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The Presence of Antithyroid Antibodies in Euthyroid Patients with Unexplained Infertility

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ABSTRACT

Objective: To determine the presence of antithyroid antibodies in euthyroid patients presenting with unexplained infertility. **Study Design:** Cross-sectional study. **Place and Duration of Study:** This study was conducted at the Department of Obstetrics and Gynecology, Jinnah Postgraduate Medical Centre (JPMC), Karachi, over a period of six months following approval of synopsis. **Methodology:** A total of 156 infertile women aged 20 to 45 years with confirmed euthyroid status and no identifiable cause of infertility were included through random sampling. Patients with known causes of infertility or any signs of thyroid dysfunction were excluded. After obtaining informed consent, participants underwent thyroid function tests including TSH, FT3, and FT4, along with anti-thyroid peroxidase (TPO) antibody measurement using ELISA. Data were recorded using a structured proforma and analyzed using SPSS version 26. **Results:** Out of 156 euthyroid women with unexplained infertility, 29 (18.6%) tested positive for TPO antibodies while 127 (81.4%) were negative. TPO antibody positivity was significantly associated with infertility duration greater than two years (p=0.03) and history of early pregnancy loss (p=0.02). Mean age, TSH, FT4, and FT3 levels showed no statistically significant differences between the two groups. **Conclusion:** A considerable proportion of euthyroid women with unexplained infertility were found to have TPO antibody positivity, suggesting an underlying immunological mechanism. Routine screening for antithyroid antibodies should be considered in the diagnostic workup of unexplained infertility to enable early intervention and improve reproductive outcomes.

INTRODUCTION

Infertility is a problem that affects 10-15% of individuals and couples worldwide and causes emotional, social, and psychological burden, especially in women living in developing countries like Pakistan. Unexplained infertility (otherwise known as the inability to conceive after trying for 1 year, without any obvious cause [3]) is known as one of the potential reasons couples have trouble getting pregnant and is responsible for nearly 15–30% of cases of infertility globally [1].

Thyroid hormones have an essential role in female reproductive physiology, affecting ovulation, implantation, and the maintenance of pregnancy. Overt and subclinical thyroid dysfunctions impair fertility 1 but recent research suggests that even in women with normal thyroid function (euthyroid) the presence of antithyroid antibodies (ATAs), including anti-thyroid peroxidase (TPO-Ab), might be related to subfertility 2 3.

ATAs reflect thyroid autoimmunity and are frequently present in euthyroid women of reproductive age[4]. Several reports have found association between these antibodies and poor reproductive outcome such as miscarriage, failure of implantation and poor IVF outcome[5,6]. Their baseline appearance may be a sign of an immune dysfunction that affects the endometrial receptivity and oocyte quality in a negative way[7].

Even with normal hormonal profiles and ovulation, euthyroid women with idiopathic infertility also present higher ATAs titters than fertile women[8,9]. These antibodies could have either local inflammatory action or interfere with folliculogenesis and cause subtle, still clinically relevant reproductive disorders[10]. A 2022s study from Egypt found ATA positivity in nearly 18.6% of euthyroid women with unexplained infertility compared with 0% in the controls[11]. However, the information on South Asian populations is limited.

Owing to this paucity of regional literature, this study has been undertaken to determine the proportion of euthyroid females (20–45 years) presenting with unexplained infertility who are seropositive for antithyroid antibodies. Prompt recognition of this immunologic marker could influence possible therapeutic options, namely immunomodulation, or increased vigilance for the assisted reproductive technologies[12].

METHODOLOGY

This was a cross-sectional study carried out over a span of six months from Jan 2024 to September 2024 at the Department of Obstetrics and Gynaecology, Jinnah Postgraduate Medical Centre (JPMC), Karachi. Its aim was to assess the prevalence of antithyroid antibodies in euthyroid women with unexplained infertility. Total 156 subjects were randomly sampled for the study. The sample size was determined with 5% of error, a 95% confidence interval and an estimated prevalence of subclinical hypothyroidism (11.4%) reported in previous studies.

Strict inclusion and exclusion criteria were used to select participants. The selection criteria for patients were infertile female patients aged between 20 and 45 years, without known reason for infertility and with euthyroid state. Exclusion criteria were women with known causes of infertility (e.g., tubal blockage, endometriosis, ovulatory disturbances), signs or symptoms of thyroid dysfunction, women being outside the age range, fertile women. The study was approved by the institutional review board and written informed consent was provided by all participants prior to enrolment.

A detailed history and clinical evaluation was performed in all the patients. Hormonal profile: Serum levels of luteinizing hormone (LH), follicle-stimulating hormone (FSH), prolactin, estrogen, and progesterone were measured. Before the experiment, TSH, free thyroxine (FT4), and free triiodothyronine (FT3) were measured to ensure euthyroidism. Moreover, anti-thyroid peroxidase (TPO) antibody was determined by enzyme-linked immunosorbent assay (ELISA). Blood samples were drawn, clotted, centrifuged, and stored between 2–8°C for maximum of five days, and –20°C for longer periods, before analysis.

A predesigned proforma was used to document all clinical and laboratory data. In order to reduce bias and confounding, all of the subjects were prescreened and excluded using rigorous clinical and laboratory criteria. Statistical analysis was performed using SPSS, version 26. Continuous variables are described as the mean (SD) and categorical variables as number and percentage. Chi-square test for post-stratification was used to compare results of antibody positive with antibody-negative groups. P value of <0.05 was considered significant.

RESULTS

The study participants: a hundred fifty-six cases of women aged from 20 to 45 years of unexplained infertility and the confirmed absence of disturbances in the thyroid function. Participants were on average 32.6 ± 5.1 years old. Of these, anti-thyroid peroxidase (TPO) antibodies were positive in 29 women (18.6%), whereas in 127 women (81.4%) the antibodies tested negative (Table 1).

All participants had normal thyroid function tests, with mean TSH levels of 2.1 ± 0.8 mIU/L, FT4 levels of 17.3 ± 2.6 pg/mL, and FT3 levels of 5.1 ± 1.1 pg/mL, confirming euthyroidism. No participants exhibited clinical signs or symptoms of thyroid dysfunction during the study period.

A comparative study, according to two groups of pre-Tx treatment (antibody-positive and antibody-negative), revealed that TPO-Ab presence was more frequent in women with a long period of infertility (>2 years) (p=0.03). Similarly, there was a statistically significant relationship between serum antibody positivity and prior history of early pregnancy loss or biochemical failure of pregnancy among participants without evidence of clinical thyroid disease (p=0.02) (Table 2).

The distribution of TPO-Ab status is presented in Graph 1, with an obviously increased ratio of TPO-Ab negative (but WNL) subjects, although a significant proportion remained TPO-Ab positive despite normal thyroid activity. The data indicate an increased prevalence of thyroid autoimmunity among unexplained sub fertile women, thus supporting the need for thyroid autoantibodies determination in clinical infertility.

Table 1

Frequency Distribution of TPO Antibody Status

TPO Antibody Status	Number of Patients	Percentage (%)
Positive	29	18.6
Negative	127	81.4

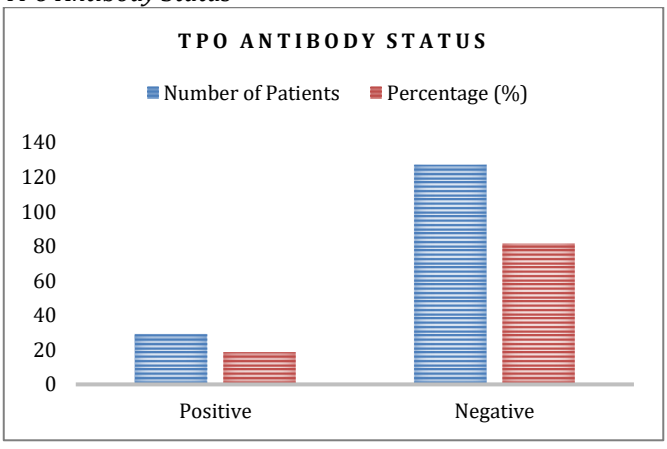
Table 2

Comparison of Clinical Parameters Between Antibody-Positive and Antibody-Negative Groups

Parameter	TPO-Ab Positive (n=29)	TPO-Ab Negative (n=127)	p-value
Mean Age (years)	33.2	32.5	0.12
Mean TSH (mIU/L)	2.3	2.0	0.08
Mean FT4 (pg/mL)	17.1	17.4	0.36
Mean FT3 (pg/mL)	5.0	5.1	0.52
Infertility Duration >2 years (%)	62.1%	41.7%	0.03*
History of Pregnancy Loss (%)	44.8%	25.2%	0.02*

Figure 1

TPO Antibody Status



DISCUSSION

The results of our study bring to light a statistically significant prevalence of anti-thyroid peroxidase (TPO) antibodies in euthyroid women with unexplained infertility and all its consequences and indirectly acknowledge an immunological cause of infertility apart from the overt state of thyroid diseases. In our population, individuals with positive TPO-Ab represented 18.6%, a

proportion concordant, although somewhat lower than prevalence estimates from other countries.

Our results support the suggestion for thyroid autoimmunity as contributing factor in female subfertility, even with euthyroid thyroid hormone levels. Several possible models have been suggested. It is thought that antithyroid antibodies, could interfere with the ovarian and endometrial function directly through the mechanism of autoimmune mediated inflammation or indirectly, altering the profiles of cytokines and endometrial receptivity[13]. TPO-Ab is also reported to have adverse effects on implantation and early pregnancy loss in euthyroid women who are receiving ART[14].

A recent study by Medenica et al. highlighted the relationship of TPO Ab with decreased pregnancy rates in IVF treatment with an over –threefold decrease in both clinical pregnancy and live birth (LB) rates among TPO-Ab positive women[15]. This reinforces the importance of thyroid autoantibody testing, even without clinical hypothyroidism. Similarly, Zhang et al. described a higher rate of unexplained infertility and miscarriage in women with elevated TPOAb levels, thus corroborating the reproductive sequelae of subclinical thyroid autoimmunity[16].

Another possible reason of our results is based on the commentary of shared autoimmunity. It has been shown that women with one organ-specific autoimmunity, such as autoimmune thyroid disease, are at higher risk of second organ-specific autoimmunity related with fertility (i.e. antiphospholipid syndrome or ANA positivity) [17]. These immune changes may affect folliculogenesis, endometrial receptive or early embryonic development.

The inferences form our study are important in South Asian setting which does not have a prevalent practice of routine TPO-Ab screening in infertility workup. Integration of thyroid autoantibody testing into initial work-up may identify patients who are at risk and benefit from immunomodulatory therapy or close monitoring when attempting pregnancy[18].

Nevertheless, our study has limitations. It was carried out at one referral center only, limiting generalizability. Moreover, we did not evaluate the long-term effects of TPO-Ab positivity including pregnancy and the rate of miscarriage. Larger longitudinal studies with follow-up data are needed to clarify causality and the effectiveness of intervention.

CONCLUSION

This study indicates a high prevalence of anti-TPO antibodies in euthyroid women with unexplained infertility, indicating a possible association of thyroid autoimmunity with infertility even in the absence of autonomic thyroid diseases. The results enable us to recommend the routine testing of TPO antibody in infertile women at risk of immunological factors and may provide an opportunity for more personalized treatment if appropriate. Early detection of thyroid autoimmunity could lead to the optimization of reproductive success and minimize psychological distress in these couples. More large multicentre and prospective studies are suggested to evaluate the effect of TPOAb on the pregnancy rate and treatment results in this population.

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