status, cardiovascular risk, and thyroid antibody status. Accurate prediction of disease progression may improve patient risk stratification, monitoring strategies, and timely initiation of therapy.[19]

Therefore, this prospective cohort study was undertaken to evaluate the role of thyroid autoantibodies, particularly TPOAb and TgAb, in predicting progression from subclinical hypothyroidism to overt hypothyroidism.

Methodology:

The present study was conducted as a prospective cohort study to evaluate the role of thyroid autoantibodies in predicting the progression of subclinical hypothyroidism to overt hypothyroidism. The study was designed to assess the association between baseline thyroid autoantibody status and subsequent changes in thyroid function parameters during the follow-up period.The study was conducted in the Department of medicine at tertiary care centre.

Study Population

The study population consisted of patients diagnosed with subclinical hypothyroidism based on biochemical evidence of elevated serum TSH levels with normal free thyroxine (FT4) concentrations. Eligible participants were enrolled after obtaining informed consent and were followed to assess progression to overt hypothyroidism.

Study Methodology

Patient Recruitment

Patients presenting with abnormal thyroid function tests were screened for eligibility. Individuals fulfilling the diagnostic criteria for subclinical hypothyroidism were recruited for the study after obtaining informed consent.Baseline demographic details, clinical history, examination findings, and relevant laboratory investigations were recorded for all enrolled participants.

Diagnostic Criteria for Subclinical Hypothyroidism

Subclinical hypothyroidism was defined as:

- Serum thyroid-stimulating hormone (TSH) concentration above the laboratory reference range.
- Normal serum free thyroxine (FT4) concentration.

Patients were categorized according to their baseline TSH levels as per the study protocol.

Inclusion Criteria

1. Adults aged ≥18 years.
2. Patients diagnosed with subclinical hypothyroidism based on elevated serum TSH levels with normal FT4 levels.
3. Patients who provided written informed consent for participation and follow-up.
4. Patients who were not receiving thyroid hormone replacement therapy at the time of enrollment.

Exclusion Criteria

1. Patients with overt hypothyroidism at baseline.
2. Patients already receiving levothyroxine therapy.
3. Pregnant women or women planning pregnancy during the study period (if applicable).
4. Patients with previous thyroid surgery.
5. Patients with a history of radioactive iodine therapy.
6. Patients receiving medications known to affect thyroid function, such as lithium or amiodarone.
7. Patients with severe systemic illness affecting thyroid function tests.
8. Patients unwilling to provide consent or complete follow-up.

Baseline Assessment

Clinical Assessment

The following clinical parameters were recorded:

- Age and sex
- Duration of symptoms, if present
- Family history of thyroid disease
- History of autoimmune disorders
- Previous exposure to thyroid-related medications or treatments
- Symptoms suggestive of hypothyroidism

A complete physical examination was performed, including assessment of thyroid enlargement and other clinical signs of hypothyroidism.

Investigations

Baseline blood samples were collected from all participants for estimation of:

- Serum thyroid-stimulating hormone (TSH)
- Free thyroxine (FT4)
- Thyroid peroxidase antibodies (TPOAb)
- Thyroglobulin antibodies (TgAb)

Additional investigations, including lipid profile and other biochemical parameters, were performed wherever indicated.

Measurement of Thyroid Autoantibodies

Serum thyroid autoantibody levels were measured using standardized laboratory methods. Thyroid peroxidase antibody (TPOAb) and thyroglobulin antibody (TgAb) levels were estimated using automated methodology.

Follow-up and Outcome Assessment

All enrolled participants were followed prospectively at predefined intervals. During follow-up visits, clinical assessment and thyroid function tests were repeated.The primary outcome was progression from subclinical hypothyroidism to overt hypothyroidism, defined as:

- Persistent elevation of serum TSH levels along with
- Reduction in serum FT4 concentration
- The time to progression and association with baseline thyroid autoantibody status were evaluated.

Study Outcomes

Primary Outcome

To determine whether baseline thyroid autoantibody positivity predicted progression from subclinical hypothyroidism to overt hypothyroidism.

Secondary Outcomes

- Natural course of subclinical hypothyroidism
- Rate of progression to overt hypothyroidism
- Association between TSH, FT4, demographic factors, and disease progression
- Comparison of progression between antibody-positive and antibody-negative patients

Statistical Analysis

Data were entered into a database and analyzed using SPSS. 26 statistical software. Continuous variables were expressed as mean ± standard deviation or median (IQR). Categorical variables were expressed as frequencies and percentages.Appropriate statistical tests (Chi-square test, Student's t-test, Mann–Whitney U test) were applied as required. Multivariate analysis was performed to identify independent predictors of

progression. A p-value <0.05 was considered statistically significant.

Results:

A total of 200 patients with subclinical hypothyroidism were enrolled in the study and followed prospectively for a mean duration of 24 ± 6 months. During the follow-up period, 46 (23%) patients progressed to overt hypothyroidism, whereas 154 (77%) patients did not show progression and continued to have subclinical hypothyroidism or normalized thyroid function.The mean age of the study population was 38.6 ± 11.4 years, and females constituted the majority of participants (158, 79%). The baseline demographic and clinical characteristics of the study participants are summarized in Table 1, Figure 1

Table 1. Baseline demographic and clinical characteristics of study participants (n=200)

Variables	Total population (n=200)
Age (years), mean ± SD	38.6 ± 11.4
Female sex, n (%)	158 (79%)
Male sex, n (%)	42 (21%)
Family history of thyroid disease, n (%)	62 (31%)
Presence of goiter, n (%)	28 (14%)
Fatigue, n (%)	76 (38%)
Cold intolerance, n (%)	52 (26%)
Dry skin, n (%)	48 (24%)
Menstrual abnormality (among females), n (%)	34/158 (21.5%)

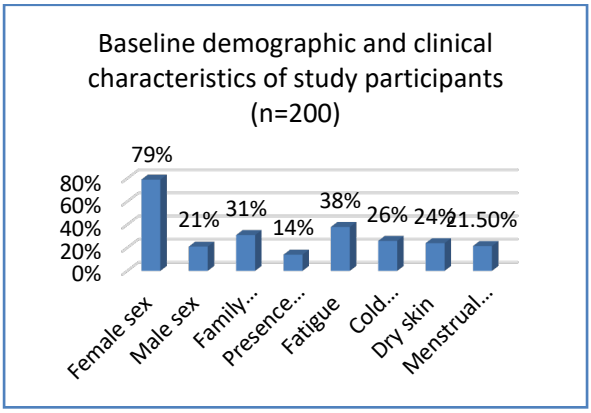

Figure 1 Baseline demographic and clinical characteristics of study participants (n=200)

Baseline Thyroid Function Profile and Autoantibody Status

At enrollment, the mean serum TSH concentration was 8.1 ± 2.3 mIU/L, while the mean FT4 level was within the normal range (1.12 ± 0.18 ng/dL). Thyroid autoantibody positivity was observed in 92 (46%) patients. TPOAb positivity was more frequent than TgAb positivity, with 78 (39%) patients positive for TPOAb and 54 (27%) patients positive for TgAb. The baseline biochemical characteristics are presented in Table 2, Figure 2

Table 2. Baseline thyroid profile and thyroid autoantibody status among study participants

Parameters	Mean ± SD / n (%)
TSH (mIU/L)	8.1 ± 2.3
FT4 (ng/dL)	1.12 ± 0.18
TPOAb positive, n (%)	78 (39%)
TgAb positive, n (%)	54 (27%)
Any thyroid antibody positive, n (%)	92 (46%)
Both TPOAb and TgAb positive, n (%)	42 (21%)

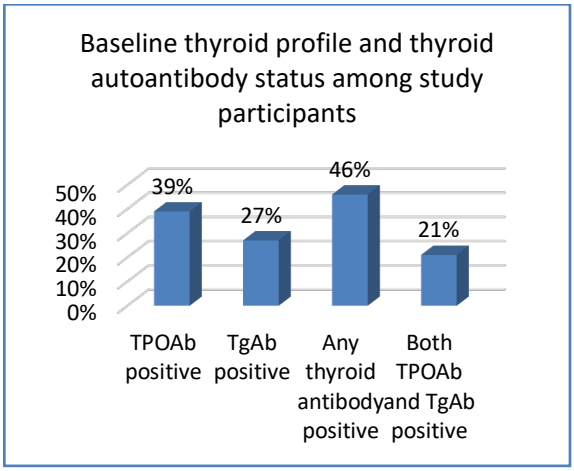

Figure 2 Baseline thyroid profile and thyroid autoantibody status among study participants

Progression of Subclinical Hypothyroidism During Follow-up

During the follow-up period, 46 patients (23%) progressed from subclinical hypothyroidism to overt hypothyroidism. The median time to progression was 18 months (range: 6–36 months).Progression

was significantly higher among patients with baseline thyroid autoantibody positivity compared with antibody-negative patients (35.9% vs 12.0%, p<0.001). The clinical outcome of participants during follow-up is shown in Table 3, Figure 3

Table 3. Clinical outcomes of patients with subclinical hypothyroidism during follow-up

Outcome	Number (%)
Progression to overt hypothyroidism	46 (23%)
Persistent subclinical hypothyroidism	118 (59%)
Normalization of thyroid function	36 (18%)
Mean follow-up duration (months)	24 ± 6

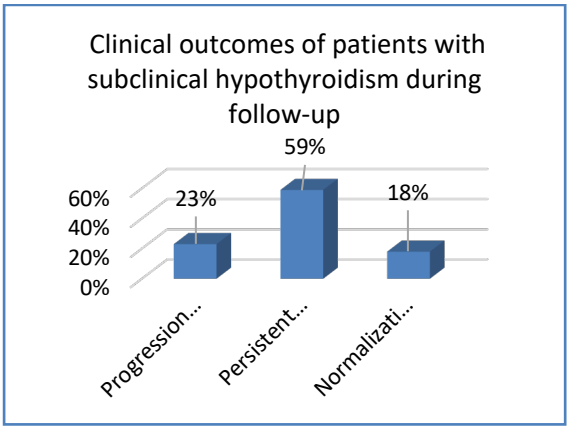

Figure 3 Clinical outcomes of patients with subclinical hypothyroidism during follow-up

Association Between Thyroid Autoantibodies and Progression to Overt Hypothyroidism

Patients who progressed to overt hypothyroidism had significantly higher baseline TSH levels and increased frequency of thyroid autoantibody positivity compared with those who did not progress. Among the 46 patients who progressed, 32 (69.6%) were TPOAb positive, whereas only 46 (29.9%) of non-progressors showed TPOAb positivity. Similarly, TgAb positivity was significantly higher among patients who progressed (47.8% vs 20.8%, p<0.001) (Table 4).

Table 4. Comparison of baseline characteristics

Variables	Progressors (n=46)	Non-progressors (n=154)	p-value
Age (years)	42.3 ± 10.8	37.5 ± 11.3	0.01
Female sex, n (%)	40 (87%)	118 (77%)	0.15
Baseline TSH (mIU/L)	10.2 ± 2.6	7.5 ± 1.9	<0.001
FT4 (ng/dL)	1.05 ± 0.16	1.14 ± 0.18	0.002
TPOAb positive, n (%)	32 (69.6%)	46 (29.9%)	<0.001
TgAb positive, n (%)	22 (47.8%)	32 (20.8%)	<0.001

Risk of Progression According to Thyroid Autoantibody Status

The risk of progression to overt hypothyroidism was significantly increased among patients with positive thyroid autoantibodies. Patients with positive TPOAb had approximately 5-fold higher odds of progression compared with antibody-negative individuals. On univariate analysis, higher baseline TSH, TPOAb positivity, TgAb positivity, and older age were significantly associated with progression. These findings are summarized in Table 5.

Table 5. Factors associated with progression from subclinical hypothyroidism to overt hypothyroidism (Univariate analysis)

Predictor	Odds Ratio (OR)	95% CI	p-value
Age >40 years	2.1	1.1–4.0	0.02
Female sex	1.8	0.8–4.1	0.12
TSH ≥10 mIU/L	3.7	1.8–7.5	<0.001
TPOAb positivity	5.4	2.7–10.8	<0.001

TgAb positivity	3.5	1.7–7.1	<0.001
Low-normal FT4	2.6	1.3–5.2	0.006

Multivariate logistic regression analysis for predictors of progression to overt hypothyroidism

Multivariate logistic regression analysis demonstrated that TPOAb positivity and baseline TSH concentration ≥10 mIU/L remained independent predictors of progression to overt hypothyroidism after adjustment for age, sex, FT4 levels, and TgAb status. Patients with positive TPOAb had significantly increased risk of progression (adjusted OR: 4.8; 95% CI: 2.2–10.3; p<0.001) (Table 6).

Table 6. Multivariate logistic regression analysis for predictors of progression to overt hypothyroidism

Variable	Adjusted OR	95% CI	p-value
Age >40 years	1.6	0.8–3.2	0.18
Female sex	1.3	0.5–3.1	0.56
TSH ≥10 mIU/L	3.2	1.5–6.8	0.002
TPOAb positivity	4.8	2.2–10.3	<0.001
TgAb positivity	1.9	0.8–4.5	0.13
Low-normal FT4	2.1	1.0–4.3	0.04

Discussion

Subclinical hypothyroidism (SCH) represents a transitional phase between normal thyroid function and overt hypothyroidism (OH), with a variable clinical course. Some patients experience spontaneous normalization of thyroid function, whereas others progress to overt disease. Therefore, identifying reliable predictors of progression is important for risk stratification and individualized management. The present prospective cohort study evaluated the role of thyroid autoantibodies, particularly thyroid peroxidase antibodies (TPOAb) and thyroglobulin antibodies (TgAb), in predicting progression from SCH to OH.In this study, 200 patients with SCH were followed prospectively for a mean duration of 24 ± 6 months. During follow-up, 46 patients (23%) progressed to OH, 118 patients (59%) remained in the subclinical hypothyroid state, and 36 patients (18%) achieved normalization of thyroid function. These findings demonstrate the heterogeneous natural history of SCH. Similar variability has been reported in previous longitudinal studies. Huber et al. [20] followed 82 women with SCH for a mean duration of 9.2 years and reported that progression to OH was strongly associated with higher baseline TSH levels and positive thyroid antibodies. The 10-year incidence of OH was 42.8% among patients with TSH 6–12 mIU/L and 76.9% among those with TSH >12 mIU/L. The progression rate observed in the present study (23% over approximately two years) was comparable with previous Indian data. Singh et al. [21] reported progression to OH in 18.97% of SCH patients followed for one year, with anti-TPO antibody positivity being the only significant predictor of progression (OR 4.58; 95% CI 1.14–18.28). Variations in progression rates among studies may be related to differences in baseline TSH levels, follow-up duration, sample size, and patient selection criteria. Thyroid autoantibody positivity was observed in 46% of participants, with TPOAb positivity in 39% and TgAb positivity in 27%. Among patients who progressed to OH, TPOAb positivity was significantly higher than in non-progressors (69.6% vs 29.9%, p<0.001). Multivariate analysis demonstrated that TPOAb positivity remained an independent predictor of progression after adjustment for age, sex, FT4 concentration, and TgAb status (adjusted OR 4.8; 95% CI 2.2–10.3; p<0.001). This finding supports the role of autoimmune-mediated thyroid destruction in the progression of SCH. Huber et al. [20] similarly demonstrated that patients with positive microsomal antibodies had a significantly higher incidence of overt hypothyroidism compared with antibody-negative individuals (58.5% vs 23.2%, p=0.03). The Whickham survey also identified thyroid antibody positivity and elevated TSH as independent predictors of future thyroid dysfunction. Baseline TSH concentration was another important determinant of progression. Patients who developed OH had significantly higher baseline TSH levels compared with non-progressors (10.2 ± 2.6 vs 7.5 ± 1.9 mIU/L; p<0.001), and TSH ≥10 mIU/L remained independently associated with progression (adjusted OR 3.2; 95% CI 1.5–6.8). These findings are consistent with previous studies demonstrating that higher initial TSH levels increase the risk of thyroid failure. Although TgAb positivity was higher among patients who progressed, it did not remain an independent predictor after adjustment for other variables. This suggests that TPOAb may have greater prognostic value as a marker of active autoimmune thyroid injury.

Increasing age was associated with progression in univariate analysis; however, it was not an independent predictor after multivariate adjustment, indicating that antibody status and biochemical parameters may have stronger predictive importance.The findings are also consistent with studies evaluating mild SCH. Díez et al. [22] and other cohorts have shown that many patients remain stable or revert to euthyroidism, particularly those with lower TSH levels and absence of thyroid antibodies. A Chinese prospective cohort of 505 patients with mild SCH reported persistent SCH in 43.8%, normalization in 49.7%, and progression to OH in 3.4%, with TPOAb positivity associated with increased risk of progression.The clinical importance of this study lies in identifying patients at higher risk who may require closer monitoring or early treatment. The strengths of the study include its prospective design, longitudinal follow-up, and evaluation of thyroid function parameters and autoimmune markers. Limitations include the single-center design, relatively short follow-up duration, and lack of thyroid ultrasonography assessment.

Conclusion

The present study concluded that thyroid autoantibodies, particularly thyroid peroxidase antibodies (TPOAb), play an important role in predicting the progression of subclinical hypothyroidism to overt hypothyroidism. Patients with positive thyroid autoantibodies and higher baseline TSH levels were found to have an increased risk of disease progression. Assessment of thyroid autoantibody status may help identify high-risk patients requiring closer follow-up and timely intervention. Therefore, thyroid autoantibody measurement can be considered an important tool for risk stratification and individualized management of patients with subclinical hypothyroidism.

Limitations

The present study had certain limitations. It was conducted at a single center with a relatively limited sample size, which may affect the generalizability of the findings. The follow-up duration may not have been sufficient to capture long-term progression in all patients with subclinical hypothyroidism. Additionally, other factors such as genetic predisposition, dietary iodine status, and thyroid ultrasonographic features were not evaluated, which may have influenced disease progression.

Ethical approval

Ethical approval for this study was obtained from the Institutional Ethics Committee prior to the commencement of the study. The study was conducted in accordance with the ethical principles outlined in the Declaration of Helsinki and applicable institutional guidelines.

Informed Consent

Written informed consent was obtained from all participants prior to their enrolment in the study.

Funding Statement

The authors received no financial support for the research, authorship, and/or publication of this article.

Conflict of Interest Declaration

The authors declare no conflict of interest.

Data Availability Statement

All data generated or analysed during this study are available from the corresponding author upon reasonable request.

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