ASSOCIATION BETWEEN ANTI-THYROID PEROXIDASE ANTIBODY AND RECURRENT MISCARRIAGE

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ABSTRACT

Background

Thyroid disease is the second most commonly affected disease in childbearing women, after diabetes, and thyroid autoimmunity in pregnancy has been connected with adverse pregnancy outcomes such as miscarriage, recurrent miscarriage, preterm birth, and low intelligence.

Objectives

The study seeks to determine whether there is a significant correlation between anti-thyroid peroxidase antibodies and unexplained recurrent miscarriages.

Patients and Methods

A Case-control study was conducted on 124 participants, 62 women who had experienced unexplained recurrent miscarriage and 62 healthy women without a history of miscarriage, from 11.9.2021 until 10.6.2022 in the outpatient and emergency department of Sulaimani Maternity Teaching Hospital and some private clinics in Sulaimani City/Kurdistan Region /Iraq, and screening for TSH and anti–thyroid peroxidase antibody are done for both groups.

Results

The prevalence of positive anti-thyroid peroxidase antibody in women with recurrent miscarriage was 19.4 %, while in women without miscarriage was 6.5% (which is considerably higher in cases than in women without recurrent miscarriage with a p-value of 0.03 and an odd ratio of 3.48 (95% CI; 1.06-11.48).

Conclusion

There is a statistically significant relationship between anti-thyroid peroxidase antibodies and recurrent miscarriage. We recommend screening for TSH and thyroid antibodies for women with recurrent miscarriages and further studies on the effect of levothyroxine therapy for euthyroid women with antibody positive.

Keywords: *Recurrent miscarriage, Auto-immune thyroid disease (AITD), anti-thyroid peroxidase antibody (anti-TPO antibody).*

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INTRODUCTION

In general, miscarriage is the loss of a pregnancy that can occur from the onset of conception until 24 weeks of gestation; when a miscarriage happens consecutively three or more times, it is called a recurrent miscarriage. It is worth mentioning that 1% of couples suffer from recurrent miscarriages [1]. Studies has shown that various factors contribute to recurrent miscarriage, such as age, smoking, and obesity that are categorised as epidemiological reasons, antiphospholipid syndrome, genetic factor, anatomical aspects like a congenital uterine anomaly and cervical weakness, inherited thrombophilic factor, endocrine diseases like diabetes mellitus and thyroid disease [1]. However, the majority of miscarriage causes are not diagnosed. 50% or more of recurrent miscarriages occur for no apparent reason. Testing for parental karyotypes, hysterosalpingography or hysteroscopy, and antiphospholipid antibodies are performed in these couples to identify the reason for recurrent miscarriage; however, if all of the tests are negative, the miscarriage still occurs [2].

Many studies have been conducted to diagnose the cause of recurrent miscarriages in couples. One of these studies is thyroid peroxidase antibody positive in women with normal thyroid hormone levels and unexplained recurrent miscarriages. After diabetes, thyroid disorder is the second most commonly diagnosed endocrine condition in childbearing women [3]. The most frequent cause of thyroid disease is thyroid autoimmunity, which is the existence of anti-thyroid antibodies, specifically thyroid peroxidase and thyroglobulin [4]. The prevalence of thyroid autoimmunity causes negative pregnancy outcomes such as miscarriage, recurrent miscarriages, preterm birth, and low IQ [5]. Thyroid antibodies can be found in the serum of euthyroid women in about 5-20% and women with a history of recurrent miscarriage (17%-33%) [6].

Thyroid peroxidase is a crucial enzyme for iodinating tyrosine residues to form thyroid hormones. Thyroglobulin is a glycoprotein that contains these tyrosine residues. Thyroglobulin is generated by thyroid cells and stored in the thyroid colloid [7].

The mechanism underlying the relationship between recurrent miscarriage and anti-thyroid peroxidase antibody is still not detected precisely. However, there is a hypothesis that explains this association. The first hypothesis stated that thyroid peroxidase antibody reflects a different level of auto-immune process that causes subfertility or pregnancy loss. In contrast, the second hypothesis stated that the association with miscarriage is attributable to a subtle thyroid hormone deficiency [8].

Euthyroid individuals with a serum TSH level of more than 3 ml U/L have higher TPO-Ab titers and a higher rate of changing to overt hypothyroidism in the future, even though they have normal TSH [9].

Thyroxine therapy might be beneficial in preventing miscarriage and increasing the term delivery rate for women with a history of unexplained recurrent miscarriage and anti-thyroid peroxidase antibody positive before and during conception [10]. However, in contrast to these, other studies have found that thyroxine treatment is ineffective in women with a history of unexplained recurrent miscarriage and normal thyroid function test with thyroid antibody positive [11–13].

PATIENTS AND METHODS

This case-control study included 124 participants; the sample size estimation is calculated depending on the prevalence of recurrent miscarriage among reproductive-age women who relied on previous studies.

Moreover, it divided into two groups the first group of 62 cases, which included women who had three or more consecutive miscarriages before 22 weeks of gestation with or without live birth, and the second group as control of 62 healthy fertile women of multigravida who have three or more term live birth without complication such as preterm labour, premature rupture membrane, stillbirth, preeclampsia, gestational diabetes, any chronic disease and without a history of miscarriage.

Data collection started after approval of the research protocol by the research protocol ethics committee / Kurdistan Board of Medical Specialties / Ministry of Higher Education and Scientific Research /Kurdistan Region Government - İraq, with approval number (1182) at 11.9.2021 until 10.6.2022 in outpatient and emergency department of Sulaimani Maternity Teaching Hospital and some private clinics in Sulaimani City/Kurdistan Region /Iraq.

One hundred twenty-four women participated in the research after an investigation resulted in seven participants in a case group and six in a control group being excluded from the study because they had TSH >4.27 and were diagnosed with overt hypothyroidism; another participant was put in place of them.

Exclusion criteria for both groups were any woman with a history of a uterine anomaly, fibroid, cervical weakness, auto-immune diseases like antiphospholipid syndrome, overt thyroid disease (defined as TSH <0.27 OR >4.27), any chronic disease like hypertension, diabetes, or any other endocrine disease like PCOS and hyperprolactinemia either physiological or medical hyperprolactinemia and chromosomal abnormality of both parents.

Samples were collected from 124 women who voluntarily participated in the study. Their consent is obtained verbally, and they are informed that they can refuse participation in this research.

After Demographic data and BMİ full history taken from participants, past obstetric gravity, parity, and some miscarriages, it was a first-trimester miscarriage or second-trimester miscarriage (first trimester: from conception until 12+6 weeks and second trimester: from 13 weeks until 22+6 weeks) gynecological history, past surgical history, drug history, and their health record laboratory investigation of cases with recurrent miscarriage are seen and clinical examination done to exclude anyone with past medical illness or abnormal BMI or any other exclusion criteria, all data was put in questioner form example is put in Figure(1)and data saved after the entrance to excel in a google document.

Furthermore, after agreement from participants, one cc peripheral blood samples were collected from all women and the serum was separated by centrifuge and kept in a low-temperature freezer FROİLABO at – 36-degree Celsius for sample protection until the day, which is taken for a lab to investigate for titer of anti-thyroid peroxidase antibody and thyroid-stimulating hormone in serum of cases and control women which are done in Smart Medical Building /Laboratory Department.

Elecsys TSH kits measured TSH and Anti-thyroid peroxidase antibody titers and Elecsys Anti-TPO from Roche diagnostic kits, Germany... Using electrochemiluminescence (ECL) (Cobas 6000.e601 module) analyzers series, HITACHI. Both of them had run through and were guaranteed by laboratory quality control. Therefore, the titer of thyroid peroxidase antibody of more than 35 IU/ml is regarded as positive as laboratory reference range values were analyzed. Also, the normal value for the thyroid-stimulating hormone test was (0.27-4.27mIU/mL).

The statistical analysis was performed by the SPSS program, version 21 (IBM SPSS Statistical Package for the Social Sciences). Compliance of quantitative random variables with the Gaussian curve (normal distribution) was analysed using Kolmogorov-Smirnov and Shapiro – Wilk tests. The data presented in tabular forms shows the frequency and relative frequency distribution of different variables among both groups. In addition, chi-square tests were used to compare the categorical data between these two groups of study participants (cases and controls) concerning different variables. Variables showed to be normally distributed continuous quantitative variables and described by mean and SD (standard deviation).

The statistical significance of the difference in mean between the two groups (cases and controls) was assessed using an independent sample t-test. Non-normally distributed quantitative variables such as TSH and Anti-TPO antibody titer were described by the median in addition to the mean. In such conditions, the median and interquartile ranges (IQR) were used for compared groups. The difference in the Median (IQR) of the two groups was assessed by a non-parametric test (Mann- Whitney). P values of 0.05 were used as a cutoff point for the significance of statistical tests.

RESULT

A total number of 124 participants were assessed for TSH and anti-TPO titer,62 of them represented the case group that has a recurrent miscarriage, 40 of them had just first-trimester miscarriage, and 22 of them had first and second-trimester miscarriage and 62 them represented control group without miscarriage, all of them were euthyroid (TSH between 0.2-4.27mlU/L).

The sociodemographic status for both groups and a comparison of characteristics are shown in (Table 1).

The mean age of both groups was 32.59 ± 5.81 with an age range of 18 - 47 years and no significant difference between the age of both groups (33.11 ± 6.37 VS 32.06 ± 5.19) p-value;0.32. Among all participants, 76.6 % lived inside the city centre; others were around the city; 61.3% were housewives. Overall, 97 were Kurdish, the remaining 23 were Arabic, and no one was a smoker or alcoholic.

Of the 124 participants, 16 had positive anti-TPO antibody titer (12.9 %); 12 were in the case group, and four were in the control group. Among those 12 cases of positive anti-TPO in the recurrent miscarriage group, 9 of them were those with first-trimester miscarriages,

and 3 of them were those were they have first and second-trimester miscarriages.

The prevalence of positive anti-TPO antibodies in women with recurrent miscarriage was 19.4 %, while in women without miscarriage was 6.5% (which is considerably higher in cases than the control group with a p-value of 0.03) and an odd ratio of 3.48 (95% CI;1.06-11.48) (Table 2).

There was no significant association between age group and thyroid peroxidase antibody in both groups, as shown in (Table 3).

The median of TSH in women with anti-TPO positive was higher than in women with anti -TPO negative in the recurrent miscarriage group (3.15 VS 1.75) p-value 0.02, (Figure4).

The Median of TSH in cases including anti-TPO positive and negative was (2.1) while in the control group was (1.8), with a p-value of 0.15; hence there is no statistically significant difference in Median (or average) TSH between cases and controls (irrespective of anti-TPO).

Table 1. Sociodemographic status for both groups and comparison of characteristics.

		Cases	Controls	Total	P value
Age (Years)	**Mean ± SD**	33.11 ± 6.37	32.06 ± 5.19	32.59 ± 5.81	0.32
	18 - 27	14 (22.6%)	16 (25.8%)	30 (24.2%)	0.42
	28 - 37	32 (51.6%)	36 (58.1%)	68 (54.8%)	
	38 - 47	16 (25.8%)	10 (16.1%)	26 (21.0%)	
Occupation	**Housewife**	45 (72.6%)	31 (50.0%)	76 (61.3%)	0.01
	Employed	17 (27.4%)	31 (50.0%)	48 (38.7%)	
Residency	**Rural**	10 (16.1%)	19 (30.6%)	29 (23.4%)	0.06
	Urban	52 (83.9%)	43 (69.4%)	95 (76.6%)	
Gravida	**0 - 2**	2 (3.2%)	1 (1.6%)	3 (2.4%)	< 0.001
	3 – 4	22 (35.5%)	61 (98.4%)	83 (66.9%)	
	5 or more	38 (61.3%)	0 (0%)	38 (30.6%)	
Para	**nulliparous**	19 (30.6%)	0 (0%)	19 (15.3%)	< 0.001
	1 – 3	40 (64.5%)	60 (96.8%)	100 (80.6%)	
	Four or more	3 (4.8%)	2 (3.2%)	5 (4.0%)	
Previous NVD	**None**	33 (53.2%)	17 (27.4%)	50 (40.3%)	< 0.001
	One - two	21 (33.9%)	12 (19.4%)	33 (26.6%)	
	Three - four	8 (12.9%)	33 (53.2%)	41 (33.1%)	
Previous C/S	**None**	43 (69.4%)	34 (54.8%)	77 (62.1%)	< 0.001
	One - two	18 (29.0%)	13 (21.0%)	31 (25.0%)	
	Three	1 (1.6%)	15 (24.2%)	16 (12.9%)	
Total		**62 (100%)**	**62 (100%)**	**124 (100%)**	

Table 2. Comparison of anti-TPO results in study participants (Cases and controls)

		Cases	Controls	Total	P value
Anti TOP titer	**Positive**	12 (19.4%)	4 (6.5%)	16 (12.9%)	0.03
	Negative	50 (80.6%)	58 (93.5%)	108 (87.1%)	
Odds ratio (95% CI)		3.48 (1.06 – 11.48)			0.03
Total		**62 (100%)**	**62 (100%)**	**124 (100%)**	

Table 3. Association between age group and thyroid peroxidase antibody titer in both groups.

Age (years)	Anti- TPO Antibody				Total
	Cases		Controls		
	Positive	Negative	Positive	Negative	
18 - 27	1	13	1	15	**30**
28 - 37	9	23	2	34	**68**
38 - 47	2	14	1	9	**26**
P value	0.18		0.88		

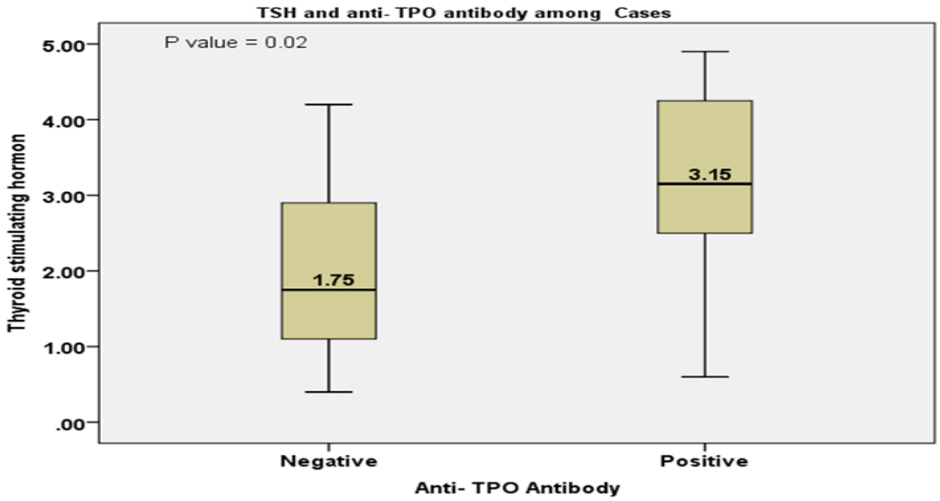

Figure 1. Compares the Median of TSH in ant- TPO positive and negative cases of recurrent miscarriage.

DISCUSSION

Many families and communities bear a heavy physical, emotional, and financial burden due to recurrent miscarriages. Therefore, proper management and treatment are needed; however, according to the 2011 RCOG guideline [(1)], 75 percent of women with unexplained recurrent miscarriages have an excellent prognosis for a successful future pregnancy with only support and no medication [(1)].

Association between anti-TPO antibodies and recurrent miscarriage has been reported by many systematic reviews and meta-analyses, in a meta-analysis done in 2011 by Shakila Thangaratinam et al.[(14)] demonstrated that; In women with normal thyroid function and thyroid autoantibodies, miscarriage risk is expected to become three times higher and the risk of preterm birth is increased twice.

In our study prevalence of thyroid antibody positive in recurrent miscarriage, women were 19.4%. This prevalence was also in the same range found in other studies varying from 17-33% [(6)].

In a case-control study done by Ticconi et al. in 2011 [(15)], the prevalence of thyroid antibodies in recurrent miscarriage was 28.7%, and the prevalence in women without abortion was 13%. In our study prevalence of anti-TPO antibodies was 6% in the control group without miscarriage which is in the same range as the total population range (6-20%) [(6)]. Ticconi et al. in 2011 [(15)] investigated anti-TPO antibodies, anti-thyroglobulin antibodies, and anti-thyroid receptor antibodies, but we did an analysis just for anti-TPO antibodies.

Thyroid peroxidase antibody is considered the most sensitive marker for detecting thyroid autoimmunity, with thyroid peroxidase antibody being the most prevalent and commonly tested for [(6)].

In 2006 Ghafoor et al. [16] did a cross-sectional analytical study on 1500 pregnant women for the role of thyroid antibodies and the outcome of pregnancy. He concluded that; thyroid auto-immune illness among pregnant women may lead to low –birth-weight of neonates and increased miscarriage frequency in pregnant women.

In contrast to our study in 1998, M S Esplin [17] did a study and found that circulating thyroid autoantibodies are not significantly different in women with recurrent miscarriage and fertile women. He also states that Testing for anti-thyroid antibodies is unnecessary for diagnosing the reason behind recurrent miscarriage.

As women get older, the quality and the number of oocytes decrease. Moreover, maternal age and number of previous miscarriages are two distinct risk factors for a subsequent miscarriage.

Women at age 35 and above are more likely to have miscarriages; in our study, there was no statistically notable difference between the ages of both groups, and we have discovered that there is no association between anti-TPO antibodies and age groups (Table 2); this is also similar to the meta-analysis of ten studies that showed, there was no significant difference between age groups and anti-thyroid peroxidase antibody [14].

Different studies have concluded that obesity increases the likelihood of sporadic and recurrent miscarriage [1]; our study excluded any women with abnormal BMI.

Another finding in our study was that the Median of TSH in women with anti-TPO positive was significantly higher than in women with anti -TPO negative in the recurrent miscarriage group (3.15 VS 1.75); this is also similar to the finding of the meta-analysis, which S. Thangaratinam does in 2011 [14] so our study more support the hypothesis that is saying the association between anti-TPO with pregnancy loss is due to the subtle deficiency in thyroid hormone.

Furthermore, till now American Thyroid Association Guidelines 2011 and the Endocrine Society Guidelines 2012 dose not recommended universal TPO-Ab testing in the evaluation of women with recurrent miscarriage or thyroxine treatment of euthyroid TPO-Ab-positive women [18].

In conclusion, there is a statistically significant association between anti-TPO antibodies and recurrent miscarriage, and we recommend TSH and thyroid antibody investigation for women with recurrent miscarriages. Also, the European Society for Human Reproduction and Embryology Recurrent pregnancy loss recommends thyroid screening with TSH and Anti TPO antibody. Moreover, we recommend further studies on the effect of levothyroxine therapy for euthyroid women with anti-thyroid peroxidase antibody positive.

In our study, we just included women with normal thyroid function tests, also clinically normal, and we excluded other Couse of recurrent miscarriage, for example, antiphospholipid syndrome, and it was complicated to find these cases in which they have an unexplained recurrent miscarriage.

In couples who have experienced consecutive pregnancy loss, chromosomal abnormalities of the embryo account for 30–57% of further miscarriages [1]; chromosomal analysis of the embryo which was miscarried was not done by all 62 recurrent miscarriage participants as it is very costly and not applicable for every miscarriage this was one of limitation of our study, another limitation was the small sample size, and all women were from a developing country.

REFERENCES

1. No RG. The investigation and treatment of couples with recurrent first-trimester and second-trimester miscarriage. RCOG: London, UK. 2011 Apr.

2. American College of Obstetricians and Gynecologists. ACOG practice bulletin. Management of recurrent pregnancy loss. Number 24, February 2001. (Replaces Technical Bulletin Number 212, September 1995). American College of Obstetricians and Gynecologists. Int J Gynaecol Obstet. 2002 Aug;78(2):179–90.

3. Ramprasad M, Bhattacharyya SS, Bhattacharyya A. Thyroid disorders in pregnancy. Indian Journal of Endocrinology and Metabolism [Internet]. 2012 [cited 2022 Jul 16];16(Suppl 2): S167. Available from: /PMC/articles/PMC3603018/

4. Xie J, Jiang L, Sadhukhan A, Yang S, Yao Q, Zhou P, et al. Effect of anti-thyroid antibodies on women with recurrent miscarriage: A meta-analysis. American Journal of Reproductive Immunology [Internet]. 2020 Jun. 1 [cited 2022 Jul. 16];83(6):e13238.

5. Chang DLF, Pearce EN. Screening for maternal thyroid dysfunction in pregnancy: A review of the clinical evidence and current guidelines. Journal of Thyroid Research. 2013;2013.

6. Dhillon-Smith RK, Boelaert K, Jeve YB, Maheshwari A, Coomarasamy A, Royal College of Obstetricians and Gynaecologists. Subclinical hypothyroidism and anti-thyroid autoantibodies in women with subfertility or recurrent pregnancy loss: Scientific Impact Paper No. 70 June 2022. BJOG: An International Journal of Obstetrics & Gynaecology. 2022 Jun. 28.

7. Matalon ST, Blank M, Ornoy A, Shoenfeld Y. The Association between Anti-Thyroid Antibodies and Pregnancy Loss. American Journal of Reproductive Immunology [Internet]. 2001 Feb 1 [cited 2022 Jul 16];45(2):72–7.

8. Vissenberg R, Manders VD, Mastenbroek S, Fliers E, Afink GB, Ris-Stalpers C, et al. Pathophysiological aspects of thyroid hormone disorders/thyroid peroxidase autoantibodies and reproduction. Human Reproduction Update [Internet]. 2015 May 1 [cited 2022 Jul 16];21(3):378–87.

9. Abdulateef DS, Mahwi TO. Assessment of subclinical hypothyroidism for a clinical score and thyroid peroxidase antibody: A comparison with euthyroidism grouped by different thyroid-stimulating hormone levels. Asian Biomedicine. 2019 Dec 30;13(3):85–93.

10. Mosaddegh MH, Ghasemi N, Jahaninejad T, Mohsenifar F, Aflatoonian A. Treatment of recurrent pregnancy loss by Levothyroxine in women with high Anti-TPO antibody. Iranian Journal of Reproductive Medicine [Internet]. 2012 Jul [cited 2022 Jul. 16];10(4):373.

11. Van Dijk MM, Vissenberg R, Fliers E, van der Post JAM, van der Hoorn MLP, de Weerd S, et al. Levothyroxine in euthyroid thyroid peroxidase antibody positive women with recurrent pregnancy loss (T4LIFE trial): a multicentre, randomised, double-blind, placebo-controlled, phase 3 trial. The Lancet Diabetes & Endocrinology. 2022 May 1;10(5):322–9.

12. Lata K, Dutta P, Sridhar S, Rohilla M, Srinivasan A, Prashad GR v, et al. Thyroid autoimmunity and obstetric outcomes in women with recurrent miscarriage: a case–control study. Endocrine Connections [Internet]. 2013 Jun 1 [cited 2022 Jul 16];2(2):118–24.

13. Dhillon-Smith RK, Middleton LJ, Sunner KK, Cheed V, Baker K, Farrell-Carver S, et al. Levothyroxine in Women with Thyroid Peroxidase Antibodies before Conception. New England Journal of Medicine [Internet]. 2019 Apr 4 [cited 2022 Jul 16];380(14):1316–25.

14. Thangaratinam S, Tan A, Knox E, Kilby MD, Franklyn J, Coomarasamy A. Association between thyroid autoantibodies and miscarriage and preterm birth: a meta-analysis of the evidence. BMJ [Internet]. 2011 May 14 [cited 2022 Jul. 16];342(7806).

15. Ticconi C, Giuliani E, Veglia M, Pietropolli A, Piccione E, di Simone N. Thyroid Autoimmunity and Recurrent Miscarriage. American Journal of Reproductive Immunology [Internet]. 2011 Dec 1 [cited 2022 Jul 16];66(6):452–9.

16. Ghafoor F, Mansoor M, Malik T, Malik MS, Khan AU, Edwards R, et al. Role of thyroid peroxidase antibodies in pregnancy outcome. J Coll Physicians Surg Pak. 2006 Jul;16(7):468–71.

17. Esplin MS, Branch DW, Silver R, Stagnaro-Green A. Thyroid autoantibodies are not associated with recurrent pregnancy loss. American Journal of Obstetrics and Gynecology. 1998 Dec 1;179(6):1583-6.

18. Mehran L, Tohidi M, Sarvghadi F, Delshad H, Amouzegar A, Soldin OP, et al. Management of thyroid peroxidase antibody euthyroid women in pregnancy: Comparison of the American thyroid association and the endocrine society guidelines. Journal of Thyroid Research. 2013;2013.