

Effects of Levothyroxine on Pregnant Women With Subclinical Hypothyroidism, Negative for Thyroid Peroxidase Antibodies

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Context: Currently, there is no consensus on universal thyroid screening and levothyroxine (LT₄) treatment of pregnant women with subclinical hypothyroidism (SCH) who are negative for thyroid peroxidase antibody (TPOAb⁻).

Objective: We aimed to evaluate the benefits of LT₄ treatment on pregnancy outcomes in SCH-TPOAb⁻ women.

Design: This study was conducted within the framework of the Tehran Thyroid and Pregnancy Study. A single-blind randomized clinical trial was undertaken in pregnant women who were SCH-TPOAb⁻.

Setting: Prenatal care centers of the Shahid Beheshti University of Medical Sciences.

Patients: Using the thyrotropin (TSH) cut point of 2.5 mIU/L, 366 SCH-TPOAb⁻ and 1092 euthyroid TPOAb⁻ women were recruited.

Intervention: SCH-TPOAb⁻ women were randomly assigned to two groups: group A (n = 183) who were treated with LT₄ and group B (n = 183) who received no treatment. A total of 1,028 euthyroid TPOAb⁻ women served as the control group (group C).

Main Outcome Measure: The primary outcome was the rate of preterm delivery.

Results: Using the TSH cutoff of 2.5 mIU/L, no significant difference in preterm delivery was observed between groups A and B [relative risk (RR): 0.86; 95% confidence interval (CI): 0.47 to 1.55; P = 0.61]. However, log-binomial model analysis based on a cut point of 4.0 mIU/L demonstrated a significantly lower rate of preterm delivery in LT₄-treated women compared with those who received no treatment (RR: 0.38; 95% CI: 0.15 to 0.98; P = 0.04).

Conclusions: Despite no beneficial effect of LT₄ therapy in reducing preterm delivery in SCH-TPOAb⁻ women with a TSH cut point of 2.5 to 4 mIU/L, LT₄ could precisely decrease this complication using the newly recommended cutoff ≥ 4.0 mIU/L. (*J Clin Endocrinol Metab* 103: 926–935, 2018)

Despite well-defined clinical guidelines on the treatment of pregnant women with an overt hypothyroid (1), endocrinologists have not yet reached a consensus on whether to treat women with subclinical hypothyroidism (SCH) who are positive or negative for thyroid peroxidase antibody (TPOAb⁺ or TPOAb⁻). The Endocrine Society recommends therapy in all pregnant women presenting with SCH, irrespective of autoimmunity status (either TPOAb⁺ or TPOAb⁻) (2), whereas the American Thyroid Association (ATA) supports treatment only for a specific subgroup of women with SCH who are TPOAb⁺ (SCH-TPOAb⁺) (1).

In addition, the thyrotropin (TSH) threshold for the definition of SCH is controversial and warrants further population-based studies. With respect to questions surrounding the previously recommended TSH cut point of 2.5 mIU/L (3, 4), a higher cutoff value of 4.0 mIU/L was recently proposed in the ATA's 2017 revisions (1).

In most, but not all, observational studies, isolated SCH has been linked to several fetomaternal complications including miscarriage, hypertension, placental abruption, and preterm birth (5–7). Although TPOAb status was not reported in many of these studies, prospective data suggest higher rates of adverse outcomes in both SCH-TPOAb⁺ and SCH-TPOAb⁻ pregnant women (8, 9). Data regarding the treatment benefits in such women are also inconclusive. To date, five randomized trials have investigated the effect of levothyroxine (LT₄) in pregnant women with SCH, of which two were conducted among TPOAb⁺ women (8, 10) and the other three trials pooled TPOAb⁺ and TPOAb⁻ subjects (11–13).

Overall, no intervention trial has assessed LT₄ impact, specifically in SCH-TPOAb⁻ pregnant women (9), which precludes making a recommendation for or against routine treatment in such women. The only study that assessed TPOAb⁻ women, conducted by Negro *et al.* (9), had no interventional component.

In the present population-based study, we aimed to determine the potential efficacy of LT₄ therapy on pregnancy outcomes in SCH-TPOAb⁻ women using both TSH cut points of 2.5 and 4.0 mIU/L according to the 2011 and 2017 ATA guidelines, respectively (1, 14).

Methods

Study design and participants

Data were extracted from the Tehran Thyroid and Pregnancy Study, a two-phase study conducted from September 2013 through February 2016. Details of the study protocol have previously been published (15); in brief, phase 1 was a population-based cross-sectional study in which 1746 pregnant women attending prenatal clinics at Shahid Beheshti University of Medical Sciences were screened for thyroid dysfunction. Pregnant women with twin pregnancies (n = 28)

and those with overt thyroid dysfunction or SCH were excluded (n = 99); women with other types of thyroid dysfunction were then invited for the second phase of the study (n = 1619) (Fig. 1), the results of which were previously reported as a randomized clinical trial among TPOAb⁺ cases (n = 134) (10). For the purpose of the current study, we reported on 393 SCH-TPOAb⁻ and 1092 euthyroid TPOAb⁻ women who entered the second phase of the study, 366 and 1028 of whom, respectively, agreed to participate (Fig. 1).

SCH-TPOAb⁻ women were assigned to two groups: group A (n = 183) treated with LT₄ and group B (n = 183) without treatment. Euthyroid TPOAb⁻ women (n = 1028) served as the control group (group C) (Fig. 1). In group A, patients were treated with an LT₄ morning dose of 1 μg/kg/d, initiated 4 to 8 days after the first prenatal visit and maintained throughout pregnancy.

Study procedure

A comprehensive questionnaire including demographics and reproductive, medical, and prenatal history was completed during face-to-face interviews, and a checklist including all potential risk factors, as recommended by the ATA, was filled out (14). Signs and symptoms of thyroid disorders were assessed thoroughly. Gestational age (GA) was calculated according to the first day of the last menstrual cycle for women with regular cycles and/or ultrasonography for those with irregular cycles or those who could not precisely recall their last menstrual cycle (n = 66).

All study participants received standard prenatal care at regular intervals as recommended by prenatal guidelines (15) and were followed up until delivery; adverse pregnancy outcomes were managed according to standard guidelines.

Overnight blood samples were collected at the first prenatal visit, second trimester (20 to 24 weeks of gestation), and third trimester (30 of 34 weeks of gestation) to measure serum levels of TSH, thyroxine (T₄), resin T uptake (RTU), and TPOAb. In group A, the second and third samples were obtained before ingestion of LT₄. After centrifugation, samples were stored at -80°C until the end of the study; TSH levels in offspring were measured 3 to 5 days after delivery. At the first prenatal visit, participants were asked to collect three casual morning urine samples (5 to 10 mL) on an every other day basis; these samples were kept frozen at -20°C until assayed at the end of the study.

Written informed consent was obtained from all participants, and the study was approved by the ethics committee of the Research Institute of Endocrine Sciences, approval no. 6ECRIES93/03/13, and was registered in the Iranian Registry of Clinical Trials (IRCT) Registry (code: IRCT2013121214849N3).

Randomization and masking

TPOAb⁻ women with SCH were randomly assigned to two groups, and a computer-generated list was created to achieve balance across treatment groups using permuted blocks of four. A sealed opaque envelope was allocated to each subject. Physicians who participated in various phases of the study were blinded to the groups to which the patients belonged. The midwife providing prenatal care, who did not participate in any subsequent phases of the study, was the only person who knew which group each patient belonged to (single blindness). Masking to treatment allocation was not possible, and only those health

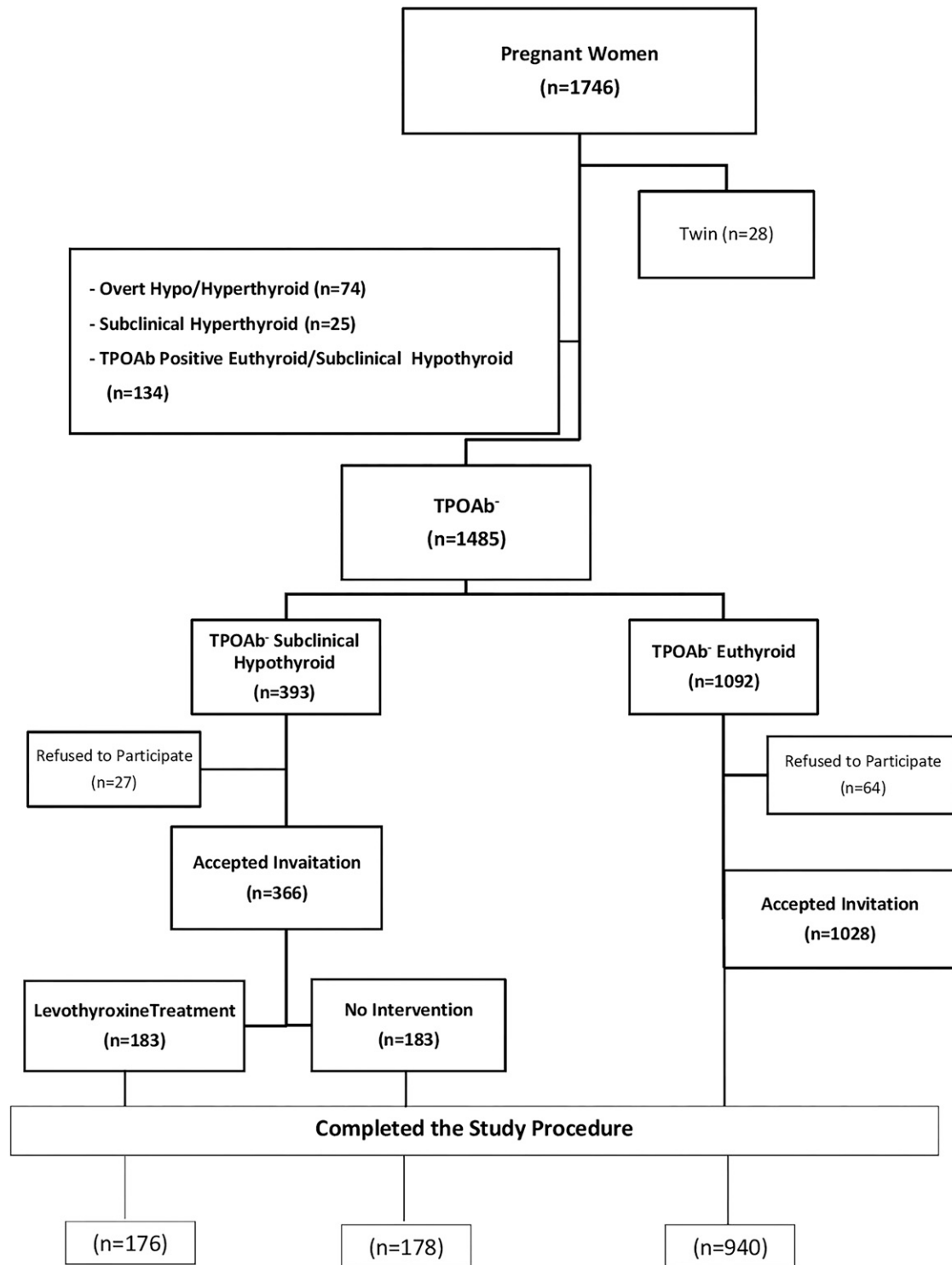


Figure 1. Study flowchart.

care workers who determined pregnancy outcomes were blinded to treatment allocation.

Assays

Using the radioimmunoassay and the immunoradiometric assay, T₄ and TSH levels, respectively, were measured with commercial kits (Izotop, Budapest, Hungary) and the γ -counter

(Dream Gamma-10; Goyang-si, Gyeonggi-do, South Korea); RTU and TPOAb were measured with an enzyme immunoassay (Diaplus, San Francisco, CA) and immunoenzymometric assay (Monobind, Costa Mesa, CA), respectively, using a calibrated enzyme-linked immunosorbent assay reader (Sunrise; Tecan Co., Salzburg, Austria). Interassay and intra-assay coefficients of variation for T₄, resin T3 uptake, TSH, and TPOAb were 1.1% and 3.9%; 2.2% and 4.3%; 1.9% and 4.7%; and 1.0%

and 1.6%, respectively. Because free T_4 immunoassays might be influenced in a method-specific manner by pregnancy-related changes of serum thyroxine-binding globulin and albumin, the free T_4 index (FT4I) was calculated instead using the following formula: $FT4I = T_4 \times RTU/100$ (16).

Urinary iodine concentration was measured using a manual method based on the Sandell-Kolthoff technique (17). In three ranges of 3.4, 12.5, and 37.1 $\mu\text{g/L}$, the intra-assay and inter-assay coefficients of variation were 8.5%, 7.2%, and 9.6% and 9.1%, 8.6%, and 12.3%, respectively.

Outcomes

In this study the primary outcome was preterm delivery, defined as birth before 37 weeks of gestation. Secondary outcomes were placental abruption, stillbirth, and neonatal admission.

Definitions

Women with baseline concentrations of TSH of 0.1 to 2.5 mIU/L, FT4I of 1 to 4.5, and TPOAb <50 IU/mL were considered euthyroid TPOAb⁻ and served as controls. Overt hyperthyroidism was defined as a TSH level <0.1 mIU/L and an FT4I >4.5. Overt hypothyroidism was defined as a TSH level >10 mIU/L or TSH level >2.5 mIU/L and an FT4I <1. SCH was defined as a normal FT4I (1 to 4.5) despite an elevated TSH level (2.5 to 10 mIU/L). SCH was defined as a normal FT4I (1 to 4.5) despite a reduced TSH level (<0.1 mIU/L). TPOAb level >50 IU/mL was considered TPOAb positivity.

Statistical analysis

Sample size was calculated according to an intention-to-treat analysis with superiority assumption (in terms of primary outcome). A sample of 366 subjects (183 per group) was necessary to detect a 0.5 difference rate in preterm delivery, with a two-sided 5% significance level, a power of 85%, a loss to follow-up rate of 10%, and a superiority proportion of 0.05.

Continuous variables were checked for normality using the one-sample Kolmogorov-Smirnov test; categorical variables were expressed as percentages and were compared using the Pearson χ^2 test. Continuous variables with normal distribution were compared between groups using one-way analysis of variance and were expressed as mean (standard deviation). Nonnormally distributed variables were expressed as median (interquartile) and were compared between groups using the Mann-Whitney test.

Analysis of variance was used to determine whether there were any differences between the means of pregnancy outcomes (GA, birth weight, height, and head circumference) in the three study groups.

To identify the impact of intervention based on a TSH cut point of 2.5 mIU/L, the primary outcome was calculated as event numbers and percentages by treatment allocation. Effect measures [relative risks (RRs)] were calculated with 95% confidence intervals (CIs), with expectant management as the reference group. No participants were excluded from the primary intention-to-treat analysis for protocol violations. There was no imputation for missing outcomes. Participants with missing data were excluded from the analysis. Number needed to treat was defined as the numeric cohort of patients who needed to be treated to prevent the occurrence of the primary outcome.

Data were reanalyzed using the TSH cutoff value of 4.0 mIU/L. The effect of treatment on binary pregnancy outcomes was estimated using the log-binomial model, which is known to

be a useful approach for computing an adjusted RR (18). This analysis was performed with the following predictors: TSH (<4 and ≥ 4 mIU/L), group status (receiving LT_4 or no intervention), and an interaction term of these two (TSH \times group status). The same analysis was done for urinary iodine using a cutoff value of 150 $\mu\text{g/L}$. To determine the potential effect of the nonsignificant differences of GA between groups A, B, and C (50.3%, 39%, and 65%, respectively) on pregnancy outcomes, we constructed a binary variable for GA (value 1 for GA ≥ 12 weeks and value 0 for GA <12 weeks). Then we adjusted it as a predictor in the log-binomial model to explore the effect of treatment on pregnancy outcomes.

Statistical analysis was performed using the STATA software package (version 12; STATA Inc., College Station, TX); the significance level was set at $P < 0.05$ and the CI at 95%.

Results

The mean (standard deviation) of age and body mass index of the study subjects in groups A, B, and C were 27.0 (5.3), 26.9 (4.7), and 27.1 (5.2) years and 25.8 (4.9), 26.0 (4.6), and 24.8 (4.6) kg/m^2 , respectively. Mean (standard deviation) of GAs in the three groups were 11.4 (4.1), 12.2 (4.3), and 11.2 (4.1) weeks, respectively (Table 1). Median (interquartile) urine iodine levels in groups A, B, and C were 140 (89 to 219), 123 (86 to 220), and 120 (79 to 184) $\mu\text{g/L}$, respectively.

Medians (interquartiles) for TSH levels in group A in the first, second, and third trimesters were 3.7 (2.8 to 4.8), 2.5 (1.7 to 3.7), and 2.1 (1.3 to 2.7) mIU/L, respectively; in group B, they were 3.6 (2.1 to 4.2), 3.7 (2.7 to 4.5), and 3.2 (2.1 to 5.1) mIU/L, respectively, and in group C, they were 1.5 (0.8 to 1.9), 1.8 (1.2 to 2.4), and 1.7 (1.0 to 2.3) mIU/L, respectively. The values for FT4I in the three trimesters for groups A, B, and C were 2.7 (2.3 to 3.2), 3.6 (2.9 to 3.9), and 3.0 (2.4 to 3.3); 2.9 (2.4 to 3.2), 3.1 (2.6 to 4.0), and 2.7 (2.4 to 3.1); and 3.0 (2.4 to 3.4), 3.1 (2.6 to 3.5), and 3.0 (2.6 to 3.2), respectively. Medians (interquartiles) for T_4 level in group A in the first, second, and third trimesters were 10.1 (8.5 to 12.4), 12.4 (11.1 to 14), and 11.5 (10.2 to 12.4) $\mu\text{g/dL}$, respectively; in group B, they were 10.7 (8.7 to 12.2), 11.2 (9.1 to 13.6), and 11 (9.8 to 12.2), respectively, and in group C, they were 10.4 (8.7 to 12.5), 12.1 (10 to 14), and 10.6 (9.2 to 12), respectively.

Figure 2 shows (a) TSH, (b) FTI, and (c) T_4 values during gestation among study groups. Although baseline median TSH value in group A was not significantly different from that in group B [3.8 (2.8 to 4.8) vs 3.6 (3.1 to 4.2) mIU/L, respectively; $P = 0.78$] after treatment, it was significantly lower in group A in the second and third trimesters [2.4 (1.7 to 3.7) vs 3.7 (2.7 to 4.5) and 2.1 (1.3 to 2.7) vs 3.2 (2.1 to 5.1) mIU/L, respectively; $P < 0.001$] (Fig. 2a). Median baseline T_4 value in group A was not significantly different from that in group B [10.1 (8.5 to

Table 1. Characteristics of Study Participants

Characteristic	Group A (n = 183)	Group B (n = 183)	Group C (n = 1028)
Maternal age, mean (SD), y	27.0 (5.34)	26.9 (4.74)	27.1 (5.17)
Maternal BMI, mean (SD), kg/m ²	25.8 (4.95)	26.0 (4.64)	24.8 (4.61)
Parity, mean (SD)	0.64 (0.73)	0.7 (0.84)	0.92 (0.97)
GA at first visit, mean (SD), wk	11.4 (4.10)	12.2 (4.36)	11.2 (4.11)
GA at first visit, n (%)			
≤8 wk	38 (21.7)	43 (23.6)	328 (30.0)
8–10 wk	32 (18.3)	12 (6.6)	228 (20.9)
10–12 wk	18 (10.3)	16 (8.8)	164 (15.0)
12–14 wk	36 (20.6)	32 (17.6)	111 (10.2)
14–20 wk	51 (29.1)	79 (43.4)	261 (23.9)
History of infertility, n (%)	9 (4.9)	8 (4.4)	48 (4.4)

Group A: treated with LT₄; group B: no intervention; group C: controls.

Abbreviations: BMI, body mass index; SD, standard deviation.

12.4) vs 10.7 (8.7 to 12.2) μg/dL, respectively; $P = 0.30$]. After treatment, it was significantly higher in group A in the second trimester than in group B [12.4 (11.1 to 14) vs 11.2 (9.1 to 13.6) μg/dL, respectively; $P = 0.006$]. Comparison of the median T₄ level in group A with that of group B showed no statistically significant difference in the third trimester [11.5 (10.2 to 12.4) vs 11 (9.8 to 12.2) μg/dL, respectively; $P = 0.08$] (Fig. 2c).

Neonatal birth weight, head circumference, and TSH levels were not improved by maternal LT₄ therapy in group A compared with group B. Also, we found no significant correlation between maternal and neonatal TSH values among our study groups (Table 2).

On the basis of a TSH cutoff value of 2.5 mIU/L, LT₄ replacement therapy had no statistically significant effect on reduction of the preterm delivery rate in group A compared with the rate in group B (RR: 0.86; 95% CI: 0.47 to 1.55; $P = 0.61$), although there was a significant difference in group A compared with group C (RR: 1.91; 95% CI: 1.14 to 3.18; $P = 0.01$). There was also a significant difference in the rate of preterm delivery between groups B and C (RR: 2.22; 95% CI: 1.37 to 3.60; $P = 0.001$) (Fig. 3a).

To further explore the impact of treatment based on a TSH cutoff value of 4.0 mIU/L and a urinary iodine cut point of 150 μg/L, a log-binomial model was fitted and adjusted for GA at the time of recruitment. Data analysis indicated no significant difference in progression of outcomes, including preterm delivery and neonatal admission, between groups A and B with TSH levels <4.0 mIU/L (Table 3); however, women in group A with baseline TSH values ≥4.0 mIU/L were less likely to experience preterm delivery than were women in group B (RR: 0.38; 95% CI: 0.15 to 0.98; $P = 0.04$); in other words, the risk of preterm delivery in group A decreased about 62% compared with the risk in group B with TSH values ≥4.0 mIU/L. Also, preterm delivery was less common among women in group B whose TSH values were <4.0 mIU/L compared with

those in group B with TSH values ≥4.0 mIU/L (RR: 0.44; 95% CI: 0.2 to 0.97; $P = 0.04$). Although the risk of preterm delivery in group A decreased about 43% in women with TSH values ≥4.0 mIU/L compared with those with TSH values <4.0 mIU/L, the difference was not statistically significant (RR: 0.57; 95% CI: 0.22 to 1.46; $P = 0.24$) (Fig. 3b; Table 3). There was no statistically significant difference in preterm delivery between women in group B whose urinary iodine level was <150 μg/L and those whose urinary iodine level was ≥150 μg/L (RR: 0.55; 95% CI: 0.18 to 1.62; $P = 0.3$) (Table 3).

Considering TSH values ≥4.0 mIU/L, the number needed to treat for preterm delivery was 3.8 (95% CI: 2.2 to 67.5).

Discussion

Although a majority of researchers studying the association between SCH and pregnancy outcomes have pooled TPOAb⁺ and TPOAb⁻ cases, we examined the impact of LT₄ on obstetric outcomes specifically among TPOAb⁻ women.

In this population-based study, we showed that based on a TSH cut point of 2.5 mIU/L, LT₄ therapy did not lower the rate of adverse pregnancy events in SCH-TPOAb⁻ women. However, increasing the cut point to 4.0 mIU/L, as currently recommended by the ATA, could decrease the rate of preterm delivery (RR: 0.38; 95% CI: 0.15 to 0.98; $P = 0.04$).

Multiple studies have investigated the association between TSH elevation and preterm delivery. Consistent with our results, a prospective study of Texan women ($n = 17,298$) concluded that women with a TSH level at or above the 97.5th percentile before 20 weeks of gestation were two times more likely to experience preterm birth before 34 weeks of gestation (5); however, their findings were not stratified according to TPOAb status. In contrast, Negro *et al.* (9) failed to show any difference in the

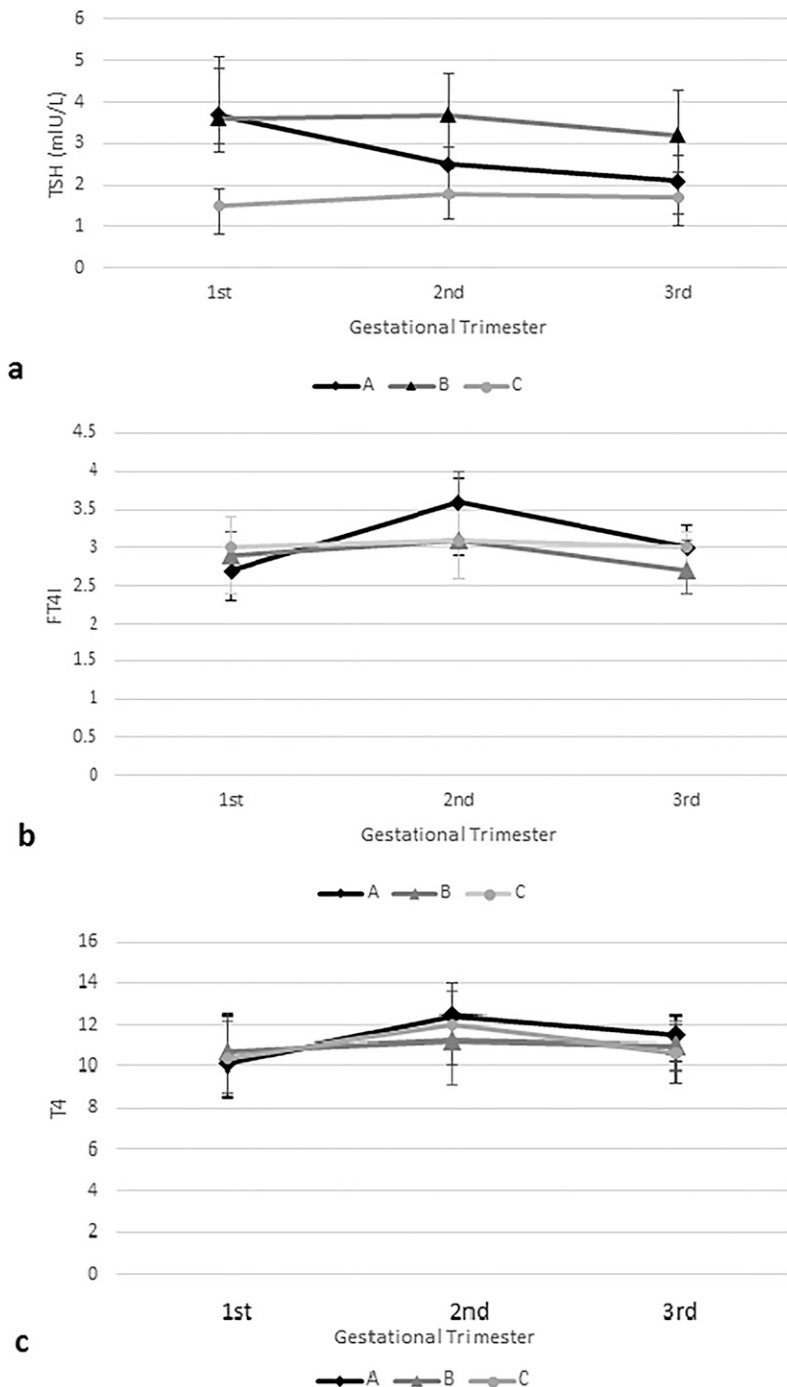


Figure 2. (a) TSH, (b) FT4I, and (c) T₄ values during gestation in study groups. Group A: treated with LT₄; group B: no intervention; group C: controls.

rate of preterm delivery in TPOAb⁻ women with TSH levels of 2.5 to 5.0 mIU/L compared with those with normal TSH levels (<2.5 mIU/L); however, no intervention was provided for the patients in their study. The difference between their findings and those of the current study might be due to their inclusion of women with TSH ranges between 2.5 and 5.0 mIU/L, whereas we defined SCH as TSH levels of 2.5 to 10 or 4.0 to 10 mIU/L. Likewise, Cleary-Goldman *et al.* (19) found no association between TSH

elevation and pregnancy complications (*e.g.*, prematurity and low birth weight); however, their analysis was performed on a small proportion of women (~30% of the initial study cohort), which limited data interpretation. These conflicting results on the relationship between SCH and preterm delivery (19–21) may be explained in part by variable cut points used across studies to define SCH and also by the pooling of TPOAb⁺ and TPOAb⁻ cases in the data analysis.

Of the limited randomized trials that have so far assessed the use of LT₄ in women with SCH, two that evaluated pregnancy outcomes were conducted among TPOAb⁺ women (8, 10), and the other three studies were not stratified according to anti-TPOAb status (11–13). In the first trial, Negro *et al.* (8) randomly assigned 4562 high-risk women to universal thyroid screening and a case-finding approach and found that the use of LT₄ to treat SCH-TPOAb⁺ women (TSH level >2.5 mIU/L) was associated with a reduced rate of pregnancy complications. Similarly, our earlier report from the Tehran Thyroid Study on TPOAb⁺ women supported a 70% reduction in preterm delivery rate in LT₄-treated patients (10).

In another trial by Ma *et al.* (11), lower odds of miscarriage and macrosomia were reported in women who received intervention (odds ratio: 0.34; CI: 0.21 to 0.56 and odds ratio: 0.46; CI: 0.28 to 0.74, respectively). However, a limitation of their study was the asynchronous assignment of women into screening and control groups (at gestational weeks 11 and 7, respectively), which may explain why their control group experienced more early pregnancy

losses. In the Controlled Antenatal Thyroid Screening study, women with SCH were treated regardless of their antibody status, and the cognitive function of children at age 3 years did not differ between children born to treated mothers and those born to their untreated counterparts (13). However, a 24% follow-up loss of the initial cohort was a limitation for their intention-to-treat analysis.

Finally, in a placebo-controlled study by Casey *et al.* (12) investigating 677 LT₄-treated mothers, findings were

Table 2. Pregnancy Outcomes in Study Groups

Adverse Pregnancy Outcome	Group A (n = 183)	Group B (n = 183)	Group C (n = 1028)
Preterm delivery, ^a n (%)	18 (9.8)	21 (11.5)	53 (5.6) ^b
Neonatal admission, ^a n (%)	8 (4.5)	9 (4.9)	75 (8.0)
Placental abruption, ^a n (%)	3 (1.6)	0	5 (0.5)
Stillbirth, ^a n (%)	0	0	2 (0.2)
GA ^c mean (SD)	38.03 (1.4)	37.9 (1.5)	39.4 (1.4)
Birth weight, ^c mean (SD)	3190.82 (455.13)	3203.1 (497.1)	3236.6 (448.8)
Neonate height, ^c mean (SD)	50.1 (2.3)	50.2 (2.7)	50.1 (2.3)
Birth head circumference, ^c mean (SD)	34.6 (1.4)	34.7 (1.6)	34.7 (2.3)
Neonatal TSH, mIU/L	1.1 (0.5–1.9)	1.1 (0.5–2.1)	0.90 (0.4–1.7)

Group A: treated with LT₄; group B: no intervention; group C: controls.

Abbreviation: SD, standard deviation.

^aχ² test was used to compare groups.

^bGroup C vs group B: *P* = 0.02.

^cAnalysis of variance test was used to compare groups.

similar to those of the Controlled Antenatal Thyroid Screening study. Although this study had the strength of a 5-year follow-up with a >90% follow-up rate, results were confined by the late initiation of replacement therapy at a mean of 17 weeks. Also, the primary endpoints of the two latter trials were IQ and cognitive performance of children born to LT₄-treated mothers with SCH. Moreover, lack of TPOAb data was an important limitation of these two studies, which precluded a comparison of their data with our findings.

On the whole, despite the mentioned discrepancies, data generally confirm the beneficial effect of treatment in women with SCH in terms of reducing preterm birth rates (1). However, different factors, including antibody positivity, timing of treatment initiation, and cut point values used, must be considered when interpreting the results.

Our findings conform closely to the latest ATA guideline that recommends intervention only in SCH-TPOAb⁺ women and not in those who are TPOAb[−] and have TSH levels <4.0 mIU/L (1). The underlying cause for this recommendation may be reports that present higher rates of adverse outcomes attributable to TPOAb positivity (22).

Moreover, since the target became to keep TSH concentrations below 2.5 mIU/L, the accuracy of this threshold has been questioned in recent years. Because response to treatment varies significantly within the wide TSH range of 2.5 to 10 mIU/L (4), LT₄ therapy in those with serum concentrations of 2.5 to 4.0 mIU/L may result in overtreatment of euthyroid women (3, 4).

In the current study, when the TSH cutoff value of 2.5 mIU/L was considered, administration of LT₄ showed no effect on pregnancy outcomes, whereas treatment reduced the rate of preterm delivery in women with TSH levels ≥4.0 mIU/L. Reports showed varying TSH upper limits ranging between 2.1 and 4.6 mIU/L in different

cohorts, of which 90% were higher than the common cutoff value of 2.5 mIU/L (3). Apparently, use of the fixed cut point of 2.5 mIU/L according to the 2014 European Thyroid Association and 2011 ATA guidelines (14, 23) appears to no longer be valid among populations such as Iranians (16, 24).

This evidence is further confirmed by a recent study by Maraka *et al.* (4) assessing the effectiveness of LT₄ among women with SCH (n = 5405), which demonstrated the treatment benefit only in women with baseline TSH levels of 4.1 to 10.0 mIU/L but not in those with pretreatment TSH concentrations of 2.5 to 4.0 mIU/L. Also, data from the Generation R study suggested that although TSH concentrations >2.5 mIU/L did not affect rate of preterm delivery, the risk was 2.5-fold higher among women with TSH levels ≥4.0 mIU/L (25); the authors noted that when TPOAb⁺ women were excluded from the analysis, this association did not remain.

Our prior study assessing treatment effects on TPOAb⁺ women revealed that LT₄ therapy can result in an 80% reduction in neonatal admission rate, mainly in women with a TSH level ≥4.0 mIU/L (10). In the current study, however, treatment had no effect on the rate of neonatal admission, a result that may be partly due to the higher GA of newborns at the time of delivery or the exclusion of TPOAb⁺ women who were more likely to suffer perinatal complications irrespective of thyroid hypofunction mechanisms (5, 6, 26, 27). Thyroid antibodies can alter the secretion of interleukin-4, interleukin-10, and interferon γ, which in turn can change the immune response of the uterus in up to 40% of women with thyroid autoimmunity (28). Elevated thyroid antibodies are associated with increased TSH levels (29) and decreased thyroid ability to function adequately during pregnancy, leading to overt hypothyroidism (30). These results are comparable to those of

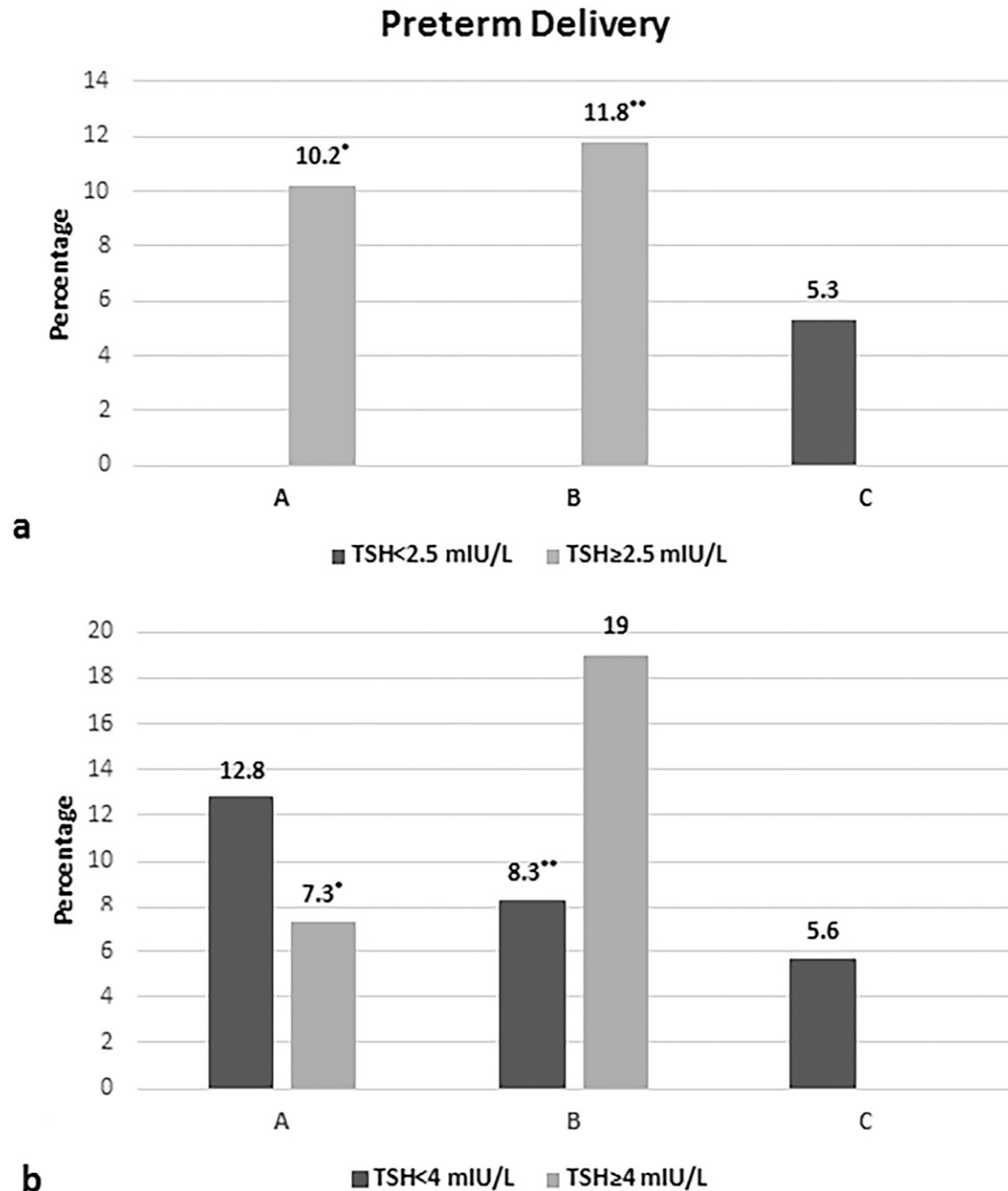


Figure 3. Comparison of preterm delivery rates among study groups according to TSH cut points of (a) 2.5 mIU/L and (b) 4.0 mIU/L. Group A: treated with LT_4 ; group B: no intervention; and group C: controls. *Group A with $TSH \geq 4$ mIU/L in comparison to group B with $TSH \geq 4$ mIU/L; $P = 0.04$. **Group B with $TSH < 4$ mIU/L in comparison to group B with $TSH \geq 4$ mIU/L; $P = 0.04$.

Liu *et al.* (22), who reported that women with TSH values within the range of ≥ 2.5 to < 5.22 mIU/L did not have an increased risk of pregnancy loss, whereas the concomitant thyroid antibodies exacerbated the effect and increased the risk of adverse events.

The strength of this study lies in its methodology as a population-based study conducted mainly on pregnant women in their first trimester. Comprehensive thyroid assessments, including a detailed history, physical examination, and thyroid function tests (TSH, T_4 , T-uptake,

TPOAb, and urinary iodine concentration) were performed for all study participants. However, the following limitations should be considered when extrapolating the results of this study: First, because of the nontimely referral of the women, meaningful results were not obtained on miscarriage rates; hence, these data are not shown here. Second, we were unable to measure antithyroglobulin antibodies; along with TPOAb, these measurements could have more effectively differentiated women with thyroid autoimmunity. However, using only TPOAb would miss a

Table 3. Log-Binomial Model Analysis for Pregnancy Outcomes in Study Groups Based on TSH Cutoff Value of 4.0 mIU/L and Urinary Iodine Concentration of 150 µg/L, Adjusted for GA at Time of Recruitment

Preterm Delivery	RR (95% CI)	P Value
TSH subgroups		
TSH ≥4 mIU/L and without LT ₄ treatment	Reference Group	
TSH ≥4 mIU/L and with LT ₄ treatment	0.39 (0.15–0.98)	0.04
TSH <4 mIU/L and without LT ₄ treatment	0.44 (0.23–0.97)	0.04
TSH <4 mIU/L and with LT ₄ treatment	0.68 (0.32–1.44)	0.32
Urinary iodine subgroups		
Urine iodine <150 µg/L and without LT ₄ treatment	Reference Group	
Urine iodine <150 µg/L and with LT ₄ treatment	0.66 (0.28–1.54)	0.34
Urine iodine ≥150 µg/L and without LT ₄ treatment	0.55 (0.19–1.60)	0.30
Urine iodine ≥150 µg/L and with LT ₄ treatment	0.73 (0.33–1.76)	0.50
Neonatal Admission		
TSH subgroups		
TSH ≥4 mIU/L and without LT ₄ treatment	Reference Group	
TSH ≥4 mIU/L and with LT ₄ treatment	1.05 (0.18–6.18)	0.95
TSH <4 mIU/L and without LT ₄ treatment	1.67 (0.31–7.85)	0.60
TSH <4 mIU/L and with LT ₄ treatment	1.56 (0.17–2.35)	0.58
Urinary iodine subgroups		
Urine iodine <150 µg/L and without LT ₄ treatment	Reference Group	
Urine iodine <150 µg/L and with LT ₄ treatment	0.49 (0.1–2.66)	0.41
Urine iodine ≥150 µg/L and without LT ₄ treatment	0.83 (0.15–4.33)	0.82
Urine iodine ≥150 µg/L and with LT ₄ treatment	0.83 (0.2–3.61)	0.81

small number (5%) of pregnant women with isolated antithyroglobulin antibodies (31). Third, we did not use our local trimester-specific cutoff values for TSH and FT4I (16), as these values were introduced after initiation of the current study. Fourth, the number of samples was insufficient to examine other rare pregnancy complications (e.g., preeclampsia, stillbirth). Finally, it should be noted that lack of knowledge regarding other risk factors of premature delivery could influence the results of this study, although the randomized allocation of study participants minimized this effect.

In conclusion, this study provides evidence that replacement therapy with LT₄ in pregnant women with TSH concentrations of 2.5 to 4.0 mIU/L who are negative for TPOAb could not improve pregnancy outcomes, whereas treatment in women with TSH concentrations ≥4.0 mIU/L was beneficial in reducing preterm delivery. Confirmatory randomized clinical trials are critically needed to investigate the benefit of LT₄ treatment initiated preconception and in early pregnancy.

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