International Journal of Clinical Obstetrics and Gynaecology

ISSN (P): 2522-6614
ISSN (E): 2522-6622

www.gynaecologyjournal.com
2021; 5(2): 80-84
Received: 11-02-2021
Accepted: 14-03-2021

Dr. Sura Munem Naeem
M.B.CH. B, Resident Doctor at Al-Affiliation: Elwiyah Maternity Teaching Hospital, Baghdad, Iraq. Iraqi Council of Medical Health Specialization

Dr. Ulfat MA J Al-Nakkash
M.B.Ch.B, C.A.B.O.G, Consultant at Al-Elwiyah Maternity Teaching Hospital, Baghdad, Iraq
Affiliation: Iraqi Council of Medical Health Specialization

Corresponding Author:
Dr. Sura Munem Naeem
M.B.CH. B, Resident Doctor at Al-Elwiyah Maternity Teaching Hospital, Baghdad, Iraq
Affiliation: Iraqi Council of Medical Health Specialization

The association of Maternal Alpha fetoprotien and BHCG with umbilical artery pattern in mid trimester

Dr. Sura Munem Naeem and Dr. Ulfat MA J Al-Nakkash

DOI: https://doi.org/10.33545/gynae.2021.v5.i3b.906

Abstract
Background: Mid-trimester maternal serum markers have been used for prenatal aneuploidy screening for more than 20 years. In the absence of aneuploidy or neural tube defect, these serum markers have also been associated with several placenta-mediated adverse pregnancy outcomes, including preeclampsia, intrauterine growth restriction, and stillbirth.
Objective: To assess the effect of maternal alpha fetoprotein and B-HCG in umbilical artery Doppler change in 2nd trimester.
Patients and Method: An observational prospective cohort study, conducted at Al-Elwiya maternity teaching hospital Obstetrics department, from the first of February 2019 to the end of Jan. 2020.
Results: The current study was included 50 patients within the mean age (25.7±6.2) years old and the main age group was between 21-30 years (62.0%), normal pregnancy outcome in (88.0%), while preeclampsia found in 4 (8.0%) patients and IUGR in 2 (4,0%). Significant association were found between elevated level of serum HCG and Alpha feto protein with adverse pregnancy outcome (P=0.005).
Conclusion: Alpha feto protein and HCG were not significantly related to Doppler change in the current study.

Keywords: Maternal Alpha fetoprotein, HCG, umbilical artery, mid trimester

Introduction
Mid-trimester maternal serum markers have been used for prenatal aneuploidy screening for more than 20 years [1] In the absence of aneuploidy or neural tube defect, these serum markers have also been associated with several placenta-mediated adverse pregnancy outcomes, including preeclampsia, intrauterine growth restriction, and stillbirth. The Society of Obstetricians and Gynecologists of Canada suggests that an unexplained elevation of maternal serum alpha-fetoprotein (> 2.5 MoM) and/or human chorionic gonadotropin (> 3.0 MoM) is associated with an increased frequency of PMAPOs. On the other hand, recent reviews and meta-analyses found that neither of these two markers used in isolation is a good predictor of adverse pregnancy outcomes [2].
A growing body of evidence suggests that deep placentation disorders, which have been typically associated with preeclampsia and are mainly found in the preterm forms of the disease, are also found in all "great obstetrical syndromes" including all placenta-mediated adverse pregnancy outcomes (PMAPOs), spontaneous preterm labour and preterm premature rupture of membranes [3] These findings are also supported by ultrasound studies showing that abnormal first trimester and mid-trimester uterine artery Doppler velocimetry is associated more with the preterm forms of preeclampsia than the term forms, and is associated more with preterm stillbirths than term [4]. There is an increased interest in the early prediction of preterm PMAPO because of the recent evidence showing that most could potentially be prevented with the use of low-dose aspirin initiated at or before 16 weeks of gestation [5].

Umbilical artery
Doppler sonography is used for non-invasive assessment of circulation in many clinical conditions. This technique has been used for studying most of the major fetal circulatory systems, including the umbilical artery (UA), umbilical vein, aorta, heart, and middle cerebral artery. Doppler sonography provides a unique opportunity to investigate human fetal Hemodynamics and to use these findings for fetal surveillance.

In pregnancies complicated by placental insufficiency, meta-analyses of randomized trials have shown that fetal umbilical artery Doppler is an effective test for improving perinatal mortality and morbidity [6].

The umbilical artery (UA) is the major vascular pathway connecting the fetus and placenta. The fetus obtains nutrients and oxygen through the umbilical circulation. The systolic/diastolic ratio (S/D), pulsatility index (PI), and resistance index (RI) are the hemodynamic indices of the fetoplacental circulation [7].

Alpha-fetoprotien

Alpha-fetoprotein (AFP) was first described in 1963 by Abelev *et al*. and is normally produced in the fetal liver and yolk sac. AFP in human fetal blood rises rapidly from the end of the first trimester and begin to fall after 32 weeks of gestation. After birth, serum AFP level continues to fall with a half-life of 3-4 days, and by the second year of life to the normal range [8, 9].

Maternal Serum AFP

AFP is a glycoprotein and it is a member of the albuminoid gene family [AFP, albumin (ALB), alpha albumin (αALB), vitamin D binding protein (DBP)]. In adults AFP expression is often associated with hepatocellular cancer, non-germinomatous germ cell tumors and gastrointestinal cancers. However, AFP can be found in non-neoplastic conditions such as hepatitis, cirrhosis and pregnancy. During pregnancy it is synthesized by the fetus. AFP is normally produced in early pregnancy primarily by the fetal liver and yolk sac. It is also produced to a lesser extent by the fetal gastrointestinal tract. As the yolk sac involutes at the 9th week of gestation, the fetal liver is the principal source of AFP during development. AFP synthesis by the proliferating fetal liver actually increases through the 20th week of gestation, after which it remains fairly constant until 32nd week. AFP is excreted into fetal urine and transported to maternal serum through the placenta or by diffusion across fetal membranes [9].

The transplacental passage of AFP involves additional and more complicated mechanisms. It is asymmetrical and unidirectional, displaying a faster transfer rate of AFP from the fetal to maternal circulation. A small amount of AFP is transported and can be measured in the maternal serum. Despite the decrease in fetal serum AFP (fs-AFP) during mid-trimester of gestation, ms-AFP continues to rise until the 32nd week of gestation. After the 32nd week of gestation, ms-AFP begins to decline until parturition. AFP is involved with ontogenic and oncogenic growth. It can bind and transport a multitude of ligands (bilirubin, fatty acids, retinoids, steroids, heavy metals, dyes, flavonoids, phytoestrogens, dioxin and various drugs). Also, it is capable of regulating growth in reproductive, hematopoietic, placental, hepatic, inflammatory and lymphatic cells. However, AFP regulating function during pregnancy remains controversial and relatively unknown. AFP involvement in the regulation of placental growth is also unknown [10].

Maternal Serum hCG

hCG is a member of the glycoprotein hormone family [hCG, Follicle Stimulating Hormone (FSH), Luteinizing Hormone (LH), Thyroid Stimulating Hormone (TSH)]. All of them are dimers consisting of a common α-subunit and distinct β-subunits that are associated non-covalently. The distinct β-subunits confer biological activity and display various degrees of homology. In adults hCG expression is often associated with pregnancy. However, hCG can be found in other conditions such as gestational trophoblastic disease and non-germinomatous germ cell tumors. During pregnancy hCG is produced almost exclusively by the syncytiotrophoblast of the placenta. However, it is synthesized by the fetal kidney and fetal liver. Most of the hCG in circulation is metabolized by the liver. Also, 20% of the circulating hCG is excreted by the kidneys. However, hCG appears early during pregnancy. Its concentration increases gradually by reaching a peak at 8 to 10 weeks of gestation. After that peak, it progressively declines to reach a plateau at 18 to 20 weeks of gestation. During early pregnancy, the main function of hCG is the maintenance of the corpus luteum. It takes over from LH in promoting progesterone production by ovarian corpus luteal cells, preventing menstrual bleeding. It promotes progesterone production only for 3-4 weeks following pregnancy implantation. Also, hCG receptor gene expressed by uterine spiral arteries and hCG acts on them promoting angiogenesis of uterine vasculature and uterine growth in line with fetal growth. During pregnancy hCG also promotes: cyto-trophoblast differentiation, immunosuppression and blockage of phagocytosis of invading trophoblast cells [10].

Aim of the study

To assess the effect of maternal alpha fetoprotein and B-HCG in umbilical artery Doppler change in 2nd trimester.

Patients and method: Study design and setting

Observational prospective, conducted at Al-Elwiya maternity teaching hospital at the Obstetrics department, from the first of February 2019 to the end of Jan. 2020.

Ethical consideration: Verbal consent was taken with reassurance that interpret gained will be kept confidentially and not to be used for other research object. By no mean, the research data interfere with patient's management and all patients received their management irrespective to the study approved by each of the local thesis committee Iraqi ministry of health, council of Iraqi board of health specialization and Al-Elwia Maternity teaching hospital.

Patients

50 pregnant women in 2nd trimester were enrolled in the current study.

Inclusion criteria

1. Singleton pregnancy
2. 2nd trimester pregnancy

Exclusion criteria:

1. Twin pregnancy
2. Congenital anomalies
3. Previous history of preterm labor
4. Previous history of IUGR
5. Maternal disease, hysterectomy, DM, HT, and other

A specially designed questionnaire form was used to collect information about (age, BMI, GA, Obstetrical history, smoking history and Birth weight of the neonate in the last pregnancy).

Method

5 cc of maternal venous blood was collected from all patients to measure serum level of both AFP and HCG. Serum samples were kept frozen at -40°C until assayed. The AFP level was determined by an enzyme immunoassay with a fluorometric end-point. The levels of hCG were assay using monoclonal antibodies, Doppler velocimetry of the umbilical artery was

performed to all patients by a specialist. Measurements were performed using a continuous-wave Doppler with a 4-MHz probe. The S/D ratio, reflecting resistance to flow distal to the point of measurement, was calculated. An abnormal value was considered as two standard deviations from the mean value for the gestational age. In order to incorporate all the velocimetry determinations performed along the pregnancy for each patient, a scoring system was devised. The 'Velocimetry Score' (VS) was defined as follows:

$$VS = \frac{\text{Number of tests with normal values}}{\text{Total number of tests}} \times 100$$

Statistical analysis

After the data were entered in a table developed by the researchers, the analysis was done by using the SPSS program, version 23 and for qualitative variables, we used frequencies and percentages, and for the quantitative variables, we used measures of central tendency and dispersion (standard deviation). For the inferential statistics, the tests were used of chi-square test (with a significance of P ≤0.05). Validity test were done to measure the (sensitivity, specificity, NPV, PPV and accuracy of the test.

Results

The current study were included 50 patients within the mean age (25.7±6.2) years old and For the pregnancy outcome it was found normal in the majority of the patients (88.0%), while preeclampsia found in 4 (8.0%) patients and IUGR in 2 (4,0%) (figure 1).

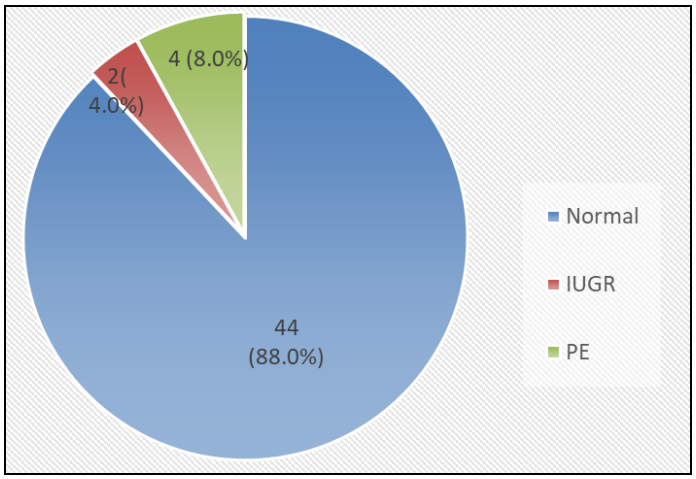

Fig 1: Distribution of the studied group according to pregnancy outcome

Table 1 show that there is highly significant association were found between S. Alpha feto protein and HCG with parity (p<0.001), but no association were found regarding the umbilical artery Doppler change and parity (p>0.05)

Table 1: Relation between different parameters and parity

	Parity			P value
	Nulli para	1-3	4-6	
	Mean±SD	Mean±SD	Mean±SD	
S. Alpha feto protein	107±31	154±44	166±56	<0.001
S. HCG	23047±2106	27283±7105	16695±4407	<0.001
PSV (cm/s)	44.47±8.18	38.19±10.17	38.93±8.83	0.784
SDV (cm/s)	14.74±7.12	16.87±7.79	11.72±4.48	0.77
S/D ratio	4.56±1.43	2.64±0.88	3.79±0.71	0.88
RI (Resistance index)	0.736±0.181	0.577±0.13	0.7±0.074	0.842
PI (Pulsatility index)	1.207±0.487	0.932±0.3	1.243±0.195	0.721

Significant association were found between S. alpha feto protein, S/D ratio, PI and gestational age (p<0.05) (table 2)

Table 2: Relation between different parameters and gestational age

	Gestational age (w)		P value
	≤22	>22	
	Mean±SD	Mean±SD	
S. Alpha feto protein	132±47	180±38	0.013
S. HCG	21444±3699	26655±11139	0.234
PSV (cm/s)	39.46±7.17	40.75±12.51	0.685
SDV (cm/s)	14.85±6.71	14.21±5.54	0.738
S/D ratio	4.04±2.81	2.99±0.55	0.01
RI (Resistance index)	0.693±0.174	0.663±0.079	0.2
PI (Pulsatility index)	1.126±0.442	0.978±0.19	0.03

As show in table 3 there is no significant association were found between S. Alpha feto protein and umbilical artery Doppler change (p>0.05)

Table 3: Association between S. Alpha feto protein and umbilical artery Doppler change.

	S. Alpha feto protein		P value
	<100	>100	
	Mean±SD	Mean±SD	
PSV (cm/s)	39.87±6.8	39.9±6.79	0.97 NS
SDV (cm/s)	14.8±6.5	14.5±6.09	0.7 NS
S/D ratio	3.9±1.02	3.8±1.11	0.5 NS
RI (Resistance index)	0.672±0.71	0.613±0.03	0.4 NS
PI (Pulsatility index)	1.02±0.41	1.01±0.12	0.8 NS

Significant association were found between elevated level of serum Alpha feto protein and adverse pregnancy outcome (P=0.005), while no association regarding HCG (table 4)

Table 4: Association between S. Alpha feto protein and HCG with adverse pregnancy outcomes

Parameters	Mean±SD			P value
	Normal	PE	IUGR	
S. Alpha feto protein	147±49	220±34	250±57	0.005
S. HCG	22345±7075	23223±1422	24429±5632	0.5

The acceptable cut off points and the corresponding validity tests values S. Alpha feto protein in prediction of IUGR from healthy pregnant women were shown in table 5.

The cutoff S. Alpha feto protein level of >199.9 had acceptable validity results (38.0% sensitivity, 93.0% specificity, 48.0%PPV, 90.0% NPV and accuracy 70.0%). While cutoff for S. HCG >20128 was (30.0% sensitivity, 90.0% specificity, 37.0% PPV, 87.0% NPV and accuracy 73.0%).

Table 5: Coordinates of the ROC Curve of S. Alpha feto protein and HCG in IUGR

Cutoff point	Sensitivity	Specificity	PPV	NPV	Accuracy
Alpha feto protein > 199.9	38.0%	93.0%	48.0%	90.0%	70.0%
HCG >20128	30.0%	90.0%	37.0%	87.0%	73.0%

The acceptable cut off points and the corresponding validity tests values S. Alpha feto protein in prediction of PE from healthy pregnant women were shown in table [6], cutoff S. Alpha feto protein level of >218 had acceptable validity results (48.0% sensitivity, 90.0% specificity, 55.0% PPV, 78.0% NPV and accuracy 70.0%). While cutoff for S. HCG >21655 was not good (25.0% sensitivity, 45.0% specificity, 46.0%PPV, 37.0% NPV and accuracy 40.0%)

Table 6: Coordinates of the ROC Curve of S. Alpha feto protein and HCG in PE

Cutoff point	Sensitivity	Specificity	PPV	NPV	Accuracy
Alpha feto protein > 218	48%	90%	55%	78%	70%
HCG > 21655	25%	45%	46%	37%	40%

No association were found between doppler change and level of S.HCG (P>0.05) (table 7)

Table 7: Association between S.Alpha feto protein and umbilical artery

Parameters	S.HCG (mIU/ml)		P Value
	≤ 20128	>20128	
	Mean±SD	Mean±SD	
PSV (cm/s)	39.87±0.3	39.9±5.9	0.95 Ns
SDV (cm/s)	14.8±4.3	14.6±6.12	0.7 Ns
S/D ratio	3.8±1.09	3.9±1.0	0.49 Ns
RI (resistance index)	0.68±0.77	0.64±0.8	0.7 Ns
PI (pulsatility index)	1.03±0.01	1.03±0.05	1 Ns

Discussion

Alpha-fetoprotein (AFP) is a plasma protein produced by the embryonic yolk sac and the fetal liver. AFP levels in serum, amniotic fluid, and urine functions as a screening test for congenital disabilities, chromosomal abnormalities, as well as some other adult occurring tumors and pathologies. This tumor marker is a glycoprotein encoded by the *AFP* gene on chromosome 4q25. Prenatal levels in developing human embryo rise from the end of the first trimester and begin to fall after 32 weeks of gestation. Maternal serum AFP forms part of the triple or quadruple screening tests for fetal anomaly [11, 12].

The current study revealed that there is adverse pregnancy outcome in which, preeclampsia found in (8%) and IUGR in (4%) with significant increase in serum level of Alpha feto protein in adverse outcome. This is in agreement with that mentioned by Misfud W [32] study when they believed that early placental dysfunction, which influences maternal serum AFP and hCG, can lead to placental hypo-perfusion and a maternal endothelial reaction that can result in fetal growth restriction, preeclampsia, and even fetal death. Ogge G *et al*, study suggested that the term and preterm forms of preeclampsia are different with regard to maternal characteristics and placental pathology [3].

Previous studies were describe the association between elevated maternal serum AFP levels in pregnancy (especially the 2nd trimester) and adverse pregnancy outcome [13]. As in a study carried by van Rijn M *et al*, reported that the Elevated maternal serum AFP levels are associated with placenta previa [14].

while in a study carried by Hung T *et al* and Zelop C *et al*, found that the un explained increased level of maternal serum AFP in both second and third trimester placenta previa are related to increased risk of abnormal adherence of placenta including precreta, accrete and increta [13, 14].

Other studies found a relation between elevated serum levels of AFP with many other complications of pregnancy outcomes like (gestational hypertension, pre-eclampsia, early fetal loss, late fetal loss, preterm delivery, IUGR, and placental abruption). [15-18]

Maternal serum AFP or hCG > 2.0 MoM increases the risk of preterm placenta-mediated adverse pregnancy outcomes but not term placenta-mediated adverse pregnancy outcomes in their population. They suggested that women with elevated serum AFP or hCG should receive standard pregnancy care once they have reached 37 weeks of gestation if fetal growth is in the normal range [19].

For HCG in the current study, we found that there is elevated serum level of HCG but with no significant association were found with adverse pregnancy outcome. Which is not in agreement with previous studies that found increased levels of second-trimester hCG have been related with many adverse pregnancy outcome which is likely attributed to placental dysfunction [17, 18, 20] This may be due to the increase in these markers not reach more than 2 MOM.

As previously observed with some other markers of Down syndrome, it seems that the risk of adverse effects increases with increasing HCG levels. Towner *et al*. [21] examined 344 women with high HCG levels and found no increased risk of adverse obstetric outcomes in women with HCG levels greater than 2.0 MoM. There was an increased risk of premature pre-eclampsia in women with hCG levels greater than 3.0 MoM and greater risk of preterm labor due to fetal indications in subset of women with hCG levels above 4.0 MoM.

While in Dugoff L *et al* study, revealed that there is no significant association were found between serum level of HCG (greater than 2.0 MoM) and adverse pregnancy outcome [22].

Moreover, in a study carried by Spencer K, mentioned that 28 isolated increased free serum-hCG levels (more than 2.0 MoM) were not related with an increased risk of fetal death at or after 24 weeks of gestation, low birth weight, IUGR, and preterm labor [19].

The current study revealed that in spite of little increment in uterine artery indices (PI and RI), there is no significant

association were found between S. Alpha feto protein and umbilical artery Doppler change (p>0.05). Which is not in agreement with that found by El-Aal NK *et al* when significant differences were found [23].

Limitation of the study

This is the first study carried in Iraq in this subject so there is no studies to compare with.

Conclusion

Alpha feto protein and HCG were not significantly related to Doppler change in the current study.

No conflicts of interest

Source of funding: self

Ethical clearance: was taken from the scientific committee of the Iraqi Ministry of health

References

1. Committee; CCMG Committee on Prenatal Diagnosis; SOGC Diagnostic Imaging Committee. Prenatal screening for fetal aneuploidy. SOGC Clinical Practice Guideline no. 262, July 2011. J Obstet Gynaecol Can 2011;33(7):736-50.
2. Hui D, Okun N, Murphy K, Kingdom J, Uleryk E, Shah PS. Combinations of maternal serum markers to predict preeclampsia, small for gestational age, and stillbirth: a systematic review. J Obstet Gynaecol Can 2012;34(2):142-53.
3. Ogge G, Chaiworapongsa T, Romero R, Hussein Y, Kusanovic JP, Yeo L, *et al*. Placental lesions associated with maternal underperfusion are more frequent in early-onset than in late-onset preeclampsia. J Perinat Med 2011;39(6):641-52.
4. Demers S, Bujold E, Arenas E, Castro A, Nicolaides KH. Prediction of recurrent preeclampsia using first-trimester uterine artery Doppler. Am J Perinatol 2014;31(2):99-104
5. Roberge S, Nicolaides KH, Demers S, Villa P, Bujold E. Prevention of perinatal death and adverse perinatal outcome using low-dose aspirin: a meta-analysis. Ultrasound Obstet Gynecol 2013;41(5):491-9.
6. Mone F, McAuliffe FM, Ong S. The clinical application of Doppler ultrasound in obstetrics. The Obstetrician & Gynaecologist 2015;17:13-19.
7. Fu J, Olofsson P. Relations between fetal brain-sparing circulation, oxytocin challenge test, mode of delivery and fetal outcome in growth-restricted term fetuses. Acta Obstet Gynecol Scand 2011;90:227-230.
8. Anzai H, Kazama S, Kiyomatsu T, Nishikawa T, Tanaka T, Tanaka J *et al*. Alpha-fetoprotein-producing early rectal carcinoma: a rare case report and review. World J Surg Oncol 2015;13:180.
9. Lin HH, Chang CC, Yang SH, Chang SC, Chen WS, Liang WY *et al*. Elevated serum alpha-fetoprotein in poorly differentiated adenocarcinoma with neuroendocrine differentiation of the ascending colon: a case report. World J Surg Oncol 2016;14:84.
10. Androutsopoulos G, Gkogkos P, Decavalas G. Mid-trimester maternal serum HCG and alpha fetal protein levels: clinical significance and prediction of adverse pregnancy outcome. International journal of endocrinology and metabolism. 2013;11(2):102.
11. Sharony R, Dayan D, Kidron D, Manor M, Berkovitz A, Biron-Shental T *et al*. Is the ratio of maternal serum to amniotic fluid AFP superior to serum levels as a predictor of pregnancy complications? Arch. Gynecol. Obstet 2016;293(4):767-70.
12. Öztürk H, Erkaya S, Altınbaş S, Karadağ B, Vanlı Tonyalı N, Özkan D. The role of unexplained high serum alpha-fetoprotein (AFP) and human chorionic gonadotropin (hCG) levels in the second trimester to determine poor obstetric outcomes. Turk J Obstet Gynecol 2014;11(3):142-147
13. Mifsud W, Sebire NJ. Placental pathology in early-onset and late-onset fetal growth restriction. Fetal Diagn Ther 2014;36(2):117-28.
14. Hung TH, Shau WY, Hsieh CC, Chiu TH, Hsu JJ, Hsieh TT. Risk factors for placenta accreta. Obstet Gynecol 1999;93:545-50.
15. van Rijn M, van Der Schouw YT, Hagenaars AM, Visser GH, Christiaens GC. Adverse obstetric outcome in low- and high- risk pregnancies: predictive value of maternal serum screening. Obstet Gynecol 1999;94:929-34.
16. Zelop C, Nadel A, Frigoletto FD Jr, Pauker S, MacMillan M, Benacerraf BR. Placenta accreta/percreta/increta: a cause of elevated maternal serum alpha-fetoprotein. Obstet Gynecol 1992;80:693-4.
17. Krause TG, Christens P, Wohlfahrt J, Lei U, Westergaard T, Norgaard-Pedersen B *et al*. Second-trimester maternal serum alpha-fetoprotein and risk of adverse pregnancy outcome. Obstet Gynecol 2001;97:277-82.
18. Chandra S, Scott H, Dodds L, Watts C, Blight C, Van Den Hof M. Unexplained elevated maternal serum alpha-fetoprotein and/or human chorionic gonadotropin and the risk of adverse outcomes. Am J Obstet Gynecol 2003;189:775-81.
19. Spencer K. Second-trimester prenatal screening for Down syndrome and the relationship of maternal serum biochemical markers to pregnancy complications with adverse outcome. Prenat Diagn 2000;20:652-6.
20. Tancrède S, Bujold E, Giguère Y, Renald MH, Girouard J, Forest JC. Mid-trimester maternal serum AFP and hCG as markers of preterm and term adverse pregnancy outcomes. Journal of Obstetrics and Gynaecology Canada. 2015;37(2):111-6.
21. Towner D, Gandhi S, El Kady D. Obstetric outcomes in women with elevated maternal serum human chorionic gonad- otropin. Am J Obstet Gynecol 2006;194:1676-82.
22. Dugoff L, Hobbins JC, Malone FD, Vidaver J, Sullivan L, Canick JA *et al*. Quad screen as a predictor of adverse pregnancy outcome. Obstet Gynecol 2005;106:260-7.
23. El-Aal NK, El-Halaby AE, Taie AH. Combined second trimester maternal serum α-fetoprotein and uterine artery Doppler in the prediction of pre-eclampsia. Menoufia Medical Journal 2019;32(2):430.