Research Article

Role of C-Reactive Protein, Erythrocyte Sedimentation Rate, Progesterone and Estradiol Hormone Levels in First Trimester Threatened Abortion

Peran C-Reactive Protein, Laju Endap Darah, Progesteron dan Estradiol pada Abortus Imminens Trimester Pertama

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Abstract

Objective: To assess correlation of High sensitivity C-Reactive Protein (Hs-CRP), erythrocyte sedimentation rate (ESR), progesterone and estradiol levels in the first trimester threatened abortion incidence in Dr. Zainoel Abidin Hospital, Banda Aceh.

Methods: Case control design used in this study divided threatened abortion into a case group and normal pregnancy into a control group with a total of 20 subjects for each group. This research was conducted in the Emergency Room and Obstetric Ward of Dr. Zainoel Abidin hospital Banda Aceh in 2019. Eta correlation test was conducted to find out the link between variables towards threatened abortion with 95% confidence level followed by the Receiver Operating Curve (ROC) analysis to find out the cut off points.

Results: Progesterone levels (14.76 ng/mL), estradiol (427.61 pg/mL), Hs-CRP (2.57 mg/L) and ESR (28.75 mm/hour) case group were lower compared to the control group. Incidence of threatened abortion correlates to progesterone and estradiol with the correlation strength respectively -0.838 and -0.416.

Conclusions: Progesterone and estradiol correlate negatively with first-trimester abortion incidence. Evaluation of these two hormones levels is useful for diagnostic purposes and screening of threatened abortion with a cut point of progesterone 23.03 ng/mL and estradiol 468.8 pg/mL.

Keywords: C-Reactive Protein, erythrocyte sedimentation rate, estradiol, progesterone, threatened abortion

Abstrak

Tujuan: Penelitian ini bertujuan untuk menilai korelasi antara kadar High sensitivity C-Reactive Protein (Hs-CRP), Laju Endap Darah (LED), progesteron dan estradiol terhadap kejadian abortus imminens pada trimester pertama di RSUD Dr. Zainoel Abidin Banda Aceh.

Metode: Desain kasus kontrol digunakan dalam penelitian ini dimana kejadian abortus imminens menjadi kelompok kasus dan kehamilan normal menjadi kelompok kontrol dengan jumlah masing-masing kelompok sebanyak 20 sampel. Penelitian ini dilakukan di Instalasi Gawat Darurat dan ruang rawat Obstetri Rumah Sakit Umum Dr. Zainoel Abidin (RSUDZA) Banda Aceh pada tahun 2019. Uji korelasi Eta digunakan untuk mengetahui korelasi antar variabel terhadap abortus imminens dengan tingkat kepercayaan 95% dilanjutkan dengan analisis Receive Operating Curve (ROC) untuk menentukan titik potong.

Hasil: Kadar progesteron (14,76 ng/mL), estradiol (427,61 pg/mL), Hs-CRP (2,57 mg/L) dan LED (28,75 mm/jam) kelompok kasus lebih rendah dibandingkan kelompok kontrol. Kejadian abortus imminens berkorelasi terhadap progesteron dan estradiol dengan kekuatan korelasi (R) secara berurutan -0,838 dan -0,416.

Kesimpulan: Progesteron dan estradiol berkorelasi negatif terhadap kejadian abortus imminens pada trimester pertama kehamilan. Evaluasi kadar kedua hormon tersebut bermanfaat untuk kepentingan diagnostik dan penapisan abortus imminens dengan titik potong progesteron 23,03 ng/mL dan estradiol 468,8 pg/mL.

Kata kunci: abortus imminent, C-Reactive Protein, estradiol, laju endap darah, progesteron.

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INTRODUCTION

Abortion is known as a common complication during pregnancy. Abortion not only can cause morbidity but also has impacts towards social, psychological and overall quality of life. In 20% of cases, abortion is able to cause stress conditions. The cause of abortion has always been a question for couples who experience it. In general, abortion is caused by abnormalities in the results of conception or by maternal factors which cause pregnancy to not continue. Abortion is generally caused by various chromosomal abnormalities such as trisomy, monosomy and polypodia, which predicted to occur occur in 60% of cases.[1] In addition, various other factors such as chronic diseases, obesity, alcohol consumption, the consumption of certain drugs, infections and hormonal disorders are reported to increase the incidence of first-trimester abortion.[2-4]

Inflammatory processes which trigger early abortion allows the development of inflammatory biomarkers in predicting pregnancy failure. Using acute and chronic inflammatory parameters has been recommended lately as in the increased activity of fibrinogen and plasma globulin which shows that inflammation can be seen with an increase in erythrocyte sedimentation rate (ESR). The use of other biomarkers that also has a fairly good correlation with ESR is C-Reactive Protein (CRP), which is a parameter that has been widely used to help establish an early diagnosis and treatment of an inflammatory process and also infection. Elevating the levels of CRP has been known to show a risk of endothelial thrombosis that causes complications of pregnancy failure. Other research also reports some genotypes in CRP is significantly correlated with abortion and higher CRP findings during acute chorioamnionitis that play an important part in pathological abortion during early pregnancy.[5, 6]

The involvement of the endocrine system during the pregnancy process is vital in maintaining an adequate uterine environment. The progesterone hormone helps protect the environment through secretory changes stimulation for implantation and suppressing the myometrial contractility also increasing the nitric oxide synthesis regulation in the endometrium to suppress the adverse pro-inflammatory cytokine response. Progesterone is also considered to play an important part in carrying out immunomodulating mechanisms in pregnancy that will facilitate different allogenic responses to trophoblast cells. Trophoblastic invasion of maternal spiral arteries will occur without interference with progesterone. The occurrence of vascular remodelling triggered by adequate trophoblast cells invasion can produce a good uteroplacental circulation, which by facilitating high blood flow with a low resistance system. This will then result in a good placentation process.[7, 8]

Besides progesterone, estrogen is also important during ovulation and pregnancy. Estrogen increases slowly during pregnancy with the final product being estradiol. Related to the recurrent pregnancy loss (RPL) case study, estradiol together with FSH was significantly increase. When evaluating the occurrence of spontaneous abortion in obese women, it shows that estradiol levels that are high significantly increase spontaneous abortion risk. The correlation that was also reported was the mechanism of increasing levels of prostaglandin (PGF2) as an effect of increasing concentrations of estrogen receptors (nucleus and cytosol estradiol prematurely with oxytocin) which can trigger spontaneous abortion in pregnant women.[9] The purpose of this study was to evaluate changes in biomarkers in C-Reactive Protein levels, erythrocyte sedimentation rate (ESR), progesterone and estradiol in early pregnancy and their correlation to the threatened abortion incidence.

METHODS

This research is a correlative observational study with a case control design. This research was conducted at the Obstetric Emergency Installation and Obstetric Ward of the General Hospital Dr. Zainoel Abidin (RSUDZA) Banda Aceh during 2019.

Our inclusion criteria include first-trimester pregnant women aged 20-35 years old who are experiencing threatened abortion (case group) and without threatened abortion (control group). In our study, the exclusion criteria were patients with a history of autoimmune chronic disease, chronic infection (Tuberculosis), endocrinological metabolic disorders, coronary heart disease, haematological or malignant abnormalities, multiple pregnancies, extrauterine pregnancy, mole hydatidiform, hormonal therapy or progesterone supplementation, anatomical abnormalities of reproduction and BMI = 30 kg/m2. The amount of the subjects in this research was determined based on the formula of an

unpaired numerical analytic study and obtained a sample of each group to be 20 samples.

Blood specimens for the examination of High sensitivity C-Reactive Protein (Hs-CRP) levels, erythrocyte sedimentation rate (ESR), progesterone and estradiol were taken and sent to Prodia Laboratory Banda Aceh. All data collected were analysed further using the ETA test and Receiver Operating Characteristic (ROC) curve.

RESULTS

Based on collected data at the Emergency Obstetric Installation and Obstetric Ward of the General Hospital dr. Zainoel Abidin during the research period, the data obtained as many as 40 pregnant women with and without threatened abortion. In the following table, data on the characteristics of our study is shown.

Table 1. Sample Characteristics Data

Characteristics	Threatened Abortion n = 20	Non-Threatened Abortion n = 20
Age (year)*	29.2 ± 4.26	28.2 ± 3.65
Education		
Elementary School	1 (5)	0
Middle School	2 (10)	1 (5)
High School	7 (35)	7 (35)
Diploma	4 (20)	5 (25)
Bachelor	5 (20)	7 (35)
Master	1 (5)	0

	Threatened Abortion	Non-Threatened Abortion
Occupation		
Occupied	8 (40)	6 (30)
Unemployment	12 (60)	14 (70)
Age of gestation (week)		
< 8	4 (20)	10 (50)
8-12	16 (80)	10 (50)
Parity		
Nulliparous	7 (35)	8 (40)
Multiparous	13 (65)	12 (60)
Observation		
Spontaneous Abortion	9 (45)	2 (10)
No spontaneous abortion	11 (55)	18 (90)

Based on age, it is found that the abortion group was 1 year younger than the non-threatened abortion group. The highest number of threatened abortion groups was high school graduates (35%), while the highest number of non-threatened abortion groups were high school and undergraduate graduates with the same percentage of 35%. Eighty per cent of the group with threatened abortion was with 8-12 weeks gestation. The two groups of threatened abortion and non-threatened abortion were multiparous in the order of 13 and 12 respectively. The incidence of spontaneous abortion based on observations was 9 people in the threatened abortion group and 2 people in the non-threatened abortion group.

Table 2. Correlation Analysis of Serum Progesterone, Estradiol, Erythrocyte Sedimentation Rates (ESR) and Hs-CRP with Threatened Abortion

	Average (Min – Max)		R*	P-value
	Threatened Abortion	**Non-Threatened Abortion**		
Progesterone	14.76 (2.8 – 23.40)	37.46 (21.6 – 53.38)	- 0.838	< 0.001
Estradiol	427.61 (101.9 – 1080)	771.07 (154 – 1676.5)	- 0.416	0.008
ESR	28.75 (14 – 44)	33 (22 – 44)	- 0.254	0.114
Hs-CRP	2.57 (0.3 – 4.8)	3.92 (2.8 – 5)	- 0.293	0.066

Table 2 presents the results of the correlation test of progesterone, estradiol, Eryhtrocyte Sedimentation Rates (ESR) and Hs-CRP with threatened abortion. Based on the Eta test results, both progesterone and estradiol levels between the two groups showed a negative correlation with strong and moderate correlation, respectively. Our results show no significant correlation in the levels of ESR and Hs-CRP between the two groups (p> 0.05).
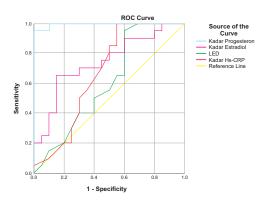

Figure 1. ROC curve of Progesterone, Estradiol, ESR and Hs-CRP levels for Threatened Abortion

Table 3. Interpretation of Area Under the Curve (AUC) Results

Variable	Area	*P-value*
Progesterone	0.995	< 0.001
Estradiol	0.740	0.009
Erythrocyte sedimentation rate	0.606	0.25
Hs-CRP	0.669	0.068

Figure 1 shows the ROC curve of progesterone, estradiol, ESR and Hs-CRP levels against threatened abortion with Area Under the Curve (AUC) as presented in Table 3. Based on the table, it is known that progesterone shows a higher level of discrimination compared to estradiol with percentages respectively of 99.5% and 74%.

Table 4. Progesterone and Estradiol Diagnostic Significance Values toward Threatened Abortion

Variable	Intersection	Sensitivity	Specification
Progesterone (g/L)	23.03	95	95
Estradiol (pg/mL)	468.8	70	70

The diagnostic significance of progesterone and estradiol for threatened abortion is presented in table 4 above. Progesterone levels 23.03 showed the best sensitivity and specificity values of 95% and estradiol levels of 468.8 showed the best sensitivity and specificity with a value of 70% threatened abortion predictors in the first trimester of pregnancy.

DISCUSSION

Based on the data obtained from this study, found that Hs-CRP levels in the group of threatened abortion patients were lower than those with non-threatened abortion group with average levels of 1.57 mg/L and 3.92 mg/L, respectively. One study reported that the expression of IL-10, IL-6, TNF-a and TNF-R1 was lower in women who is experiencing threatened abortion compared to normal pregnancies. In this regard, pro-inflammatory cytokine secretions such as IL-1, IL-6 and endothelin-1 are induced by CRP.[10] This causes disruption of trophoblastic protein synthesis and uteroplacental inflammation in the area of implantation and placentation in the first-trimester pregnancy. The study also reported that serum Hs-CRP levels also decreased in pregnancy with threatened abortion.[11]

Referring to the results of this study, there was a difference in Hs-CRP levels of 2.35 mg/L between the two groups. However, it was statistically concluded that Hs-CRP levels between the two groups did not have a significant correlation (p=0.066). The same thing was also reported where the Hs-CRP examination did not provide information in the management of threatened abortion.[11] Another study also concluded that Hs-CRP did not contribute as a predictor of complications in the early phases of pregnancy.[12]

However, the role of CRP in pregnancy has not shown consistent results. The study reported concluded that an abnormal CRP response in the first trimester of pregnancy indicates a disruption in fetomaternal relationships and is associated with the incidence of abortion. They also concluded that CRP testing could be a predictor or even a screening modality in some cases that would carry out an immunotherapy program.[13]

Elevated CRP levels in early pregnancy are not known with certainty. The main stimulant that produces CRP is IL-6 which is produced by macrophages and active monocytes. Macrophages are the main leukocyte in the decidual cells early in pregnancy. Its existence is undoubtedly a major component of interaction between the trophoblast and the maternal immune system. Increased function in producing IL-6 is believed to be the cause of increased CRP.[13, 14]

Another parameter that becomes the evaluation variable in this study is ESR. The average values of ESR in the groups with and without threatened abortion were 28.75 and 33 mm/hour, respectively. The difference between the two values does not indicate a difference that is significant statistically (p=0.144). Referring to the normal levels of ESR in the first trimester of pregnancy (4-57 mm/hour), the levels shown in this research are still in the normal range. Normal ESR levels will decrease in the second trimester (7-47 mm/hour) and will increase again in the third trimester (13-70 mm/hour).[15] A study conducted by Khazal and Zangana of 300 normal pregnant women concluded that ESR had increased during pregnancy and their levels were influenced by haemoglobin concentration and gestational age.[16]

ESR is an inexpensive and simple examination modality for evaluating inflammation and acute phase responses. This examination was first discovered in 1897, but only in 1981 this examination began to be used for scientific purposes by Robert Fahraeus who first used it for the tests on pregnancy. The value of ESR will increase by 0.85 mm/hour every 5 years due to increased levels of fibrinogen or the incidence of

asymptomatic disease in the elderly.[17, 18]

The ESR examination reflects the erythrocyte aggregation measured in a vertical tube with an internal diameter of 2.5 mm for 1 hour without any vibration at room temperature. Erythrocyte aggregation is influenced by erythrocyte cell surface shape and friction that occurs between erythrocytes. An extreme increase in ESR>100 mm/hour is associated with serious illnesses such as collagen disease in blood vessels, tumours with metastases or severe infections. Other conditions such as anaemia, macrocytosis, increased molecular weight of protein in the blood, hemodilution, hypercholesterolemia, nephrotic syndrome, severe liver disease, thyroiditis and pregnancy are the causes of increased ESR. ESR was lower in the condition of hypofibrinogenemia, hypogammaglobulinemia, polycythemia, microcytosis, hemolytic anaemia, hemoglobinopathy, heart failure, allergic diseases and the consumption of anti-inflammatory drugs that can reduce ESR levels. ESR examination can modality for evaluating inflammation and acute responses and can be used as an alternative to other more expensive new examinations such as CRP.[17]

In this study, progesterone levels were found to be lower than those without abortion with a difference of 22.7 ng/mL. Statistically, progesterone levels are negatively correlated with the occurrence of abortion in the first trimester, with a strong correlation strength. A negative correlation means that with higher levels of progesterone, the risk of threatened abortion occurrence is lowered. Conversely, lower levels of progesterone in the first trimester of pregnancy will increase the risk of abortion. The same thing was reported study evaluating fetal viability in the first trimester of pregnancy when it was concluded that progesterone levels were higher in viable pregnancies by a difference of 13.31 ng/mL when compared to non-viable pregnancies group.[19]

Progesterone is an important hormone in the process of implantation, viability and maintaining pregnancy. Low progesterone levels is used as an accurate predictor of threatened abortion even in conditions where other accompanying conditions cause luteal insufficiencies such as genetic disorders or immunological anomalies, infections of the genital tract, uterine malformations, diabetes and Cushing's syndrome. One study reports that progesterone levels <14 ng/mL were used as predictors of non-viable pregnancy.[20,21]

The cut-off point for the levels of serum progesterone as a predictor for the occurrence of abortion was 11 ng/mL.[22]

In contrast to these two studies, the cut-off point for progesterone levels as a predictor of first trimester threatened abortion is higher at 23.03 ng/mL. Progesterone is a hormone which is essential in the reproductive process. This hormone secretion influences the changes in the uterus and has an important role in the success of the embryo implantation process. Progesterone also modulates the mother's immune response in preventing embryo rejection and suppressing uterine contractions.[23]

Progesterone stimulates Th2 cytokine secretion and Th1 suppresses the maternal immunological system in preventing fetal rejection. Some studies report that progesterone supplementation can reduce the occurrence of abortion with a history of previous abortion. Progesterone and steroids similar to them (progestagens) are useful for preventing abortion and increasing the ratio of embryo implantation in pregnancy programs.[23]

This study proves that the assessment of estradiol levels is a predictor of threatened abortion and shows moderate correlation strength (r=0.416). Estradiol levels are negatively correlated to the occurrence of threatened abortion, where the lower the level of estradiol, the greater the possibility for threatened abortion. Estradiol levels of 468.8 pg/mL were used as cut off points for threatened abortion predictors with a sensitivity and specificity of 70%. Another study reported the same thing as this study, where the levels of estradiol in patients with non-viable pregnancies were lower than viable pregnancies with a difference of 307.9 pg/mL. The difference in estradiol levels in the study was not much different from this study in the amount of 343.46 pg/mL. Then further concluded that a combination of progesterone, estradiol and hCG examination could be a predictor of pregnancy viability with an accuracy of 90.9%.[19]

Various factors influence the incidence of threatened abortion, including embryonic chromosome abnormalities, environmental and immune factors and maternal endocrine dysfunction.[24] Regarding the endocrine system, interactions between the mother and the fetus are strongly influenced by reproductive hormones (sex steroids) including estradiol (E2) which is also reported as a predictor in several cases of first-trimester abortion. Therefore, recommended abortion therapy is in the form of total rest (total

bed rest), luteal support and supplementation of a combination of progesterone and estradiol.[25, 26]

Other than the importance of diagnosis, a comprehensive analysis of reproductive hormones is useful for evaluating the metabolism of reproductive which is generally influenced by 3 factors in the form of genetics, lifestyle and diet, also the environment. Because of these considerations, developing a comprehensive reproductive hormone analysis modality without invasive procedures will benefit the patient and clinical interests. Deficiency of dehydroepiandrosterone (DHEA) levels in urine will reduce estrogen activity in women with threatened abortion.[26]

Estrogen has an important role in normal pregnancy. The increasing of hormone estrogen occurs gradually during pregnancy which increases blood vessel formation and nutrient transfer and supports fetal growth. Any changes in hemostasis that occur on these hormones will have an impact on pregnancy. Low estrogen levels are found in threatened abortion patients and giving estrogen is one of the modalities to save pregnancy in these women. A study reports that hormone-HCG and estrogen are closely related to the incidence of threatened abortion.[27] Similar to this study, estradiol levels in women with threatened abortion were lower than those without threatened abortion with an average level of 427.61 pg/mL.

CONCLUSION

C-Reactive Protein and Erythrocyte sedimentation rate do not correlate with the incidence of the first trimester threatened abortion. Progesterone and estradiol negatively correlated with the incidence of the first trimester threatened abortion. Evaluation of these two hormones levels is useful for diagnostic purposes and screening of threatened abortion with a cut point of progesterone 23.03 ng/mL and estradiol 468.8 pg/mL.

SUGGESTION

Further research needs to be done with more research subjects in order to evaluate ESR with similar haematological profiles subjects (matching) and to comprehensively analyze changes in reproductive hormones in cases of threatened abortion.

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