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What Should Be the Thyroid Stimulating Hormone Cut-off Level in the First-Trimester Screening? : A Prospective Cohort Study and Mini Review of The Guidelines

Birinci Trimester Taramasında Tiroid Uyarıcı Hormon Cut-off Düzeyi Ne Olmalıdır?: Prospektif Bir Kohort Çalışması ve Kılavuzların Mini İncelemesi

Halime Sen Selim[1], Sefa Kelekci[2]

[1] Izmir Katip Celebi University, Ataturk Training and Research Hospital, Department of Gynecology and Obstetrics, Izmir, Turkey
[2] Izmir Katip Celebi University, Faculty of Medicine, Department of Gynecology and Obstetrics, Izmir, Turkey

Abstract

Aim: We aimed to evaluate the contributions of the maternal serum TSH cut-off value of 2.5 mU/L to the development of maternal and fetal complications.

Materials and Methods: We constructed the study with pregnancies in their first trimester and planned to do only observation prospectively. We excluded pregnant women with systemic disease and any history of thyroid surgery or thyroid pathology. According to the TSH level, a case group(TSH level >2.5 mU/L) and a control group (TSH level<2.5 mU/L) were created through the pregnants with normal Thyroxine (T4) levels. The cohort group were divided into four subgroups according to whether they were anti-thyroid peroxidase (anti-TPO) positive or not. We observed the fetomaternal outcomes like pregnancy loss, hyperemesis gravidarum, hypertensive disorders, gestational diabetes, prelabour rupture of membranes, placental abruption, with routine prenatal visits until delivery; also delivery style, birth weight, shoulder dystocia, newborn intensive care needs, and postpartum hemorrhage were recorded.

Results: The incidence of miscarriage in the subgroup with TSH >2.5 mU/L and anti-TPO (+) was significantly higher than in those with TSH <2.5 mU/L and anti-TPO (+) ($p<0.05$). All groups had no significant difference in other maternal or fetal/neonatal complications.

Conclusion: If only the population-based nomograms are created, we may advise maternal serum TSH level as <2.5 mU/L for first-trimester screening.

Single or multiple pregnancy status, gestational age, and the presence of thyroid peroxidase antibodies should also be taken into account when creating these nomograms.

Keywords: Anti-thyroid antibodies; hypothyroidism; postpartum hemorrhage; pregnancy loss; TSH reference range

Öz

Amaç: Bu çalışmada maternal serum TSH düzeylerinin ve anti-tiroid antikor durumunun maternal/fetal olumsuz sonuçlar üzerindeki etkisini değerlendirmeyi ve ve birinci trimester cut-off düzeyi olarak 2,5 mU/L'nin gerekliliğini değerlendirmeyi amaçladık**.**

Gereç ve Yöntemler: Birinci trimesterdeki gebelerde prospektif gözlemsel kohort çalışması yapıldı. Bilinen diyabet, hipertansiyon, tiroid cerrahisi veya herhangi bir tiroid hastalığı öyküsü olan hastalar dışlandı. Tiroksin (T4) düzeyi normal olan kadınlar başlangıç TSH düzeylerine göre vaka grubu (TSH düzeyi >2,5 mU/L) ve kontrol grubu (TSH düzeyi <2,5 mU/L) olmak üzere iki gruba ayrıldı. Serum anti-tiroid antikor durumu değerlendirildi ve anti-tiroid peroksidaz (anti-TPO) pozitif olup olmamasına göre dört alt grup oluşturuldu. Gebelik kaybı, hiperemezis gravidarum, hipertansif bozukluklar, gestasyonel diyabet, doğum öncesi membran rüptürü, plasenta dekolmanı gibi fetomaternal sonuçları doğuma kadar rutin prenatal ziyaretlerle gözlemlendi; ayrıca doğum şekli, doğum ağırlığı, omuz distosisi, yenidoğan yoğun bakım ihtiyacı ve doğum sonu kanama olup olmadığı kaydedildi.

Bulgular: TSH >2,5 mU/L ve anti-TPO (+) olan alt grupta spontan abortus insidansı, TSH<2,5 mU/L ve anti-TPO (+) olanlara göre anlamlı olarak yüksekti ($p<0,05$). Tüm gruplarda diğer maternal veya fetal/neonatal komplikasyonlarda anlamlı fark yoktu.

Sonuç: Serum TSH düzeyini <2,5 mU/L olarak önermek için fetüs sayısı, gebelik haftası ve anti-tiroid antikor durumuna göre oluşturulmuş popülasyon tabanlı nomogramlar geliştirilmesi gerekmektedir.

Anahtar Sözcükler: Anti-tiroid antikorlar; hipotiroidizm; doğum sonu kanama; gebelik kaybı; TSH referans aralığı

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Corresponding author: Halime Şen Selim, MD
Gynecology and Obstetrics Department of
Ataturk Training and Research Hospital, Basin Sitesi,
Yesilyurt, 35360 Izmir, Turkey
e-mail: dr.halime.sen.selim@gmail.com

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Introduction

As it is known, hypothyroidism is the insufficiency of thyroid hormones; increased serum TSH levels make the diagnosis. If the increase in TSH levels is combined with low serum thyroxine hormone, overt hypothyroidism is diagnosed; If serum T4 level can be normalized with TSH increase, subclinical hypothyroidism is diagnosed (1). However, some changes in thyroid physiology in pregnancy complicate the diagnosis.

Fetal and maternal thyroid glands increase thyroid hormone secretion by 40-100% to accomplish the increased requisition during pregnancy. Thyroid Stimulating Hormone (TSH) and Human chorionic gonadotropin (hCG) are glycoproteins with molecular structures composed of alpha and beta subunits. Only the beta ones of the subunits are dissimilar despite, the alpha ones being analog. Due to this structural analogy, hCG acts as a thyrotropic hormone (2,3). Thus, the maternal thyroid-stimulating grade is decreased by 80% of the pregnancies.

The presence of anti-TPO autoantibodies is related to miscarriages, perinatal mortality, and low for gestational age (LGA) infants, even though the status of thyroid hormone is in the normal range(4,5). Hypothyroidism is related to serious complications, like miscarriage, hypertensive disorders, placental abruption, postpartum hemorrhage(PPH), premature rupture of membrane (PROM), preterm delivery, increased neonatal intensive care unit (NICU) needs low birth weight, macrosomia, shoulder dystocia, increased cesarean birth rate, perinatal morbidity, and mortality (6-9).

Novel investigation and several commission opinions recommend that the TSH cut-off value should be 2.5 mU/L in the first-trimester thyroid function screening (10-15). We aimed to evaluate the contributions of the maternal serum TSH cut-off value of 2.5 mU/L to the development of maternal and fetal complications.

Materials and Methods

We constructed the study with pregnancies in their first 12 weeks who applied to our hospital from October 2012 to February 2013 and had an average TSH level according to our hospital reference range (0.35-5.50 mU/L); Furthermore, we planned to do only observation prospectively and not any treatment according to the current guidelines at the time. We didn't include in the study the pregnant diagnosed with overt hypothyroidism or hyperthyroidism. We excluded pregnant women with systemic disease and any history of thyroid surgery or thyroid pathology. According to the TSH level, a case group(TSH level: 2.5-5,5 mU/L) and a control group (TSH level:0.35-2.5 mU/L) were created through the pregnants with normal Thyroxine (T4) levels. Anti-thyroid peroxidase antibodies (anti-TPO) were also measured from the same blood samples of these pregnant women. Subgroups were formed according to whether they were anti-TPO positive or not. First-trimester hemoglobin (Hg) and fasting plasma glucose (FPG) values were also recorded. Afterward, women were routinely followed up with at least five visits during pregnancy.

This observational cohort study was carried out after obtaining approval from the ethics committee of the Obstetrics and Gynecology Department of Izmir Katip Çelebi University Atatürk Training and Research Hospital (number: 2012/70) following the principles outlined in the Declaration of Helsinki.

Outcome measures

The women were monitored for gestational hypertension until birth by performing blood pressure measurements at each visit and in their family health centers. We defined gestational hypertension (GHT) and preeclampsia based on two measurements performed at least 6 hours apart and the absence or presence of proteinuria if it is detected for the first time after the 20th week of pregnancy.

In the 24th week, a 75-g oral-glucose loading test was performed on the women, and they were evaluated for gestational diabetes by measuring blood glucose at 0, 60, and 120min.

3rd-trimester hemoglobin values and 6 hours after birth postpartum hemoglobin values were recorded. Also, women were asked about their amount of postpartum hemorrhage by recoding subjective pad account values. Those with a daily bleeding amount of more than 4-5 pads were recorded as having postpartum bleeding. The development of hyperemesis gravidarum was determined by checking the hospital records and verbal discussions with the patients.

Birth timing wasn't interfered with except for pregnant women with previous cesarean sections. As well as, when pregnant women without the last cesarean section complained of pain, amniotic fluid flow, overdue pregnancy, decreased baby movements, or bleeding, those who started labor spontaneously gave vaginal birth. Birth induction was performed in those who didn't begin spontaneous labor but had a rupture of the membrane or post-term pregnancy.

Pregnancies with indications of fetal distress, cephalopelvic incompatibility or malpresentation, preeclampsia, maternal desire/additional maternal disease, previous uterus operation, placenta previa, or vasa previa were terminated by cesarean section. Shoulder dystocia was confirmed and recorded by asking the doctors, checking patient records, and contacting each patient by phone after the birth. "Low birth weight" was described as the weight of the baby being under 2500 g and as "macrosomia" if more than 4000 g. We also recorded whether there was a need for neonatal intensive care in the first week.

Laboratory measurements

TSH measurements were made with the ADVIA Centaur TSH test. The ADVIA Centaur TPO test was used for anti-TPO measurements, respectively. The test's reference range is 0.35-5.50 mU/L for TSH, a cutoff of 60 U/ml was used for anti-TPO, and there is no specific reference ranges for pregnant and non-pregnant women.

Data synthesis and analysis

IBM SPSS Statistics version 20 was used for statistical analysis. While evaluating the study data, descriptive statistics (mean, standard deviation, and frequency) were used. A student's t-test was used for the parameters that showed a normal distribution between the two groups. A chi-squared test and risk ratios were used to compare qualitative data. In addition, the Mantel-Haenszel test was used to test the homogeneity of ORs between groups. Binary logistic regression was applied to estimate pregnancy loss with some risk factors. Results were evaluated with 95% confidence intervals, and significance was set at p <0.05. confidence intervals and significance were set at p <0.05.

Results

Basic features included in the study

The average age of pregnant women was 27 years. The mean gestational age at which serum TFT samples were taken was 8.26 weeks, and approximately 37.91% were nulliparous. The average TSH value was 2.24 ± 1.19 uIU/ml (0.35-5.34 uIU/ml). Anti-TPO was positive (+) in 25 (24%) patients in the case group and 5 (4.6%) in the control group.

The maternal and fetal outcomes are summarized in table 1 and 2

Outcomes of the first trimester

Pregnancy loss:

Except for one 19-week pregnancy loss, all other pregnancy losses occurred in the first trimester.

When we evaluated 23 cases of miscarriage according to the anti-TPO subgroups, the incidence of pregnancy loss in the subgroup with TSH>2.5 mU/L and anti-TPO(+)was significantly higher than in the subgroup with TSH <2.5 mU/L and anti-TPO(+)(24% vs. 0%) (p <0.05)

Hyperemesis gravidarum

The incidences of hyperemesis are similar in the case and control groups (33.3% vs. 35%) [OR: 1.07 95% CI (0.6-1.8)]. Also, the subgroups are very close to each other, and there is no statistically significant difference.

Maternal outcomes

Hypertensive disorders in pregnancy

While the percentage of GHT was 2% in the control group, it was 1.1% in the case group, and preeclampsia was found to be 3% and 3.4%, respectively. There was no statistically significant difference between the groups or subgroups.

Gestational diabetes

There wasn't statistically significant in the case and control group (6.8% vs. 7%) [OR: 0.97% 95 CI (0.3-3.0)]. Also, antibody positivity didn't seem to increase the risk.

Prelabor rupture of membranes (PROM)

While PROM was seen in 27 cases, all instances were term except for one preterm PROM. Having TSH>2.5 mU/L in the first trimester doesn't seem to pose any risk for PROM.

When the PROM was evaluated based on subgroups, the incidence was 15.8% (n = 15) in the subgroup with TSH <2.5 mU/L anti-TPO (-), whereas there was no PROM in the anti-TPO (+) subgroup. While the incidence was 11.8% (n = 8) in the TSH> 2.5 mU/L anti-TPO (-) subgroup, it was 20% (n = 4) in the anti-TPO (+) subgroup. Although there was no statistical difference between groups and subgroups, antibody positivity appears to increase the incidence of PROM in the group with TSH>2.5.

Placental abruption

No one had a placental abruption.

Delivery style

There wasn't any increased risk in the CS ratio when TSH > 2.5 mU/L or anti-TPO positivity.

Postpartum hemorrhage(PPH)

PPH was evaluated in 188 births. Although there is an increased risk in the case group (27.3% vs. 19%), i was not statistically significant [OR: 1.59 95% CI (0.80-3.17)]. Anti-TPO positivity increased the risk in the case group (42.1% vs. 22.1%), although this difference was not statistically significant.

Fetal/Neonatal outcomes

Birth weight

We didn't observe that either an increase in TSH or anti-TPO positivity harmed newborn birth weight.

Shoulder dystocia

Shoulder dystocia was detected in 3/76 (3.9%) cases during birth. There wasn't a statistically significant difference between the case and control groups (0% vs. 7.3%) or between subgroups.

Newborn intensive care needs:

There wasn't a statistically significant difference between case-control groups (10.2% vs. 13%) or subgroups.

Preterm birth rate

There isn't any statistically significant between the case and control groups (6.8% vs. 8%). When increased TSH > 2.5 mU/L and anti-TPO positivity was found together, there was a partial increase in risk in terms of preterm birth (8%vs 5.2%), but there was no statistically significant difference.

Stillbirth

There wasn't any stillbirth

Discussion

In our study, the anti-TPO positivity rate was found to be 4.62% in the group with TSH <2.5 mU/L, which was accepted as euthyroid. This is similar to the results of Negro et al. (16). The ATA reported that 10-20% of all ongoing first-trimester pregnancies are thyroid-antibody positive and euthyroid (17). Lazarus et al. reported that positivity for TPO antibodies was present in 10% of 14 weeks of pregnancies (18). In our study, the positivity rate of TPO antibodies was found to be 14.2%. The reason for our antibody-positivity rate being moderately high may be the fact that half of the pregnant women consisted of cases with TSH>2.5 mU/L.

***Miscarriage*:** Hypothyroidism has been related to raised abortion rate (19). In our study, 23 (10.9%) of 211 pregnancies resulted in abortion. Although there was an increased risk in the group with TSH>2.5 mU/L(14.6% vs. 7.4%), it wasn't statistically significant [OR: 2.13% 95 CI (0.86, 5.2)]. Similarly, in a study by Baker et al., although

the risk of abortion tended to rise with the increase in TSH level, it was not statistically significant (20).
Negro et al. compared pregnant women with TSH 2.5-5.0 mU/L and anti-TPO(-) and with TSH <2.5 mU/L mU/L; the incidence of miscarriage was found to be higher in the first group (6.1% vs. 3.6%).(21). Similarly, increased rates

Table 1: Comparison of the perinatal outcomes between Control and Study groups.
(Each column proportions do not differ significantly from each other at the .05 level)

	Control group TSH<2.5 mU/L (n = 108)	**Study group TSH>2.5 mU/L (n = 102)**	**Odds ratio**
Pregnancy loss	7.4% (n=8)	14.6%(n=15)	OR: 2.13% 95 CI (0.86, 5.2)
Hyperemesis gravidarum	33.3%(n=36)	35% (n=36)	OR: 1.07 95% CI (0.6-1.8)
Hypertensive disorders in pregnancy • **GHT** • **preeclampsia**	 2%(n=2) 3,0%(n=3)	 1.1%(n=1) 3,4%(n=3)	
Gestational diabetes	7% (n=7)	6,8% (n=6)	OR: 0.97% 95 CI (0.3-3.0)
Prelabor rupture of membranes (PROM):	15 (15%)	12 (13.5%)	OR: 0.88, 95% CI (0.38-2.00).
Placental abruption	0% (n = 0)	0% (n = 0)	
preterm birth	8%(n = 8)	6.8%(n = 6)	OR: 0.77, 95% CI (0.25-2.31)
Delivery style • **cesarean section(CS)** • **vaginal delivery(VD)** • **Pregnancy loss**	 56,5% (n=61) 36.1% (n=39)	 49.5% (n=51) 35.9% (n=37)	
Delivery style (reassessed (CS) indications) • **cesarean section(CS)** • **vaginal delivery(VD)**	 43,5% (n=30) 56,5% (n=39)	 31,5% (n=17) 68,5% (n=37)	OR: 0.77, 95% CI (0.25-2.31)
Birth weight • **low birth weight (LBW)** • **normal birth weight** • **macrosomic**	 4% (n=4), 88% (n=88) 8% (n = 8)	 2.3% (n=2) 96.6% (n=85) 1.1% (n=1)	
Shoulder dystocia	7,3% (n = 3)	0% (n = 0)	OR: 0.4, 95% CI (0.31-0.51)
Newborn intensive care needs	13% (n=13)	10.2% (n=9)	OR: 0.4, 95% CI (0.31-0.51)
Postpartum hemorrhage(PPH):	19% (n=19)	27.3% (n=24)	OR: 1.59, 95% CI (0.80-3.17)

were observed in our study, (11.5% vs. 7.8%). The miscarriage rate is higher in pregnancies accompanied by chronic autoimmune thyroiditis (22). Similarly, in our study, regardless of TSH level, anti-TPO(+) pregnant women were found to have higher abortion incidence (20% vs. 9.4%). It is stated in a meta-analysis that; the thyroid autoantibodies' existence, even if normal thyroid hormone status increased the risk of spontaneous abortion by 2-3 times compared to women without antibodies (23,24).
A meta-analysis of 4 observational studies examined 1098 women with in vitro fertilization(IVF); the risk of abortion was found to be two times higher in anti-TPO(+) euthyroid patients than in anti-TPO(-) patients (RR 1.99, 95% CI 1.42-2.78) (25). Temur et al. didn't find a statistically significant difference in terms of abortion between pregnancies with thyroid disease and euthyroid cases in their study(26). In our study, although we detected an increased risk of miscarriage for pregnancies with a high TSH level and anti-TPO positivity, this difference was not statistically significant. However, meaningful results might be obtained if the number of samples was increased.

In a prospective study, 115 anti-TPO(+) women were randomized (half were treated with T4, and the other half were untreated), and they were all compared to 869 anti-TPO(-) women. Miscarriage incidence were 3.5% in the anti-TPO(+) treated group, 2.4% in anti-TPO(-) group, and 13.8% in the anti-TPO(+) untreated group (27).

Whereas thyroid autoantibody-positive women have a risk of abortion of 13-22%, autoantibody-negative euthyroid women have a risk of 3.3-8.4% (28). In our study, similar rates were found respectively (20-24% vs. 7.8%).

Table 2-Comparison of the perinatal outcomes between subgroups.

	Control group (n = 108)		**Study group (n = 102)**		**P Value**
	Anti TPO(-) n=103	**Anti TPO(+) (n=5)**	**Anti TPO(-) (n=77)**	**Anti TPO(+) (n=25)**	
Pregnancy loss	7.8% (n=8)	**0% (n = 0)**	11.7% (n=9)	**24% (n=6)**	**p <0.05**
Hyperemesis gravidarum	33,0% (n=34)	40,0% (n=2)	35,1% (n=27)	36,0% (n=9)	p >0.05
Ongoing pregnancy	N=95	N=5	N=68	N=19	
Hypertensive disorders	2,1% (n=2)	0,0% (n=0)	1,5% (n=1)	0,0% (n=0)	
• **GHT**					p >0.05
• **preeclampsia**	3,2% (n=3)	0,0% (n=0)	4,4% (n=3)	0,0% (n=0)	p >0.05
Gestational diabetes	7,4% (n=7)	0,0% (n=0)	8,8% (n=6)	0,0% (n=0)	p >0.05
Prelabor rupture of membranes (PROM):	15,8% (n=15)	0,0% (n=0)	11,8% (n=8)	20,0% (n=4)	p >0.05
Placental abruption	0% (n = 0)	0% (n = 0)	0% (n = 0)	0% (n = 0)	p >0.05
Preterm birth	7,8% (n = 8)	0,0% (n = 0)	5,2% (n = 4)	8,0% (n = 2)	p >0.05
Delivery style					
• **cesarean section(CS)**	55,3% (n = 57)	80,0% (n = 4)	50,6% (n = 39)	44,0% (n = 11)	p >0.05
• **vaginal delivery(VD)**	36,9% (n = 38)	20,0% (n = 1)	37,7% (n= 29)	32,0% (n= 8)	p >0.05
• **Pregnancy loss**	7,8% (n = 8)	0,0% (n = 0)	11,7% (n = 9)	24% (n = 6)	p >0.05
Birth weight					
• **low birth weight (LBW)**	4,2% (n = 4)	0,0% (n = 0)	2,9% (n = 2)	0,0% (n = 0)	p >0.05
• **normal birth weight**	88,4% (n = 84)	80,0% (n = 4)	95,6% (n = 65)	100,0 (n= 19)	p >0.05
• **macrosomic**	7,4% (n = 7)	20,0% (n = 1)	1,5% (n = 1)	0,0% (n = 0)	p >0.05
Shoulder dystocia	7,7% (n = 3)	0,0% (n = 0)	0,0% (n = 0	0,0% (n = 0)	p >0.05
Newborn intensive care needs	12,6% (n = 12)	20,0% (n = 1)	11,8% (n = 8)	5,3% (n = 1)	p >0.05
Postpartum hemorrhage(PPH):	20,0% (n = 19)	0,0% (n = 0)	22,1% (n = 15)	42,1% (n = 8)	p >0.05

Interestingly, in our study, regardless of TSH levels, when pregnancy loss and smoking relationships were evaluated, 63.6% (n = 23) of all abortions were in the non-smoking group, while 36.4% were in the smoking

group. This difference was statistically significant (P <0.05). Negro et al. evaluated the relationship between pregnancy loss risk and TSH levels, and the smoking rate was higher in groups without pregnancy loss (21). From this point of view, smoking seems to be a protective factor.

On the other hand, in our study, 8.3% of non-smoker pregnant women had abortions, while 20.5% of smokers had abortions. Non-smokers were likelier to have no pregnancy loss [OR: 1,3 95% CI (0.94-1.8)]. Smoking's effects are widely accepted information, and it is conversant as a risk factor for abortion in the existence of thyroid autoantibodies. In abortion mechanisms related to autoimmunity, smoking may suppress immunity and prevent some of these abortions. The relationship should be clarified by doing more detailed studies on this subject.

***Hyperemesis gravidarum*:** Akdemir et al. evaluated thyroid dysfunctions in hyperemesis gravidarum and found significantly lower TSH values in the hyperemesis group (29). Our study didn't support this.

Panesar et al. found that anti-TPO levels were higher in pregnant women with hyperemesis gravidarum (30). Similarly, in our study, anti-TPO positivity was higher in pregnant women who developed hyperemesis than in non-hyperemesis patients (15.3% vs.13.8%), but statistical significance wasn't obtained. Fell et al. evaluated the risk factors of hyperemesis (31), and smoking was reported to decrease the risk of hyperemesis. Still, in our study, hyperemesis was observed at a higher rate in the smoker group, but the result wasn't statistically significant (38.5% vs. 33.1%).

***Postpartum hemorrhage*:** PPH was detected in 22.22% (4/18) diagnosed with hypothyroidism in a study of 633 women by Bostanci et al. (32). A similar rate of 27.3% was found in our case group. Leung et al. found zero PPH cases in the subclinical hypothyroid group (33).

It is anticipated that complications of hypothyroidism may be more moderate in subclinical hypothyroidism. There are many studies on this subject. In the FASTER study, Cleary-Goldman et al. didn't find a consistent result in the pattern of their undesirable consequences with maternal subclinical hypothyroidism, and subclinical hypothyroidism wasn't related to a rise in complications, like preterm labor, preterm delivery, abortion, and EMR (34). On the other hand, Casey et al. found subclinical hypothyroidism in 2.3% of 17,298 women screened before the first half of pregnancies. They also found that the rates of preterm delivery, ablation placenta, and NICU needs were higher than the control group (35). In our study, if we accept pregnant women as having subclinical hypothyroidism with TSH> 2.5 mU/L, the incidence of abortion, PPH, and PROM appears to be increased in the subclinical hypothyroid group.

There are different opinions on the reference ranges that should be used to diagnose subclinical hypothyroidism; Soldin et al. said that "to define aberrancies in thyroid hormone levels for the pregnancy process, it was stated that it is necessary to define reference intervals specific to each trimester, and these reference intervals will be different in iodine-insufficient populations" (22).

The ATA stated that "trimester-specific reference intervals should be used for TSH due to changes in thyroid physiology during pregnancy" in both the 2011 and 2017 guidelines (36,37). Pearce et al. said that the TPO-Ab status of pregnant women should be revised when creating trimester-specific reference intervals since increased TPO-Ab is related to rised TSH and low T4 levels. (38). Mandel et al. emphasized the need for more longitudinal studies on TSH levels during pregnancy without evidence of autoimmune thyroid diseases in areas of adequate iodine to improve trimester-specific TSH reference intervals (39).

Soldin et al. reported that the stimulative impact of hCG on the thyroid tissue during normal pregnancy at the end of the first trimester induces partial TSH suppression and falls below the non-pregnant reference range (mean 0.89 mU/L) (40). They also stated that the trimester-specific TSH concentrations significantly differed between the first and third trimesters (all P ≤ 0.05). But between the second and third trimesters, ıt wasn't significantly different from each other (all P> 0.25) (40-42).

The reference range for TSH in the population without autoimmune antibody and iodine sufficiency was found using immune assays as 0.24–2.99 mU/L for the first trimester, 0.46–2.95 mU/L for the second trimester, and 0.43–2.78 mU/L for the third trimester (22).

Dashe et al. calculated the average reference interval for TSH and stated that TSH was significantly decreased during the first trimester, and this reduce was higher in twin pregnancies (P <0.001). They set an upper limit of approximately 4.0 MoM for single pregnancies in the first trimester and 3.5 MoM for twins (43). First-trimester-specific reference ranges were presented in several studies (43-46).

Some of the complications in our anti-TPO+ group may haven't occurred due to the low number of cases in these subgroups. When we started this study, a guide published by the ATA in 2011 discussed the question of the first-trimester TSH interval in pregnant women for TSH (36). For the population with average iodine intake, the specified trimester-specific reference range should be applied (LEVEL-B). If the trimester-specific reference range for TSH isn't available in the laboratory, for the 1st, second and third trimesters, it should be respectively 0.1-2.5 mU/L; 0.2-3.0 mU/L; 0.3-3.0 mU/L be used as a reference range at LEVEL I. After that, the ATA published a new guideline in 2017 and said that "more recent studies on pregnant women in Asia, India, and the Netherlands have demonstrated only a modest reduction in the upper reference limit". They revised their recommendation as follows: " the panel recommends using the following trimester-specific ranges and cutoffs when local assessments are unavailable. In the first trimester, the lower reference range of TSH can be reduced by approximately 0.4 mU/L,

while the upper reference range is reduced by approximately 0.5 mU/L. For the typical patient in early pregnancy, this corresponds to a TSH upper reference limit of 4.0 mU/L" (37).

The degree of the decreasing shift in the TSH reference range for pregnancy is changing in different racial and ethnic groups (37,47). Other studies on diverse groups suggest a mild decrease in the upper limit of normal TSH of only 0.5 to 1.0 mU/L (48,49). Finally, the Turkish Society of Endocrinology and Metabolism recommends the use of Trimester-specific TSH references during pregnancy in the "diagnosis and treatment guideline for thyroid diseases" published in 2020; if this is not possible, 0.1-2.5 mU/L for the first trimester; 0.2-3.0 mU/L for the second trimester; of 0.3-3.0 mU/L values for the third trimester were recommended, and these recommendations are still up-to-date (50).

According to all these studies, TSH values in pregnant women seem to be lower than in the non-pregnant population. However, these values may differ from society to society, especially in areas of iodine insufficiency, depending on whether the pregnancy is multiple or not. It is understood that anti-TPO positivity can change these reference ranges.

It can be interpreted that thyroid function tests depend on the number of fetuses and gestational weeks. The person's antibody status determines the community-specific nomograms, which significantly improve the detection of the disease. Pregnancy has many effects on thyroid function, with significant changes in iodine metabolism and clearance and serum thyroid-binding protein levels. This effect is more pronounced in areas of variable iodine deficiency. When confronted with the demands of pregnancy, the maternal thyroid gland should have adequate iodine support, be disease-free, and be capable of responding adequately (19).

In our study, the incidence of pregnancy loss in the group with TSH>2.5 mU/L and anti-TPO(+)was statistically significantly higher than in the group with TSH <2.5 mU/L and anti-TPO(+). High TSH and/or anti-TPO(+) status appears to have an increased risk among all groups in terms of pregnancy loss incidence. Although the group with the highest incidence of developing PROM was the subgroup with TSH> 2.5 mU/L and anti-TPO(+), no statistically significant difference was found. A statistically significant difference might be achieved by increasing the number of cases in this group.

The incidence of postpartum hemorrhage increases in groups with both TSH>2.5 mU/L and anti-TPO (+). When evaluated in general terms, anti-TPO positivity increases the risks when TSH >2.5 mU/L and complications have a high incidence. Antibody positivity doesn't increase the risks in groups with TSH <2.5 mU/L.

Leading endocrine and thyroid societies such as the Endocrine Society and the ATA don't recommend routine first-trimester TSH screening in non-risky pregnant women. Although there are articles on routine anti-TPO screening, the general acceptance is that there is no need for routine screening in the first trimester of pregnant women.

It seems that the debate will continue about the issue of diagnosis and treatment according to trimester-specific reference intervals in the definition of subclinical hypothyroidism in TFT, for which routine screening isn't recommended.

Because of our study is a prospective cohort study, some patients were excluded. Although there weren't many, this is a limitation. Furthermore, although the number of pregnant women in our case and control groups is sufficient in statistical calculations, when we divided them into subgroups, some complications with low incidence had a low number of pregnant women in the subgroups. This may have caused some complications to be absent in these subgroups or to be sufficient in number and not show statistically significant results. Unfortunately, this is another limitation of our study.

Also, since Turkey is a region with a medium-high deficiency of iodine, there are no reference ranges determined for the country, so the practice in this subject is up to the choice of the clinician. Consequently, to recommend using 0.1-2.5 mU/L mU/L as the first-trimester reference range in pregnant women, there is a need for community-specific nomograms that should be developed by considering the number of fetuses, gestational weeks, and the antibody status of the patient.

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References

1.Cooper DS, Biondi B. Subclinical thyroid disease. Lancet 2012; 379:1142.

2.Ballabio M, Poshychinda M, Ekins RP. Pregnancy-induced changes in thyroid function: role of human chorionic gonadotropin as a putative regulator of maternal thyroid. J Clin Endocrinol Metab 1991;73:824.

3.Yamazaki K, Sato K, Shizume K, Kanaji Y, Ito Y, Obara T et al. Potent thyrotropic activity of human chorionic gonadotropin variants in terms of 125I incorporation and de novo synthesized thyroid hormone release in human thyroid follicles. J Clin Endocrinol Metab 1995; 80:473.

4.Portmann L, Hamada N, Heinrich G, DeGroot LJ. Anti-thyroid peroxidase antibody in patients with autoimmune thyroid disease: possible identity with anti-microsomal antibody. J Clin Endocrinol Metab. 1985; 61: 1001-3.

5.Czarnocka B, Ruf J, Ferrand M, Carayon P, Lissitzky S. Purification of the human thyroid peroxidase and its identification as the microsomal antigen involved in

autoimmune thyroid diseases. FEBS Lett. 1985;190:147-52.
6.World Health Organization.Assessment of iodine deficiency disorders and monitoring their elimination. A guide for program managers. Geneva: WHO,2007:1-108
7.Laurberg P, Cerqueira C, Ovesen L, Rasmussen LB, Perrild H, Andersen S et al. Iodine intake as a determinant of thyroid disorders in populations. Best Pract Res Clin Endocrinol Metab. 2010;24:13-27.
8.Stockigt JR. Free thyroid hormone measurement. A critical appraisal. Endocrinol Metab Clin North Am. 2001; 30:265-89
9.Benvenga S, Cahnmann HJ, Robbins J. Characterization of thyroid hormone binding to apolipoprotein-E: localization of the binding site in the exon 3-coded domain. Endocrinology. 1993;133:1300-5.
10.Dufour DR. Laboratory tests of thyroid function: uses and limitations. Endocrinol Metab Clin North Am. 2007; 36:579-94.
11.Caldwell G, Kellett HA, Gow SM, Beckett G J, Sweeting V M, Seth J et al. A new strategy for thyroid function testing. Lancet. 1985;1:1117-9
12.Surks MI, Ortiz E, Daniels GH, Sawin CT, Col NF, Cobin RH et al. Subclinical thyroid disease: scientific review and guidelines for diagnosis and management. JAMA. 2004;291:228-38
13.Walsh JP, Bremner AP, Feddema P, Leedman PJ, Brown SJ, O'Leary P. Thyrotropin and thyroid antibodies as predictors of hypothyroidism: a 13-year, longitudinal study of a community-based cohort using current immunoassay techniques. J Clin Endocrinol Metab. 2010;95:1095-104.
14.Persani L, Borgato S, Romoli R, Asteria C, Pizzocaro A, Beck-Peccoz P.Changes in the degree of sialylation of carbohydrate chains modify the biological properties of circulating thyrotropin isoforms in various physiological and pathological states. J Clin Endocrinol Metab. 1998;83:2486-92.
15.Persani L, Ferretti E, Borgato S, Faglia G, Beck-Peccoz P.Circulating thyrotropin bioactivity in sporadic central hypothyroidism. J Clin Endocrinol Metab. 2000;85:3631-5.
16.Negro R, Schwartz A, Gismondi R, Tinelli A, Mangieri T, Stagnaro-Green A. Universal screening versus case finding for detection and treatment of thyroid hormonal dysfunction during pregnancy. J Clin Endocrinol Metab. 2010;95:1699-707.
17.Stagnaro-Green A, Abalovich M, Alexander E, Azizi F, Mestman J, Negro R et al. Guidelines of the American Thyroid Association for the diagnosis and management of thyroid disease during pregnancy and postpartum. Thyroid 2011; 21:1081.
18.Lazarus JH. Thyroid disorders associated with pregnancy: etiology, diagnosis, and management.Treat Endocrinol. 2005;4:31-4
19.Monzani F, Del Guerra P, Caraccio N, Pruneti CA, Pucci E, Luisi M et al. Subclinical hypothyroidism: neurobehavioral features and beneficial effect of L-thyroxine treatment. Clin Investig. 1993;71:367-71.
20.Baker VL, Rone HM, Pasta DJ, Nelson HP, Gvakharia M, AdamsonGD. Correlation of thyroid-stimulating hormone (TSH) level with pregnancy outcome in women undergoing in vitro fertilization. Am J Obstet Gynecol. 2006;194:1668-74.
21.Negro R, Schwartz A, Gismondi R, Tinelli A, Mangieri T, Stagnaro-Green A. Increased pregnancy loss rate in thyroid antibody negative women with TSH levels between 2.5 and 5.0 in the first trimester of pregnancy. J Clin Endocrinol Metab. 2010; 95:E44.
22.Soldin OP. Thyroid function testing in pregnancy and thyroid disease: trimester-specific reference intervals. The Drug Monit. 2006;28:8-11.
23.Chen L, Hu R. Thyroid autoimmunity and miscarriage: a meta-analysis. Clin Endocrinol (Oxf) 2011;74:513.
24.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 evidence. British Medical Journal. 2011; 342:d2616.
25.Toulis KA, Goulis DG, Venetis CA, Kolibianakis EM, Negro R, Tarlatzis BC et al. Risk of spontaneous miscarriage in euthyroid women with thyroid autoimmunity undergoing IVF: a meta-analysis. Eur J Endocrinol. 2010;162:643.
26.Temur M, Cengiz H, Arici B., Yaşar L, Özdemir İA. Erken gebelikte tiroid fonksiyon bozukluğunun tespiti. Gazi Medical Journal. 2012; 23:6-9.
27.Negro R, Formoso G, Mangieri T, Pezzarossa A, Dazzi D, Hassan H. Levothyroxine treatment in euthyroid pregnant women with autoimmune thyroid disease: effects on obstetrical complications. J Clin Endocrinol Metab. 2006;91:2587-91.
28.Abramson J, Stagnaro-Green A., Thyroid antibodies and fetal loss: an evolving story.Thyroid. 2001;11:57-63.
29.Akdemir N, Bilir C. Thyroid dysfunction in hyperemesis gravidarum: a study in Turkish pregnant women. J Turkish-German Gynecol Assoc. 2011;12:140-3.
30.Panesar NS, Chan KW, Li CY, Rogers MS. Status of anti-thyroid peroxidase during normal pregnancy and in patients with hyperemesis gravidarum. Thyroid. 2006;16:481-4.
31.Fell DB, Dodds L, Joseph KS, Allen VM, Butler B. Risk factors for hyperemesis gravidarum requiring hospital admission during pregnancy. Obstet Gynecol. 2006; 107:277.
32.Bostancı MS, Taşkesen F. Gebelikte tiroid fonksiyon bozuklukları ve sonuçlarının değerlendirilmesi. Klinik ve Deneysel Araştırmalar Dergisi. 2011;2:196-201.
33.Leung AS, Millar LK, Koonings PP, Montoro M, Mestman JH. Perinatal outcome in hypothyroid pregnancies. Obstet Gynecol. 1993; 81:349.
34.Cleary-Goldman J, Malone FD, Lambert-Messerlian G, Sullivan L, Canick J, Porter TF, et al. Maternal thyroid hypofunction and pregnancy outcome. Obstet Gynecol. 2008; 112:85.

35.Casey BM, Dashe JS, Wells CE, McIntire DD, Byrd W, Leveno KJ et al. Subclinical hypothyroidism and pregnancy outcomes. Obstet Gynecol. 2005;105:239-45.
36.Stagnaro-Green A, Abalovich M, Alexander E, Azizi F, Mestman J, Negro R et al. Guidelines of the American Thyroid Association for the diagnosis and management of thyroid disease during pregnancy and postpartum. Thyroid. 2011; 21:1081.
37.Alexander EK, Pearce EN, Brent GA, Brown RS, Chen H, Dosiou C et al. 2017 Guidelines of the American Thyroid Association for the diagnosis and management of thyroid disease during pregnancy and the postpartum. Thyroid. 2017; 27:315.
38.Pearce EN, Oken E, Gillman MW, Lee SL, Magnani B, Platek D, et al., Association of first-trimester thyroid function test values with thyroperoxidase antibody status, smoking, and multivitamin use. Endocr Pract. 2008;14:33-9.
39.Mandel SJ, Spencer CA, Hollowell JG. Are detection and treatment of thyroid insufficiency in pregnancy feasible?. Thyroid. 2005;15:44-53.
40.Soldin OP, Tractenberg RE, Hollowell JG, Jonklaas J, Janicic N, Soldin SJ. Trimester-specific changes in maternal thyroid hormone, thyrotropin, and thyroglobulin concentrations during gestation: trends and associations across trimesters in iodine sufficiency. Thyroid. 2004;14:1084-90.
41.Soldin OP, Hilakivi-Clarke L, Weiderpass E, Soldin SJ. Trimester-specific reference intervals for thyroxine and triiodothyronine in pregnancy in iodine-sufficient women using isotope dilution tandem mass spectrometry and immunoassays. Clin Chim Acta. 2004;349:181-9.
42.Soldin OP, Tractenberg RE, Soldin SJ. Differences between measurements of T4 and T3 in pregnant and nonpregnant women using isotope dilution tandem mass spectrometry and immunoassays: are there clinical implications? Clin Chim Acta. 2004;347:61-9.
43.Dashe JS, Casey BM, Wells CE, McIntire DD, Byrd EW, Leveno KJ et al. Thyroid-stimulating hormone in singleton and twin pregnancy: importance of gestational age-specific reference ranges. Obstet Gynecol. 2005; 106:753-7.
44.Spencer C, Lee R, Kazarosyan M, Bergoglio L, Braverman L, Mereshian P et al. Thyroid reference ranges in pregnancy: Studies on an iodine sufficient cohort. Thyroid. 2005;15(Supp 1):1-16.
45.Walker JA, Illions EH, Huddleston JF, Smallridge RC. Racial comparisons of thyroid function and autoimmunity during pregnancy and the postpartum period. Obstet Gynecol. 2005;106:1365-71.
46.Haddow JE, Knight GJ, Palomaki GE, McClain MR, Pulkkinen AJ. The reference range and within-person variability of thyroid stimulating hormone during the first and second trimesters of pregnancy. J Med Screen. 2004;11:170-4.
47.Medici M, Korevaar TI, Visser WE, Visser TJ, Peeters RP. Thyroid function in pregnancy: what is normal? Clin Chem. 2015; 61:704.
48.Korevaar TI, Schalekamp-Timmermans S, de Rijke YB, Visser WE, Visser W, de Muinck Keizer-Schrama SM et al. Hypothyroxinemia and TPO-antibody positivity are risk factors for premature delivery: the generation R study. J Clin Endocrinol Metab 2013; 98:4382.
49.Li C, Shan Z, Mao J, Wang W, Xie X, Zhou W et al. Assessment of thyroid function during first-trimester pregnancy: what is the rational upper limit of serum TSH during the first trimester in Chinese pregnant women? J Clin Endocrinol Metab. 2014; 99:73.
50.Akarsu E, Alagöl F, Altun B, Aral F, Ayvaz G, Boztepe, H et al. (2016). Türkiye Endokrinoloji ve Metabolizma Derneği. Tiroid hastalıkları tanı ve tedavi klavuzu. ISBN:978-625-401-061-3.
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