Endothelin-1 Level In Early Onset Preeclampsia

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Abstract

Introductions: Preeclampsia is a specific syndrome in pregnancy as a result of abnormal placental invasion leading to placental hypoperfusion. Persistent hypoxia of the placenta causes the release of various inflammatory mediators into the circulation and results in local endothelial dysfunction. Increased endothelin-1 (ET-1) secretion and increased inflammatory mediators occur in preeclampsia.
Aims: To analyze the relationship between endothelin-1 and early onset preeclampsia.
Methods: This crosssectional study included 50 pregnant women with early onset preeclampsia (n=25) and normal pregnancies (n=25). Pregnant women aged 20-35 years with single intrauterine fetus, primigravida and multigravida who experienced early onset preeclampsia < 34 weeks of gestation were included in this study. Endothelin-1 levels was measured using the Enzyme Linked Immunosorbant Assay (ELISA) method. The analysis was performed using the Mann-Whitney test. *Receiving Operator Characteristic* (ROC) curve analysis was used to find the cut-off value and diagnostic accuracy of endothelin-1 levels.
Results: The mean of ET-1 level was significantly higher in early onset preeclampsia (0.732 ± 0.56 pg/mL) compared to normal pregnancy (0.318 ± 0.09 pg/mL) with value of p = 0.000. The ROC analysis showed the AUC value of 87.8% (p = 0.000, 95% CI 78.6%-97.1%). The cut-off value for ET-1 was 0.385 pg/mL, with 80% sensitivity and 68% specificity (PR=3.14; 95% CI 1.40-7.03).
Conclusion: Increased levels of endothelin-1 (ET-1) significantly associated with early onset preeclampsia. ET-1 level ≥ 0.385 pg/mL in pregnant women with < 34 weeks of gestation is potential biomarkers to predict the occurrence of early onset preeclampsia with a risk up to 3.14 times.

Keywords: *Early onset preeclampsia, Endothelin-1 (ET-1*

INTRODUCTION

The three main causes of maternal mortality in the world are gestational bleeding (30%), hypertension in pregnancy (25%) and infection (12%). (POGI & HKFM, 2016)Preeclampsia is a specific syndrome in pregnancy as a result of abnormal placental invasion causing placental hypoperfusion.

Persistent hypoxia of the placenta causes the release of various inflammatory mediators into maternal circulation and results in local endothelial dysfunction. Alteration of vasoactive substances such as thromboxan A2, increased lipid peroxide, increased secretion of endothelin-1 (ET-1) and increased inflammatory response mediators such as C-Reactive Protein (CRP) occur in preeclampsia.(Mao et al., 2010; Sağsöz & Küçüközkan, 2003) ET-1 is a potent circulating vasoactive agent 10 times higher than angiotensin II. ET-1 is involved in vasoconstriction of uterine artery vessels as well as systemic hypertension.(Dechanet et al., 2011; George & Granger, 2011; Zeng et al., 2015)

Early preeclampsia does not cause any symptoms and signs. Proper primary prevention is needed to early detect the incidence of preeclampsia.1 Several studies have examined the increase in ET-1 levels in pregnant women with preeclampsia.(Abouzaid et al., 2014; Cuningham.F.Gary, 2014; Zeng et al., 2015) In this study, we aimed to analyze the relationship between ET-1 levels and early onset preeclampsia.

METHODS

A crosssectional study of 50 pregnant women with early onset preeclampsia (n=25) and normal pregnancies (n=25) who came to the Department of Obstetrics and Gynecology, Dr. Kariadi Semarang, Puskesmas Halmahera, Puskesmas Ngesrep and Puskesmas Bulu, Semarang, Indonesia as well as a private midwife practice during the study period. Pregnant women aged 20-35 years with single intrauterine fetus, primigravida and multigravida with early onset preeclampsia < 34 weeks gestation who were willing to be the study subjects were included in this study. Exclusion criteria included unclear first day of last menstrual period (LMP), history of preeclampsia, obesity (BMI > 30 kg/m2), presence of intrauterine fetal death (IUFD), premature rupture of membranes, intrauterine infection and history of chronic diseases, such as diabetes mellitus, cerebrovascular disease, chronic hypertension, chronic renal failure and chronic infections.

Early onset preeclampsia was assessed by measuring blood pressure at gestational age < 34 weeks using a digital manometer.

Measurements were made on the right and left arms using a cuff to fill 1/3 of the upper arm at the level of the right atrium after 15 minutes of rest lying on the left side. Preeclampsia was defined when systolic blood pressure ≥ 140 mmHg and diastolic ≥ 90 mmHg accompanied by proteinuria (urine dipstick > +1).1,9 All study subjects who met the study criteria underwent blood serum sampling. The serum obtained was stored in a refrigerator at a temperature of ≤ -200C. Endothelin-1 levels was measured using the Enzyme Linked Immunosorbant Assay (ELISA) method on serum obtained at the GAKI Laboratory, Semarang, Indonesia.

All demographic data such as maternal age, body mass index, gravidity, education level, gestational age were recorded and analyzed. The mean endothelin-1 levels were analyzed using the Mann-Whitney test. Receiving Operator Characteristic curve analysis was used to find the cut-off value and diagnostic accuracy of endhotelin-1 in early onset preeclampsia.

RESULTS

The mean age of pregnant women in this study was 28.54 ± 4.04 years old, body mass index (BMI) was 25.69 ± 2.87 kg/m2, endothelin-1 (ET-1) level was of 0.52 ± 0.45 pg/mL and a gestational age of 29.36 ± 3.09 weeks. Statistical analysis of early-onset preeclampsia group and the normal pregnancy group found that age, BMI, education, occupation, gravidity and gestational age were not significantly different (p > 0.05). (Table 1)

Table 1. Subjects characteristics

Variable	Early Onset Preeclampsia (*n*=25)	Normal Pregnancy (*n*=25)	p
Maternal Age	29,56 ± 3,42	27,52 ± 4,41	0,086‡
Body mass index	26,45 ± 2,45	24,94 ± 3,10	0,123‡
Education			
Elementary	2 (8)	2 (8)	0,724¥
Junior High School	4 (16)	5 (20)	
Senior High School	12 (48)	15 (60)	
Diploma	2 (8)	1 (4)	
Undergraduate	5 (20)	2 (8)	
Employment Status			
Unemployed	10 (40)	14 (56)	0,258¥
Employed	15 (60)	11 (44)	
Gravidity			
Primigravida	8 (32)	10 (40)	0,556¥
Multigravida	17 (68)	15 (60)	
Gestational Age	29,76 ± 3,41	28,96 ± 2,74	0,188‡

Notes: ‡ Mann-Whitney; ¥ Chi-square

Table 2. The difference of ET-1 levels in early-onset preeclampsia and normal pregnancy

Group	n	Min	Max	Mean±SD (pg/mL)	Median	p
Normal Pregnancy	25	0,18	0,45	0,318 ± 0,09	0,31	0,000

Early onset preeclampsia	25	0,31	2,43	0,732 ± 0,56	0,55

Table 2 showed the mean level of ET-1 were higher in early onset preeclampsia (0.732 ± 0.56 pg/mL) compared to normal pregnancies (0.318 ± 0.09 pg/mL). Analysis showed that the ET-1 levels between the early-onset preeclampsia group and the normal pregnancy group had a significant difference with a value of p = 0.000. ROC curve analysis showed that ET-1 has a fine diagnostic value because the curve is far from the 50% line and close to 100%. Based on this curve (Figure 1), the AUC value was 87.8% (95% CI 78.6%-97.1%) with a p value = 0.000. The cut-off value of ET-1 was 0.385 pg/mL, with 80% sensitivity and 68% specificity.

Table 3. The prevalence ratio of ET-1 levels with the incidence of early onset preeclampsia

ET-1 level	Incidence		∑	p	PR
	Early onset preeclampsia	Normal Pregnancy			
Tinggi (≥0,385 pg/mL)	20 (71.4%)	8 (28.6%)	28	0.002 *	3,14
Rendah (< 0,385 pg/mL)	5 (22.7%)	17 (77.3%)	22		
∑	25	25			

*chi-square

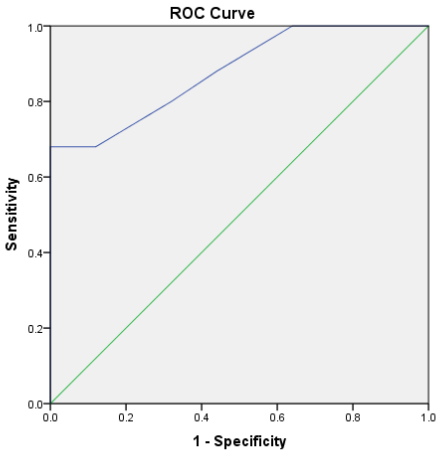

Figure 1. Receiving Operator Characteristic (ROC) curve analysis of endothelin-1 level in early onset preeclampsia.

Analysis showed a relationship between ET-1 levels and early onset preeclampsia with a value of p = 0.002 (p < 0.05). Based on the table above, (PR = 3.14; 95% CI 1.40 - 7.03), pregnant women with < 34 weeks of gestation with ET-1 levels ≥ 0.385 pg/mL, posses 3.14 times greater risk of becoming early onset preeclampsia compared to pregnant women < 34 weeks gestation with ET-1 levels < 0.38 pg/mL.

DISCUSSION

The increased risk of preeclampsia was found to almost double in pregnant women aged 40 years. Young age does not significantly increase the risk of preeclampsia. Meanwhile, nulliparous has almost threefold risk compared

to primiparous or multiparous(Duckitt & Harrington, 2005; Septiani et al., n.d.). Septiyani et al, showed that pregnant women aged < 20 and > 35 years old had a 15,731-fold risk of experiencing preeclampsia compared to respondents aged 20–35 years. A high BMI before pregnancy is a risk factor for preeclampsia. Obesity is another risk factor for preeclampsia. Obesity increases the risk of preeclampsia by 2.47 times, whereas women with a BMI before pregnancy > 35 have a 4-fold risk of preeclampsia.(Septiani et al., n.d.)

Saraswati et al, showed that the level of education was not related to the incidence of preeclampsia in pregnant women.(Saraswati & Mardiana, 2016) Chang et al, also mentioned that occupation was not related to the incidence of preeclampsia and gestational hypertension but in this study the level of activity and the level of work stress was not measured.(Chang et al., 2010) Primigravida posses 2.173 times of experiencing preeclampsia compared to multigravidas.(Septiani et al., n.d.) The results of this study were also in accordance with the research conducted at Sanglah Hospital in 2017, it was found that the majority of preeclamptic patients occurred in primigravidas.(Vincent et al., 2018)

Preeclampsia is hypertension in pregnancy characterized by proteinuria and extensive endothelial dysfunction. (George & Granger, 2011) Preeclampsia initiated by abnormal placental invasion which causes placental hypoperfusion, triggers persistent hypoxia in the placenta. Following this hypoxia, various inflammatory mediators released to the maternal circulation causing local endothelial dysfunction. Preeclampsia altered the production of vasoactive substances such as thromboxan A2, increased lipid peroxide, increased secretion of ET-1 and increased inflammatory response mediators such as C-Reactive Protein (CRP).(Abouzaid et al., 2014; Sağsöz & Küçüközkan, 2003) Endothelial dysfunction increases the level of ET-1, a potent circulating vasoactive agen, 10 times higher than angiotensin II. ET-1 involved in vasoconstriction of uterine artery vessels as well as systemic hypertension.(Dechanet et al., 2011; George & Granger, 2011; Zeng et al., 2015)

ET-1 is a vasoconstrictor that can be used as a predictor of hypertension and preeclampsia.(George & Granger, 2011) Several studies have shown a relationship between ET-1 and preeclampsia. ET-1 levels in preeclampsia increased significantly compared to normal pregnancy.(George & Granger, 2011; Jain, 2012) The release of ET-1 by endothelial cells, both directly by endothelial cells or through reduced availability of VEGF can cause hypertension and may contribute to proteinuria by modulating nephrine expression. ET-1 causes inhibition of cell proliferation and vitality, triggers oxidative stress on endothelial cells or placenta cells by changing the balance of oxidant levels (increased the level of malondialdehyde) and antioxidants. This condition changes the defense system of the placental tissue against oxidative stress, resulting in placental tissue dysfunction which triggers the production of substances that trigger maternal syndrome in preeclampsia.(Aggarwal et al., 2012; Chang et al., 2010)

Aggarwal et al, compared ET-1 levels in preeclampsia (1.52 ± 0.55 pg/mL) higher than normal pregnancy (0.88 ± 0.35 pg/mL).(Aggarwal et al., 2012)Sheng et al, showed that ET-1 levels in early onset preeclampsia (8.3 ± 7.8 pg/g) is higher than normal pregnancy (18.8 ± 1.3 pg/g).(Sheng et al., 2019) Our results are consistent with previous studies, where the level of ET-1 in patients with early onset preeclampsia was found to be higher than normal pregnancy.

Slowinski et al, stated that ET-1 levels ≥ 0.30 pg/mL had a 4.6 times greater risk of becoming preeclampsia.(SLOWINSKI et al., 2002) After analyzing the ROC curve, our study showed a cut-off value of ET-1 levels of 0.385 pg/mL, with 80% sensitivity and 68% specificity. This showed that pregnant women with < 34 weeks gestation with ET-1 levels ≥ 0.385 pg/mL, posses 3.14 times greater risk of developing early onset preeclampsia compared with pregnant women with < 34 weeks gestation with ET-1 levels < 0.385 pg/mL.

Our discussion pointed that ET-1 levels play a significant role to predict preeclampsia because of its high sensitivity and specificity. ET-1 levels increased in pregnant women with early onset preeclampsia and there was a

relationship between ET-1 levels and early onset preeclampsia.19,20 ET-1 level was a potential biomarker to predict the occurrence of early onset preeclampsia. The limitation of this study, we didn't analyze genetic influence as one of the factors that play a role in the incidence of preeclampsia. Our study did not use physical examination and laboratory investigations to screen for exclusion criteria. Further comprehensive studies are needed to examine the effect of ET-1 and other anti-angiogenic factors on preeclampsia to asses causative relationship simultaneously.

CONCLUSION

Increased levels of endothelin-1 (ET-1) significantly associated with early onset preeclampsia. ET-1 level ≥ 0.385 pg/mL in pregnant women with < 34 weeks of gestation is potential biomarkers to predict the occurrence of early onset preeclampsia with a risk up to 3.14 times.

We suggest that pregnant women who have preeclampsia risk factors at gestational age > 20 weeks should be tested for ET-1 levels. In addition, it is necessary to study ET-1 levels with a prospective cohort study approach for gestational age < 20 weeks until the early onset preeclampsia established.

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