Placental Growth Factor in 24 to 30 Weeks of gestation as a predictor of Pre Eclampsia

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Abstract:
Background: *Pre-eclampsia is one of the leading causes of maternal mortality and is lest understood in aspects of physiology; it awaits sensitive and specific tests for its prediction.*
Objectives: *1.Placental growth factor is one test that is evolving as a predictor of preeclampsia. Present study aims to evaluate this aspect.*
2. To evaluate the value of placental growth factor in predicting the severity of pre eclampsia.
3. To determine the association of placental growth factor with variables such as blood pressure and BMI.
Materials and methods: *This prospective study was conducted among 80 pregnant mothers between 24 and 30 weeks of gestation to evaluate Placental Growth Factor in predicting the occurrence of preeclampsia. Fasting blood samples were collected from randomly selected pregnant mothers between 24 and 30 weeks and they were followed up for occurrence of preeclampsia and gestational age at which termination of pregnancy was done.*
Results: *Lower level of PlGF was found to be associated with pre eclampsia when compared to those who did not develop pre eclampsia.*
Conclusion: *PlGF as a predictor of pre eclampsia is more promising; the cut of level of PlGF in our population has to be established by more studies.*
Key words: *Placental Growth Factor, Predictor of Pre eclampsia.*

I. Introduction

Preeclampsia is a multi-system pregnancy-specific disease which affects 2 to 8% of all pregnancies.[1]It is characterized by placental and maternal vascular dysfunction. It is one of the leading cause of both maternal and fetal morbidity and as well as their mortality in the developing world.[2] There is need to balance the risks between maternal and perinatal outcomes, as expectant management improves neonatal outcome, but at the same time worsens maternal conditions.[3,4,5.] Clinical diagnosis of preeclampsia in late stages gives very little scope for expectant management.[6] Hence a pre-clinical diagnostic test which is able to predict pre eclampsia and its severity is needed. Evaluation of bio-markers and imaging techniques has shown that none have adequate sensitivity and specificity or the convenience for prediction or diagnosis of pre eclampsia but they help only in diagnosis of advanced disease with established end organ damages.

In the development of placenta, there is invasion of the cytotrophoblasts into the maternal spiral arterioles and the small caliber resistance vessels are transformed to high caliber conduit vessels. This is one of the early steps in placental development which starts from first trimester (10-12 weeks) and continues till 18 to 20 weeks of gestation. The two main components in the pathogenesis of pre eclampsia are the abnormal placentation and endothelial dysfunction. Abnormal placentation is often quoted as cause of pre eclampsia rather than the fetus. Symptoms of pre eclampsia persisted until the delivery of the placenta even in rare cases of extra uterine pregnancies. This brings to light the fact that the presence of fetus is not necessary for the development of pre eclampsia.[7]

The process of normal placental invasion by the cytotrophoblasts fails in pre eclampsia, leading to a shallow invasion of the spiral arterioles keeping them as small caliber resistance vessel leading to defective uteroplacental circulation and worsening placental perfusion.[7,8] Placental growth factor (PlGF) belongs to the VEGF (vascular endothelial growth factor) sub-family and it has a key role in angiogenesis and vasculogenesis. It is abundantly seen in placental trophoblast and also in villous trophoblast, in addition to other tissues. In normal pregnancy the expression of PlGF protein and its mRNA are markedly increased during the second trimester. There is detachment of these syncytial knots from the syncytiotrophoblast, which form metabolically active multinucleated aggregates loaded with sFlt1 protein and mRNA which are accelerated in preeclampsia contributing to the maternal vascular injury. PlGF synthesized by syncytiotropoblast increases

with gestataional age reaching peak by 26 to 30 weeks of gestation and declines towards term.[9] PlGF is very much lowered in pre eclampsia and it has been proved that PlGF values are markedly lowered 5 weeks before the onset of pre eclampsia which assures us of its predictive capabilities.[7]

Levels of sFlt-1 are higher and PlGF lower in preeclamptic women, suggesting that it is mainly sourced from the placenta.[10] PlGF to sFlt-1 ratio has been proposed to be one among the best methods for predicting pre-eclampsia before the onset of the disease.[11,12,13] In pre eclamptic women, the PlGF levels were significantly lower not only in the plasma, but also in the urine.[14]

Plasma PlGF in early second trimester could be an effective predictor of early onset preeclampsia in women who are overweight/obese pregnant than in wom   men with normal BMI or those who are over weight.[15] Women who had pre eclampsia had significantly lower levels of PlGF where mean was 90 pg/ml in the pre eclamptic group as to 142 pg/ml in the control group. The maximum difference in the values of PlGF was noted in the weeks preceding pre eclampsia along with an increase in the sFlt-1 levels. The study conducted by Levine et al concluded that increased levels of sFlt-1 and reduced levels of PlGF is useful as a predictor for the subsequent development of preeclampsia.[8]

II.   Aim Of The Study

The aims of study is to determine the diagnostic value of placental growth factor for predicting the occurrence of pre eclampsia in the gestational period from 24 to 30 weeks. To compare the value of placental growth factor in predicting mild or severe pre eclampsia. To determine the association of placental growth factor with variables such as blood pressure, BMI and fetal outcome.

III.   Materials And Methodology

A prospective study was done in 80 randomly selected women who attended the casualty and antenatal OPD at the Institute of Obstetrics Gynecology, Chennai during the period from August 2014 to June 2015. The study protocol was approved by the Institutional ethical committee.

The inclusion criteria were:
(1) Pregnant mothers between 24 and 30 weeks of gestation
(2) Randomly selected population
(3) Antenatal mothers who were willing to deliver in our institution.

The exclusion criteria were:
(1) Diagnosed preeclampsia
(2) Chronic hypertension
(3) Multiple Gestation
(4) Patient not willing to participate in the study

Informed and written consent for the study was obtained from all patients. History was taken in detail and patients were subjected to clinical examination. Venous blood samples of 2.5ml with EDTA were collected from randomly selected pregnant mothers between 24 and 30 weeks to determine PlGF values by fluorescent immunoassay method. PlGF values are expressed in pg/ml. The collected data were analyzed using appropriate statistical methods. All antenatal mothers who were included in the study were followed up through admission, delivery and the immediate post-partum period.

Figure-I
The age distributions of the patients are represented in the below chart.
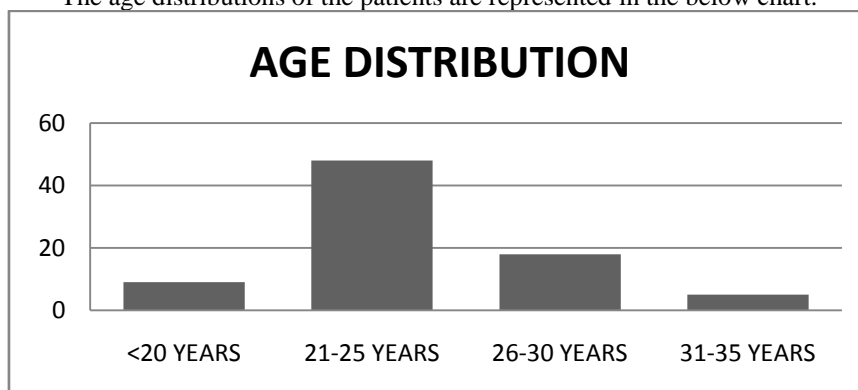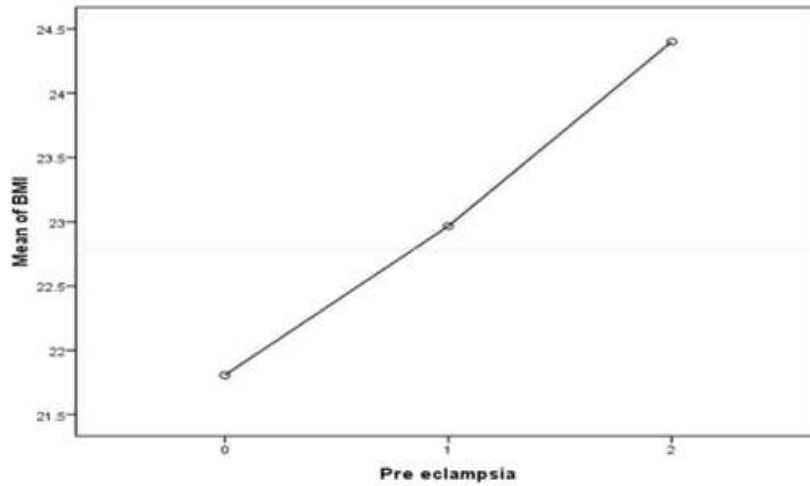
Figure-II
Mean BMI And Occurrence Of Pre Eclampsia

The mean BMI was 22.97 with a standard deviation of 2.004 among those who developed mild pre eclampsia and 24.40 with a standard deviation of 4.391 among those who developed severe pre eclampsia.

FIGURE-III
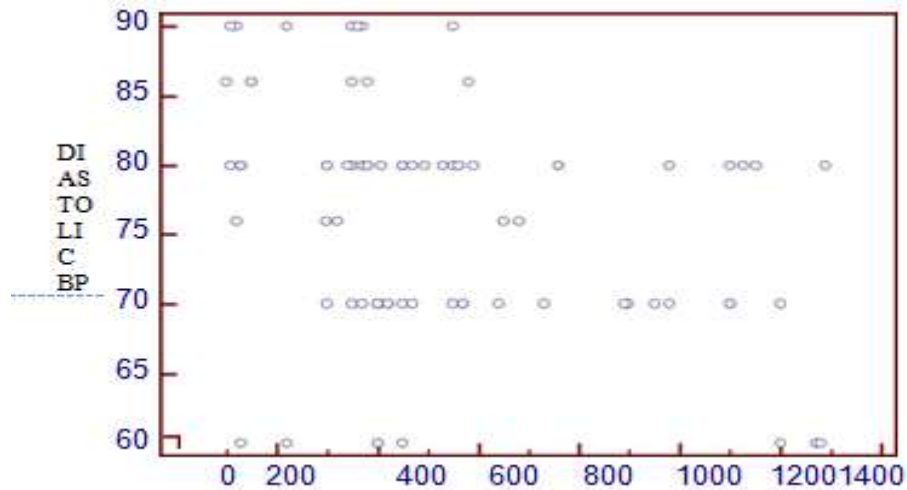**Scatter diagram showing correlation between Diastolic BP and PlGF values with a significance level of P=0.0032**

TABLE-I   **Correlation**

Sample size	80
Correlation coefficient r	-0.3258
Significance level	P=0.0032
95% Confidence interval for r	-0.5091 to -0.1143

	Variable Y		Diastolic_BP
			Diastolic BP
	Variable X		PLGF_Values
			PLGF Values

FIGURE-IV
Scatter diagram showing correlation between Systolic BP and
PlGF values with a significance level of P=0.0013

TABLE-II **Correlation**
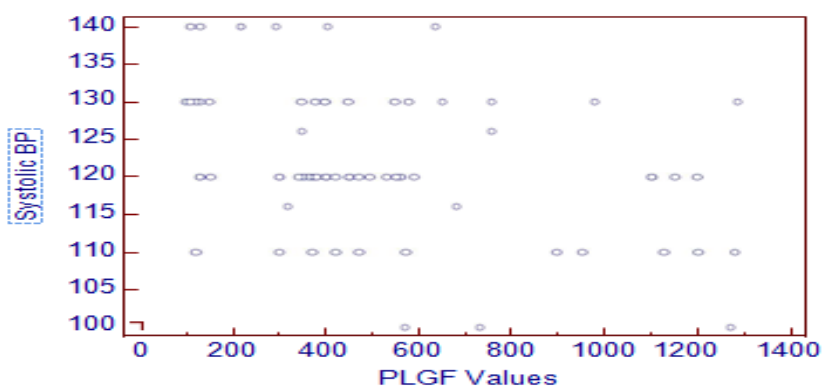
	Variable Y		Systolic_BP
			Systolic BP
	Variable X		PLGF_Values
			PLGF Values

Sample size		80
Correlation coefficient r		-0.3541
Significance level		P=0.0013
95% Confidence interval for r		-0.5324 to -0.1457

Area under the ROC curve (AUC)

| Area under the ROC curve (AUC)		0.915254
Standard Error[a]		0.0370
95% Confidence interval[b]		0.831461 to 0.965866
z statistic		11.234
Significance level P (Area=0.5)		<0.0001
[a] DeLong et al., 1988		
[b] Binomial exact		
Youden index		
Youden index J	0.7078	
Associated criterion	≤350	

21 patients out of 80 samples had developed pre eclampsia (26.5%).6 out of the 21 patients developed severe pre eclampsia.14 out of 80 patients had previous history of pre eclampsia.22 out of 80 patients had proteinuria. All 21 patients who developed pre eclampsia had proteinuria.1 patient had isolated proteinuria without pre eclampsia

TABLE- III   Criterion values and coordinates of the ROC curve

Criterion	Sensitivity	95% CI		Specificity	95% CI		+LR	-LR
<100	0.00	0.0	- 16.1	100.00	93.9 - 100.0			1.00
≤298	66.67	43.0 - 85.4		100.00	93.9 - 100.0			0.33
≤320	66.67	43.0 - 85.4		93.22	83.5	- 98.1	9.83	0.36
≤340	71.43	47.8 - 88.7		93.22	83.5	- 98.1	10.54	0.31
≤350	80.95	58.1 - 94.6		89.83	79.2	- 96.2	7.96	0.21
≤380	80.95	58.1 - 94.6		77.97	65.3	- 87.7	3.67	0.24
≤400	85.71	63.7 - 97.0		67.80	54.4	- 79.4	2.66	0.21
≤407	85.71	63.7 - 97.0		66.10	52.6	- 77.9	2.53	0.22
≤420	90.48	69.6 - 98.8		64.41	50.9	- 76.4	2.54	0.15
≤450	95.24	76.2 - 99.9		55.93	42.4	- 68.8	2.16	0.085
≤530	95.24	76.2 - 99.9		49.15	35.9	- 62.5	1.87	0.097
≤550	100.00	83.9	- 100.0	45.76	32.7	- 59.2	1.84	0.00
≤1288	100.00	83.9	- 100.0	0.00	0.0	- 6.1	1.00	

The sensitivity and specificity are 80.95% and 89.83% respectively, for PlGF criteria of ≤ 350.

FIGURE-V   PLGF Values
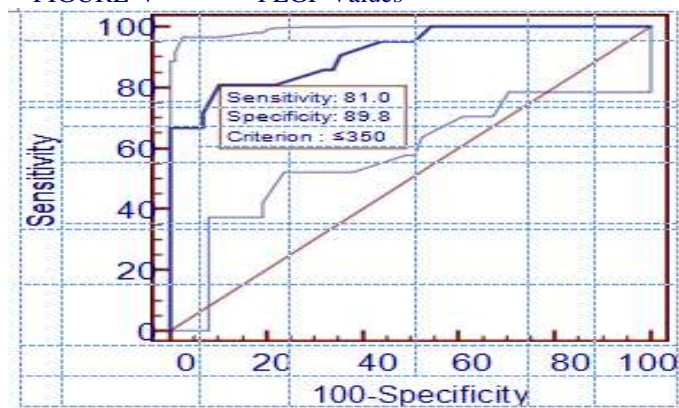
Of the 80 study patients, 14 (17.5%) had previous history of pre eclampsia and 10 (71.4%) among those with previous history of pre eclampsia, developed subsequent pre eclampsia.
So there is a statistically significant association between previous history of pre eclampsia and subsequent preeclampsia in our study.

Distribution of gestational age at which pregnancy was terminated in Normotensive mothers compared with preeclamptic mothers is shown in bar diagram below

FIGURE-VI
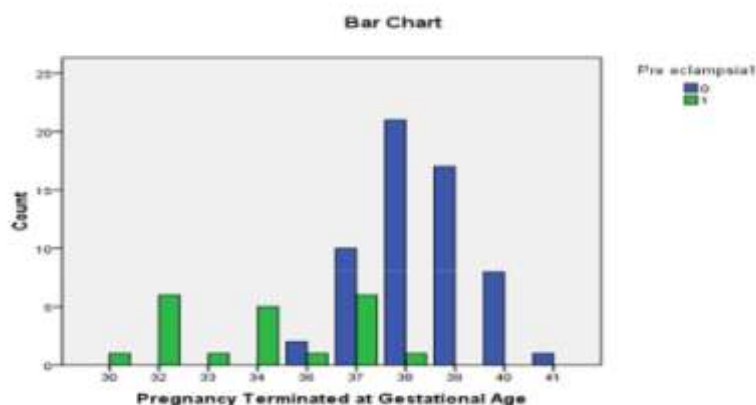
The blue bars represent those without pre eclampsia and the green bar represents the pre eclamptic patients.
Pre eclamptic patients had pregnancies terminated at earlier gestational age compared to those who were normal and carried it to term. The patients who developed severe pre eclampsia terminated by 32weeeks (n=6 ; 28.6%) where as the those who had mild pre eclampsiacarried their pregnancy till 37 weeks,(n=6 ; 28.6%).

TAB LE-IV   Anova
Pregnancy Terminated at Gestational Age

	Sum of Squares	Df	Mean Square	F	Sig.
Between Groups	286.324	2	143.162	75.059	.000
Within Groups	146.863	77	1.907		
Total	433.187	79			

More severe the degree of pre eclampsia the earlier the pregnancy was terminated.
distribution of gestational age at which pregnancy was terminated
FIGURE-VII
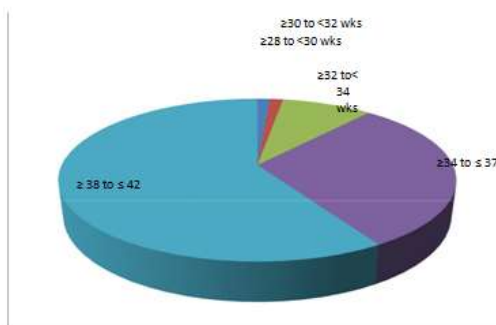
Of the 80 patients, 47 delivered at term (58.75%), 33 delivered preterm (41.25%). 3 patients had pregnancy terminated within 2 weeks of detecting low PlGF value. Out of the 3 patients, 2 had severe pre eclampsia, all 3 patients were 2ndgravida, all 3 underwent cesarean sections.2 out of the 3 babies showed features of IUGR.2 out of the 3 patients had previous history of pre eclampsia. In this study, Youden index is

0.7078 and Area under the ROC curve is 0.92 which implies the diagnostic accuracy of PlGF is significant with a sensitivity and specificity of 80.95% and 89.83% respectively, for PlGF criteria of ≤ 350.

IV.   Discussion

Serum PlGF in early second trimester could be an effective predictor of early onset preeclampsia in women who are overweight/obese pregnant than in women with normal BMI or who are underweight[15], our study correlates with this study by Ghosh et al.

In his study Akolekar concluded that a combination of maternal characteristics including the obstetric history, uterine artery PI and maternal serum PlGF taken between 11 and 13 weeks would prove to be an effective screening tool for patients suspected to develop pre eclampsia later on in their pregnancy. [16]

Pre eclampsia had an increased incidence of 3.2 times that of the control group with confidence interval of 95%,and the mean PlGF value was 62.5 pg/ml in pre eclamptic individuals. This study by Necmiye Dover concluded that low PlGF values in the second trimester have increased risk of developing pre eclampsia.[17]
Our study correlates well with this finding

 Chikako Hirashima,in his study concluded that the values of PlGF or the sFlt-1/PlGF ratio appears to be clinically useful in the second trimester and also useful in the prediction of early onset pre eclampsia .In the early third trimester the combination of angiogenesis-related factors and other risk factors yielded the best LR+.[18] Further studies are needed to confirm the findings and in combination may be a better predictor of pre eclampsia. Bolin et al has reviewed articles and has stated that in the first trimester, maternal characteristics such as mean arterial pressure, uterine artery pulsatility index and the biochemical markers PAPP-A and PlGF is of predictive value with 95% specificity and 93% sensitivity for early-onset and 36 % for late-onset pre-eclampsia.[19]

The threshold value of PlGF at which a women would develop pre eclampsia, has not been clearly defined among the Indian population which once again states the need for more work on this accurate predictive test in order to individualize the level of surveillance during pregnancy.

V.   Conclusion

Placental Growth Factor (PlGF) levels were significantly lower in patients prone to develop pre eclampsia. It also helps in risk stratification of pre eclamptic patients, as mild and severe pre eclampsia thereby early detection and appropriate management is possible in these patients. Studies with larger population size are needed in our setting to further define the diagnostic accuracy and predictive capabilities of the various angiogenic markers.

Bibilography

[1].   World Health Organization. Make Every Mother and Child Count. World Health Report, 2005. World Health Organization: Geneva, Switzerland, 2005.
[2].   Lewis G (ed). Confidential Enquiries into Maternal and Child Health. Why Mothers Die 2000–2002: The Sixth Report of United Kingdom Confidential Enquiries Into Maternal Deaths in the United Kingdom.RCOG Press: London, 2004.
[3].   Dekker G, Sibai B (2001) Primary, secondary, and tertiary prevention of pre-eclampsia. Lancet 357: 209–215. doi: 10.1016/S0140-6736(00) 03599
[4].   Belghiti J, Kayem G, Tsatsaris V, Goffinet F, Sibai BM, et al. (2011) Benefits and risks of expectant management of severe preeclampsia at less than 26 weeks gestation: the impact of gestational age and severe fetal growth restriction. Am J ObstetGynecol 205: 465 e461–466. doi: 10.1016/j.ajog.2011.06.062
[5].   Odendaal HJ, Pattinson RC, Bam R, Grove D, Kotze TJ (1990) Aggressive or expectant management for patients with severe preeclampsia between 28–34 weeks' gestation: a randomized controlled trial. ObstetGynecol 76: 1070–1075.
[6].   Jeanne Sibiude mail, Jean Guibourdenche, Marie-Danielle Dionne, Camille Le Ray, Placental Growth Factor for the Prediction of Adverse Outcomes in Patients with Suspected Preeclampsia or Intrauterine Growth Restriction; Published: November 28, 2012DOI: 10.1371/journal.pone.0050208
[7].   Lakshmi Tanuja Petla, Rosy Chikkala :Biomarkers for the management of pre-eclampsia in pregnant women, Indian J Med Res 138, July 2013, pp 60-67
[8].   Levine RJ, Maynard SE, Qian C, Lim KH, England LJ, Yu KF, Schisterman EF, Thadhani R, Sachs BP, Epstein FH, Sibai BM, Sukhatme VP, Karumanchi SA. Circulating angiogenic factors and the risk of preeclampsia. N Engl J Med.2004; 350: 672–683.
[9].   Knudsen UB, Kronborg CS, Von Dadelszen, Kopfer K, Lee SW, Vittinghus E, Allen JG, Redman CW. A single rapid point-of-care placental growth factor determination as an aid in the diagnosis of preeclampsia. Pregnancy Hypertens. 2011;2:8-15.Google Scholar
[10].   Chesley LC. History and epidemiology of preeclampsia-eclampsia. ClinObstet Gynecol. 1984;27(4):801-20.
[11].   Hertig A, Liere P23. . New markers in preeclampsia. ClinChimActa 2010; 411 : 1591-5.
[12].   Romero R, Nien JK, Espinoza J, Todem D, Fu W, Chung H, 24. et al. A longitudinal study of angiogenic (placental growth factor) and anti-angiogenic (soluble endoglin and soluble vascular endothelial growth factor receptor-1) factors in normal pregnancy and patients destined to develop preeclampsia and deliver a small-for-gestational-age neonate. J Maternal Fetal Neonatal Med 2008; 21 : 9-23.
[13].   Lapaire O, Shennan A, Stepan H. The preeclampsia biomarkers soluble fms-like tyrosine kinase-1 and placental growth factor: current knowledge, clinical implications and future application. Eur J ObstetGynecolReprodBiol 2010; 151 : 122-9
[14].   Betsy Varughese, Rajesh Kumar, NeerjaBhatla, S.N Dwivedi , ManojDhingra, RenuDhingra. Urinary Placental Growth Factor: A Promising Marker for Screening Preeclampsia;International Journal of Scientific and Research Publications, Volume 2, Issue 11, November 2012

[15]. Ghosh SK, Raheja S, Tuli A, Raghunandan C, Agarwal S. Serum placental growthfactor as a predictor of early onset preeclampsia in overweight/obese pregnantwomen. J Am SocHypertens. 2013 Mar-Apr;7(2):137-48 doi:10.1016/j.jash.2012.12.006. Epub 2013 Feb 6. PubMed PMID: 23394804.d

[16]. Akolekar R, Syngelaki A, Sarquis R, Zvanca M, Nicolaides KH. Prediction of early, intermediate and late pre-eclampsia from maternal factors, biophysical and biochemical markers at 11-13 weeks. Prenatal Diagn 2011;31(1):66-74.

[17]. Necmiye Dover,⊠ Hacer C. Gulerman, SevkiCelen, SerkanKahyaoglu, and OkanYenicesuPlacental Growth Factor: As an Early Second Trimester Predictive Marker for Preeclampsia in Normal and High-Risk Pregnancies in a Turkish PopulationJObstetGynaecol India. 2013 Jun; 63(3): 158–163.

[18]. Hirashima C, Ohkuchi A, Takahashi K, Usui R, Matsubara S, et al. (2014) Prediction of Early-Onset Preeclampsia Using Angiogenesis-Related Factors. Med J ObstetGynecol 2(2): 1025.

[19]. Bolin, M. 2012. Pre-eclampsia – Possible to Predict?: A Biochemical and Epidemiological Study of Pre-eclampsia. ActaUniversitatisUpsaliensis. Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Medicine 835. 66 pp. Uppsala. ISBN 978-91-554-8523-8