ORIGINAL ARTICLE

Thyroid function test reference ranges in the first trimester of gestation and pregnancy outcomes: Protocol and preliminary results for cohort population-based study Isfahan, Iran

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Background: This paper presents the protocol and primary findings of pregnancy cohort population-based study in Isfahan, Iran. **Materials and Methods:** In this cohort, 418 pregnant and 438 nonpregnant women were enrolled. In the first phase, serum concentrations of thyroid-stimulating hormone (TSH), free thyroxine (FT4), free triiodothyronine (FT3), thyroid peroxidase antibody, and urinary iodine concentration (UIC) were measured. Furthermore, the thyroid ultrasound was also performed. According to the results of thyroid function tests in the first phase, local reference range for TSH, FT4, and FT3 in pregnant and nonpregnant women are determined. The 2.5th and 97.5th percentiles are determined as limits of the reference ranges. In the second phase, all pregnant women underwent prenatal care visits in each trimester and they followed for 7 days after delivery and the pregnancy outcomes data are reported. **Results:** The mean ± standard deviation for TSH, FT4, FT3, and UIC in the first trimester of gestation was 1.84 ± 1.32 mIU/L, 1.01 ± 0.15 ng/dL, 4.50 ± 0.64 pmol/L, and 172.0 ± 90.29 μg/L, respectively. In nonpregnant women, these values for TSH, FT4, FT3, and UIC were 2.58 ± 1.77 mIU/L, 1.10 ± 0.21 ng/dL, 4.49 ± 0.57 pmol/L, and 190.0 ± 109.6 μg/L, respectively. **Conclusions:** The results of the present study could contribute to establish a local thyroid function tests reference ranges in the first trimester of pregnancy. It could possibly be effective on making a local reference value to prevent of thyroid disease misdiagnosis during pregnancy and adverse pregnancy outcomes.

Key words: Cohort population-based study, Iran, pregnancy outcomes, reference range, thyroid function

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INTRODUCTION

Pregnancy is one of the most important periods in a woman's life. During this period, critical but reversible physiological changes take place that can affect the thyroid economy and function tests.[1] Due to these changes, thyroid hormone secretion increases up to 50%, by the maternal thyroid.[2] In the early stage of gestation, thyroid gland is stimulated by human chorionic gonadotropin hormone (HCG). HCG has 85% structural homology.[3,4] This leads to secretion of free thyroxine (FT4) when HCG concentration increases.[5] Due to gestational-induced alteration in thyroid physiology, thyroid-stimulating hormone (TSH) concentration reduces during the first trimester of pregnancy. Therefore, normal TSH values at early gestation may be lower than for nonpregnant women. As a result, using nonpregnant reference ranges to interpret the thyroid function tests in pregnancy period results in misdiagnosis.[6] As a report of several studies, thyroid dysfunction can lead to maternal and fetal adverse effects such as miscarriage, preterm delivery, preeclampsia, eclampsia, early placental abruption,[7,8] and fetal death.[9]

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The American Thyroid Association (ATA) recommended the specific reference range for TSH in the first trimester as 0.1–2.5 mIU/L.[10] A number of studies have been reported that their local reference range was different from one that has been recommended by the ATA.[11-13] Therefore, the guidelines of the Endocrine Society, ATA, and European Thyroid Association recommended that trimester-specific reference ranges for thyroid function tests should be separately established and used for each geographic region.[10,14,15] Since there were the limited data about specific reference ranges in Iranian pregnant women in different geographic areas, the present study was designed to determine specific reference ranges for thyroid function tests in healthy pregnant women in the first trimester of gestation. We also compared the pregnancy outcomes using local and ATA first trimester-specific reference range.

Objectives

Our main objective is to determine the first trimester-specific reference ranges of TSH, FT4, and free triiodothyronine (FT3). The secondary objectives of our study are to investigate the maternal, fetal, and neonatal outcomes based on local trimester-specific reference ranges and ATA-recommended trimester-specific reference ranges. Therefore, we estimated the frequency of thyroid dysfunction, thyroid peroxidase positivity, and urinary iodine deficiency among pregnant and nonpregnant women.

Study design

This study was a population-based cohort study including two phases. The first phase of the study was a population-based cross-sectional study in pregnant and nonpregnant women, and the second phase was a longitudinal study. The reference population in the first phase of the study – determination of reference range for thyroid function tests – is pregnant and nonpregnant women, and in the second phase, only pregnant women were examined for the outcomes.

Sampling framework

The samples were selected from urban health centers I and II of Isfahan and the private gynecology and midwifery clinics. Participants were recruited using convenience sampling based on the inclusion and exclusion criteria of the study, and the consent from all pregnant and nonpregnant women was obtained.

First phase

The eligibility criteria are age ranged from 15 to 45 years at sampling time, gestational age based on the 1st day of the last menstrual period (LMP) up to completion of 14th weeks of gestation,[16] and single gestation. If LMP was not clear, the pregnancy ultrasound requested. Women with preexisting thyroid disorders, goiter or nodules, autoimmune or chronic diseases, positive thyroid peroxidase antibody (TPOAb), medication history of levothyroxine, propylthiouracil, or methimazole, and any medications affecting thyroid function tests were excluded from the study. In the first phase of the study, after matching for age and gravida, 880 women (436 pregnant and 444 nonpregnant women), who attending prenatal, mother, and child care clinics (clinics of Isfahan University of Medical Sciences and private clinics) were enrolled [Figure 1].

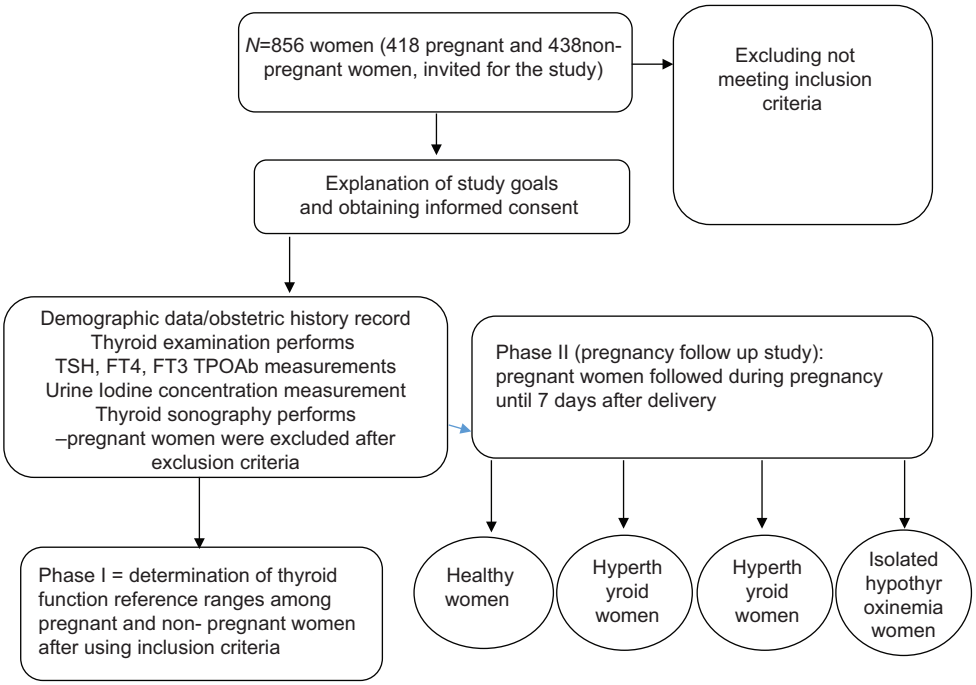

Figure 1: The flowchart of study design (Phase I and Phase II)

The study protocol and goals were explained to the participants, and written consent forms were obtained from all pregnant and nonpregnant women. The demographic characteristics (maternal age at present pregnancy, occupation, and educational level), reproductive history (gravida, para, number of abortion and/or fetal death, number of living child and/or neonatal death, methods of previous deliveries, previous pregnancy complications), history of taking any medications, previous thyroid dysfunction of herself or her first-degree relatives, and past or present history of autoimmune diseases and diabetes were taken through interviews and recorded. Symptoms and signs of hypothyroidism and hyperthyroidism including nervousness, fatigue, weakness, increased perspiration, heat intolerance, tremor, appetite change, weight change, history of menstrual disturbances, warm skin, moist skin, and smooth skin were assessed in all participants. Thyroid examination was performed. Moreover, height was measured without shoes to the nearest 0.1 cm using a tape meter against a wall whereas body weight was measured to the nearest 0.1 kg on an electronic scale which was placed on flat ground and participants wearing light clothing and standing motionless. Body mass index was calculated by body weight (kg) by height (m^2).

After taking medical history and physical examination, 5 ml of venous blood was obtained from each participant, and serum levels of TSH, FT4, FT3, and TPOAb were measured at Isfahan Endocrine and Metabolism Research Center Laboratory. All samples were taken within 7:30 a.m.–10:30 a.m. Thyroid peroxidase antibody was defined positive for values higher than 60 IU/ml.

Finally, urine samples were collected in falcon tubes, and urinary iodine level was measured by acid digestive method[17] [Table 1]. Iodine sufficiency in pregnant and nonpregnant women is defined as UIC ≥150 μg/L and UIC ≥100 μg/L, respectively. The thyroid ultrasound was performed using Philips Affiniti 70 Ultrasound System (made in the Netherlands) and using a superficial probe at 4–12 MHz. The volume of each lobe is calculated by the formula (mL) =0.000479 × length (mm) × width (mm) × thickness (mm). Pregnant and nonpregnant women with thyroid volume >30 mL by ultrasonography were excluded in this phase.

Table 1: Laboratory tests, methods were used for thyroid testing

Tests	Methods	Kits	Equipment
TSH	Immunoassay	SIMENS	ADVIA centaur CP/ SIMENS 2010-American
FT4	Immunoassay	SIMENS	ADVIA centaur CP/ SIMENS 2010-American
FT3	Immunoassay	SIMENS	ADVIA centaur CP/ SIMENS 2010-American
TPOAb	Immunoassay	SIMENS	ADVIA centaur CP/ SIMENS 2010-American
UIC	Digestive method	Sandell-Kolthoff reaction	ELISA reader

TSH=Thyroid stimulating hormone; FT4=Free thyroxine; FT3=Free triiodothyronine; TPOAb=Thyroid peroxidase antibody; UIC=Urinary iodine concentration

Pregnant and nonpregnant women with abnormal test results were referred to obstetricians or endocrinologists. The Ethics Committee of Isfahan University of Medical Sciences approved the design and protocol of the study according to the Declaration of Helsinki.

Second phase (follow-up study)

According to the results of thyroid function tests in the first phase, local reference range for TSH, FT4, and FT3 in pregnant and nonpregnant women is determined. Based on upper and lower limit of TSH and FT4, subclinical and overt hypothyroidism, subclinical and overt hyperthyroidism, and isolated hypothyroxinemia are defined.

All pregnant women underwent prenatal care visits in each trimester (first trimester ≤14 weeks, second trimester ≥15 weeks until the completion of 28 weeks of gestation, and third trimester ≥29 weeks until the completion of 42 weeks of gestation).[16] All of the pregnant women were followed within 7 days after delivery. Information of intrapartum, early postpartum, malformation of the neonate organs, and mother and neonate discharge from hospital wards are collected from the hospitals discharge units and recorded.

Exposure and outcome definitions

In the present study, thyroid disorders are considered as exposures. The maternal, fetal, and neonatal outcomes including miscarriage, preeclampsia, gestational diabetes mellitus (GDM), placenta previa, placenta abruption, premature rupture of membrane, preterm delivery, stillbirth, low birth weight, high birth weight, neonatal Intensive Care Unit admission, Apgar score <7, postpartum hemorrhage (PPH), early neonatal deaths ≤7 days after birth, and presence of neonatal hypothyroidism are also recorded. Premature neonates are followed until 10 weeks after birth, and the presence of neonatal hypothyroidism is also recorded.

Abortion is defined as a pregnancy loss with an upper limit of 20th completed weeks of gestation.[18] Preeclampsia is defined as persistent elevated blood pressure (systolic pressure ≥140 mmHg, diastolic pressure ≥90 mmHg) with proteinuria.[19] GDM is defined as a plasma glucose concentration ≥95 mg/dL after fasting, ≥180 mg/dL at 1 h after a 100-g oral glucose tolerance test (OGTT), and/or ≥155 mg/dL at 2 h after a 100-g OGTT, regardless of gestational age.[20] Placenta previa is defined as when the placenta is inserted partial or complete into the

lower segment of the uterus.[21] Placental abruption is defined as the placental lining separation from the uterine before delivery.[22] Preterm delivery is defined as pregnancy termination before 37 completed weeks of gestation can be subgrouped as extreme (<28 weeks), severe (between 28 and 32 weeks), and moderate or "near-term" (32–36 weeks).[23] Preterm premature rupture of membranes is the spontaneous rupture of the fetal membranes during pregnancy before 37 weeks' gestation in the absence of regular painful uterine contractions.[24] Stillbirth is defined as fetal death at or after 20-28 weeks of gestation.[25] PPH is commonly defined as a blood loss of 500 ml or more within 24 h after birth.[26] Low birth weight refers to birth weight below 2500 g[27] and high birth weight is defined as birth weight above 4000 g.[28]

Sample size

According to the lower limit of TSH in the first trimester of gestation reported from a study in Iran,[13] considering 0.2, adding 30% loss of follow-up (not providing hormonal profile), a total sample of 400 per pregnant and nonpregnant women group are estimated.

Statistical analyses

The data were statistically analyzed by Statistical Package for the Social Science version 16 (SPSS version, IBM, Chicago, IL, USA). Markers of thyroid function were examined for normality by Kolmogorov–Smirnov test. Numerical variables as mean (standard deviation [SD]) and median and range and nonnumerical variables as number (percentage) were also calculated. Chi-square and Kruskal–Wallis tests were used to compare nonnumerical variables between the two groups.

Plan for future analyses

For each thyroid function test, median, 2.5th, 5th, 10th, 90th, 95th, 97.5th percentiles, and first and third quartiles are calculated. The 2.5th and 97.5th percentiles are determined as limits of the reference ranges. The frequency of thyroid dysfunction, thyroid peroxidase positivity, and urinary iodine deficiency are also recorded. Mean differences between the study groups are tested by *t*-test and ANOVA. Median test and binomial test are used to compare differences, median, and selected percentiles between pregnant and nonpregnant women. Multiple logistic regression model is used to determine the risk factors of thyroid dysfunction. Variables that were significant at $P < 0.2$ on univariate analysis entered into the model. Significance level set as $P < 0.05$ for all tests.

RESULTS

A We examined 436 pregnant women (418 pregnant and 438 non–pregnant women) in the study. Mean ± SD age of the pregnant and non-pregnant women was 29.02 ± 5.01 and 29.50 ±4.90 years, respectively (range: 16–43 years) ($P = 0.107$). The median duration of gestation for pregnant women was 9 weeks and 6 days (minimum: 5 weeks; maximum: 14 weeks and 3 days). Median and minimum and maximum numbers of gravida in the pregnant and nonpregnant women were 2, 1, and 6, respectively.

The mean ± SD for TSH, FT4, and FT3 values in the first trimester of gestation was 1.84 ± 1.32 mIU/L, 1.01 ± 0.15 ng/dL, and 4.50 ± 0.64 pmol/L, respectively. In nonpregnant women, these values for TSH, FT4, and FT3 were 2.58 ± 1.77 mIU/L, 1.10 ± 0.21 ng/dL, and 4.49 ± 0.57 pmol/L, respectively. The mean ± SD for UIC in both of pregnant and nonpregnant women was 172.0 ± 90.29 µg/L and 190.0 ± 109.6 µg/L, respectively. About 14.10% and 18.72% of pregnant and nonpregnant women were passive smoker. More details of preliminary findings in the study groups are presented in Table 2.

Table 2: Descriptive characteristics, anthropometric measurements, thyroid function tests, and urinary iodine concentration of the study population

Variables	Pregnant women (*n*=418)	Nonpregnant women (*n*=438)	*P*
Age (years)*	29.02 (5.01)	29.50 (4.90)	0.107
Education**			
Illiterate	3 (0.7)	2 (0.5)	0.065
Primary school	27 (6.5)	33 (7.6)	
Under diploma	103 (24.8)	139 (31.8)	
Diploma	144 (34.6)	134 (35.2)	
University	138 (33.2)	109 (24.9)	
Current job**			
Housekeeper	382 (91.60)	412 (94.3)	0.116
Employment	31 (7.20)	15 (3.4)	
Other	5 (1.20)	10 (2.3)	
Height (m)*	1.60 (5.82)	1.61±5.75	0.001
Current weight (kg)*	64.49 (11.22)	66.01±12.03	0.056
Current BMI (kg/m^2)*	25.10 (4.26)	25.40±4.42	0.471
BMI category**			
Underweight (<18.5)	18 (4.3)	17 (3.9)	0.30
Normal (18.5-24.9)	192 (46)	190 (43.4)	
Overweight (25-29.9)	155 (37.2)	165 (37.6)	
Obesity (≥30)	52 (12.5)	66 (15.1)	
Smoking**			
Nonsmoker	357 (85.40)	337 (77.0)	0.020
Current smoker	0	1 (0.22)	0.52
Passive smoker	59 (14.10)	82 (18.72)	0.064
Tobacco user	2 (0.48)	18 (4.10)	0.001
TSH (mIU/L)*	1.84±1.32	2.58±1.77	0.001
FT4 (ng/dL)*	1.01±0.15	1.10±0.21	0.001
FT3 (pmol/L)*	4.50±0.64	4.49±0.57	0.697
UIC (µg/L)*	172.0±90.29	190.0±109.6	0.012

*Mean±SD, **Count (*n*) percentage. BMI=Body mass index; TSH=Thyroid-stimulating hormone; FT4=Free thyroxine; FT3=Free triiodothyronine; UIC=Urinary iodine concentration; SD=Standard deviation

DISCUSSION

The rationale for conducting the study, protocol, and preliminary findings of the research was reported in this manuscript. Thyroid dysfunction is a common condition in clinical practice in pregnant women and has significant maternal, fetal, and neonatal consequences.[29] Following the pregnancy-related changes in thyroid physiology, maternal thyroid dysfunction complications, especially hypothyroidism, are important to determine reference ranges for normal thyroid status during pregnancy. Since there is a wide variation of TSH values, there is no agreement on the optimal cut-off point for diagnosis and treatment of pregnant women with thyroid dysfunction.

Based on the ATA guidelines, in the first trimester of pregnancy, the reference range for TSH is 0.1–2.5 mIU/L.[10] The current study determines the reference ranges for thyroid function tests in pregnant women and also reports the reference range of these markers in nonpregnant women in Isfahan (Iran). In addition, maternal, fetal, and neonatal outcomes are reported in the second phase of the study. Several cohort, cross-sectional, and case–control studies have been conducted to determine the reference range of thyroid function in Iran. In studies conducted by Mehran *et al*.[13] and Azizi *et al*.,[30] the reference range of thyroid function tests has been reported in each trimester among pregnant woman as 5th and 95th percentiles in Tehran. In these two studies, pregnancy, fetal, and neonatal outcomes were not investigated, and considering the physiological changes in pregnancy, the researchers suggested that nonpregnant women should be sampled to be compared with pregnant women. In these studies, like our study, the criteria of National Academy of Clinical Biochemistry and National Health and National Examination Survey criteria were used to determine exclusion criteria.

In a study conducted by Mansourian *et al*.[31] in Gorgan, only TSH of 120 pregnant women was measured in the first trimester of pregnancy and the results were reported as mean and SD. Zarghami *et al*.[32] conducted their study in Tabriz and reported as mean and SD of thyroid function tests for pregnant women at different gestational age and nonpregnant women.

In recent years, some studies have assessed the influence of patterns of thyroid dysfunction on the risk of adverse maternal, fetal, and neonatal outcomes. These results are important in clinical practice because they provide new insights in potential consequences of applying nonpregnant reference ranges for pregnant population.[33]

In a study done in Italy, pregnancy loss was different in TPOAb-negative women with TSH level <2.5 mIU/L compared with pregnant women with TSH level between 2.5 and 5 mIU/L. This study demonstrated increased incidence of pregnancy loss in pregnant women with TSH level between 2.5 and 5 mIU/L although the rate of preterm birth was not significantly different.[34]

The results of a study in the USA described that the subclinical hypothyroidism before 20 weeks of gestation was associated with the higher rate of preterm delivery (4.0 vs 2.5% ,$P < 0.05$).[35] Increased in proportion of very preterm delivery (≥32 weeks) in women with TSH concentration >3.0 reported in another prospective study.[36] The results of a study in Shiraz in the second trimester of pregnancy showed that overt hypothyroidism (TSH >3 with low FT4 or TSH ≥10 mIU/L) increased risk of preterm delivery, and subclinical hypothyroidism (TSH level between 3 and 10 mIU/L) was associated with higher rate of intrauterine growth retardation (IUGR), low Apgar score, and maternal hyperthyroidism increased rate of IUGR.[37] In that study, pregnant women at 15–28 weeks of gestation were enrolled, and 2.5th, 25th, 50th, 75th, and 97.5th percentiles of TSH were calculated. The aim of the study was the prevalence of thyroid diseases and its outcomes in pregnancy.

CONCLUSIONS

As mentioned above, it seems that the reference ranges of thyroid function tests with attention to pregnancy outcomes should be determined. Despite the wide variation of TSH range, there is no agreement in the optimal cut-off point to diagnose and treat of pregnant women with thyroid dysfunction. Therefore, in the present cohort, we establish the gestational age-specific reference ranges for thyroid function tests during the first trimester of pregnancy. In addition, we compare maternal, fetal, and early neonatal outcomes in pregnant study population according to the reference range derived from the present study and the ATA. Our findings could have a significant impact on clinical practice of thyroid disorders during pregnancy.

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Conflicts of interest

There are no conflicts of interest.

REFERENCES

1. Moleti M, Lo Presti VP, Campolo MC, Mattina F, Galletti M, Mandolfino M, *et al.* Iodine prophylaxis using iodized salt and risk of maternal thyroid failure in conditions of mild iodine deficiency. J Clin Endocrinol Metab 2008;93:2616-21.
2. Lindberg BS, Johansson ED, Nilsson BA. Plasma levels of nonconjugated oestrone, oestradiol-17beta and oestriol during uncomplicated pregnancy. Acta Obstet Gynecol Scand Suppl 1974;32:21-36.
3. Glinoer D. The regulation of thyroid function in pregnancy: Pathways of endocrine adaptation from physiology to pathology. Endocr Rev 1997;18:404-33.
4. Morreale de Escobar G, Obregon MJ, Escobar del Rey F. Role of thyroid hormone during early brain development. Eur J Endocrinol 2004;151 Suppl 3:U25-37.
5. Moleti M, Trimarchi F, Vermiglio F. Thyroid physiology in pregnancy. Endocr Pract 2014;20:589-96.
6. Glinoer D, Spencer CA. Serum TSH determinations in pregnancy: How, when and why? Nat Rev Endocrinol 2010;6:526-9.
7. Ashoor G, Maiz N, Rotas M, Jawdat F, Nicolaides KH. Maternal thyroid function at 11 to 13 weeks of gestation and subsequent fetal death. Thyroid 2010;20:989-93.
8. Abalovich M, Gutierrez S, Alcaraz G, Maccallini G, Garcia A, Levalle O, *et al.* Overt and subclinical hypothyroidism complicating pregnancy. Thyroid 2002;12:63-8.
9. Lazarus JH. Thyroid function in pregnancy. Br Med Bull 2011;97:137-48.
10. 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;21:1081-125.
11. Dhatt GS, Jayasundaram R, Wareth LA, Nagelkerke N, Jayasundaram K, Darwish EA, *et al.* Thyrotrophin and free thyroxine trimester-specific reference intervals in a mixed ethnic pregnant population in the United Arab Emirates. Clin Chim Acta 2006;370:147-51.
12. Stricker R, Echenard M, Eberhart R, Chevailler MC, Perez V, Quinn FA, *et al.* Evaluation of maternal thyroid function during pregnancy: The importance of using gestational age-specific reference intervals. Eur J Endocrinol 2007;157:509-14.
13. Mehran L, Amouzegar A, Delshad H, Askari S. Hedayati M, Amirshekari G, *et al*. Trimester-specific reference ranges for thyroid hormones in Iranian pregnant women. J Thyroid Res 2013;6. Article ID 651517. Available from: http://dx.doi.org/10.1155/2013/651517.
14. Lazarus J, Brown RS, Daumerie C, Hubalewska-Dydejczyk A, Negro R, Vaidya B, *et al.* 2014 European thyroid association guidelines for the management of subclinical hypothyroidism in pregnancy and in children. Eur Thyroid J 2014;3:76-94.
15. De Groot L, Abalovich M, Alexander EK, Amino N, Barbour L, Cobin RH, *et al*. Management of thyroid dysfunction during pregnancy and postpartum: An endocrine society clinical practice guideline. J Clin Endocrinol Metab 2012;97:2543-65.
16. Cunninghan FG, Leveno KJ, Bloom SL, Hauth JC, Rouse DJ, Spong CY. Williams Obstetrics. 23rd ed. New York : McGraw Hill Medical; 2014.
17. Khazan M, Yaghmaei P, Behdadfar L, Daneshpour M, Hedayati M. Microwave digestion for urine iodine determination. Iran J Endocrinol Metab 2010;12:65-70.
18. Regan L, Rai R. Epidemiology and the medical causes of miscarriage. Baillieres Best Pract Res Clin Obstet Gynaecol 2000;14:839-54.
19. Mancia G, Fagard R, Narkiewicz K, Redon J, Zanchetti A, Böhm M, *et al.* 2013 ESH/ESC guidelines for the management of arterial hypertension: The task force for the management of arterial hypertension of the European society of hypertension (ESH) and of the European Society of Cardiology (ESC). Eur Heart J 2013;34:2159-219.
20. Sacks DB, Arnold M, Bakris GL, Bruns DE, Horvath AR, Kirkman MS, *et al.* Guidelines and recommendations for laboratory analysis in the diagnosis and management of diabetes mellitus. Clin Chem 2011;57:e1-e47.
21. Placenta Praevia, Placenta Praevia Accreta and Vasa Previa: Diagnosis and Management RCOG Green-top Guideline No. 27; January, 2011.
22. Tikkanen M. Placental abruption: Epidemiology, risk factors and consequences. Acta Obstet Gynecol Scand. 2011;90:140-9. doi: 10.1111/j.1600-0412.2010.01030.x. [Epub].
23. Leal MD, Esteves-Pereira AP, Nakamura-Pereira M, Torres JA, Theme-Filha M, Domingues RM, *et al.* Prevalence and risk factors related to preterm birth in Brazil. Reprod Health 2016;13:127.
24. Okeke TC, Enwereji JO, Okoro OS, Adiri CO, Ezugwu EC, Agu PU. The incidence and management outcome of preterm premature rupture of membranes in a tertiary hospital in Nigeria. Am J Clin Med Res 2014;2:14-7.
25. "Stillbirths". World Health Organization. Available from: http://www.who.int/maternal_child_adolescent/epidemiology/stillbirth/en/. [Last retrieved on 2016 Sep 29].
26. World Health Organization. Recommendations for the Prevention and Treatment of Postpartum Hemorrhage. 1.Postpartum Hemorrhage – Prevention and Control. 2.Postpartum Hemorrhage – Therapy. 3.Obstetric labor complications. 4.Guideline. (NLM classification: WQ 330) World Health Organization. World Health Organization; 2012.
27. Deshmukh JS, Motghare DD, Zodpey SP, Wadhva SK. Low birth weight and associated maternal factors in an urban area. Indian Pediatr 1998;35:33-6.
28. Onyiriuka AN. High birth weight babies: Incidence and foetal outcome in a mission hospital in Benin city, Nigeria. Niger J Clin Pract 2006;9:114-9.
29. Carvalho GA, Perez CL, Ward LS. The clinical use of thyroid function tests. Arq Bras Endocrinol Metabol 2013;57:193-204.
30. Azizi F, Mehran L, Amouzegar A, Delshad H, Tohidi M, Askari S, *et al.* Establishment of the trimester-specific reference range for free thyroxine index. Thyroid 2013;23:354-9.
31. Mansourian AR, Mansourian AA, Saifi A, Marjani A, Veghari GR, Ghaemi E *et al*. Maternal thyroid stimulating hormone levels during the first trimester of pregnancy at the South-East if the Caspian Sea in Iran. J Clin Diagn Res 2010;4:2472-7.
32. Zarghami N, Rohbani-Noubar M, Khosrowbeygi A. Thyroid hormones status during pregnancy in normal Iranian women. Indian J Clin Biochem 2005;20:182-5.
33. Medici M, Korevaar TI, Visser WE, Visser TJ, Peeters RP. Thyroid function in pregnancy: What is normal? Clin Chem 2015;61:704-13.
34. Negro R, Formoso G, Mangieri T, Pezzarossa A, Dazzi D, Hassan H, *et al.* Levothyroxine treatment in euthyroid pregnant women with autoimmune thyroid disease: Effects on obstetrical complications. J Clin Endocrinol Metab 2006;91:2587-91.
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, Chen X, Bogden JD, Davies TF, Scholl TO. The thyroid and pregnancy: A novel risk factor for very preterm delivery. Thyroid 2005;15:351-7.
37. Saki F, Dabbaghmanesh MH, Ghaemi SZ, Forouhari S, Ranjbar Omrani G, Bakhshayeshkaram M, *et al.* Thyroid function in pregnancy and its influences on maternal and fetal outcomes. Int J Endocrinol Metab 2014;12:e19378.