Research Article

The Role of Progesterone-Induced Blocking Factor in Threatened Abortion

Peran Progesterone-Induced Blocking Factor pada Abortus Iminens

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Abstract

Objective: To determine the role of progesterone-induced blocking factor (PIBF) in women with threatened abortion.

Methods: This was a cross-sectional study. The blood serum of two groups, the first one was women with normal gestation of ≤ 20 weeks, and the second one was those with imminent abortion in Prof. Dr. R.D. Kandou Hospital, and Subcenter Hospital in Manado, was collected. Samples were processed with PIBF ELISA-kit.

Results: PIBF serum value of women in normal gestation ≤ 20 weeks is (47.153±23.830)ng/ml and threatened abortion is (11.540± 4.892) ng/ml, with p value = 0.000.

Conclusion: PIBF serum value of women with threatened abortion is significantly lower compared to women of normal gestation ≤ 20 weeks. This study showed that PIBF has an important role in maintaining pregnancy and can be used as a biologic marker of a pathologic process in pregnancy.

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Keywords: early pregnancy, pregnancy immunology, progesterone-induced blocking factor, threatened abortion

Abstrak

***Tujuan**: Mengetahui kadar PIBF serum perempuan hamil usia kehamilan normal ≤ 20 minggu dan abortus iminens sehingga dapat menambah pemahaman mengenai PIBF sebagai petanda biologis patologi kehamilan, dan dalam upaya terapi rasional pasien dengan abortus iminens.*

***Metode**: Penelitian analitik komparatif potong lintang terhadap 32 pasien yang dibagi menjadi 2 kelompok kehamilan normal ≤ 20 minggu, dan abortus iminens di RSUP Prof Dr. R.D. Kandou, beserta RS Jejaring di wilayah Manado, dilakukan pengambilan sampel serum. Sampel dilakukan pemeriksaan kadar PIBF dengan ELISA-kit. Data diproses menggunakan program SPSS versi 22.0.*

***Hasil**: Kadar PIBF serum perempuan hamil usia kehamilan ≤ 20 minggu normal (47,153±23,830)ng/ml dan abortus iminens (11,540± 4,892) ng/ml (p=0,000).*

***Kesimpulan**: Kadar PIBF serum perempuan hamil dengan abortus iminens lebih rendah secara bermakna dibandingkan kadar PIBF serum perempuan hamil usia kehamilan≤ 20 minggu normal. Hasil ini menunjukkan bahwa PIBF berperanan dalam mempertahankan kehamilan dan dapat digunakan sebagai sarana petanda adanya proses patologis dalam suatu kehamilan.*

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***Kata kunci**: abortus iminens, imunologi kehamilan, kehamilan muda, progesterone-induced blocking factor*

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INTRODUCTION

Fetus is a semi-allogenic tissue which relies inside the body of a women whose immune-competent of producing rejection immune response, therefore modulation of maternal immune response is required to maintain pregnancy.[1]

During pregnancy, many complications may occur, which may lead to early termination of pregnancy. Abortion is defined as the loss of pregnancy either spontaneous or induction before being viable (gestational age < 20 weeks). Abortion can result in physical and emotional disturbance. The incidence of spontaneous abortion is between 15-20% from all pregnancy. Predisposing factors of spontaneous abortion include genetic factors, anatomy, endocrine, immune, infection, thrombophilia, and idiophatic.[2-4]

Maternal immune response to fetus has an important role as a predisposing factor in spontaneous abortion, and it is often unexplainable. The current hypothesis is the presence of multiple factors which may affect at systemic, and local *in utero*, determine to modulate immune response into decreasing inflammation response of trophoblasts. Factors involved in this process are hormones, particularly progesterone, embryonic hormones, and Natural Killer cells (NK cells).[1,2]

In daily clinical practice, progesterone supplement is often used as a supportive therapy for pregnant women who are diagnosed with threatened abortion. Progesterone has an important role in modulating immune response in early trimester pregnancy through modulating pro-inflammatory and anti-inflammatory cytokines from conceptus, resulting in the continuation or termination of pregnancy.

With the influence of progesterone, lymphocyte releases protein called progesterone-induced blocking factor (PIBF), which mediates the modulation of immune response and the anti-abortive effect of progesterone. This process also gives positive feedback, there by increasing the amount of progesterone receptors in the activated lymphocyte in the placental cells and $CD8^+$ cells. In return the amount of PIBF will increase along with the gestational age from week 6 to 36, and after 41 weeks, PIBF will decrease considerably and induce the parturition. PIBF also inhibits peripheral NK cells activity. The biological effects of PIBF as mentioned above indicates that PIBF has a role in maintaining pregnancy. Therefore, PIBF value in the body liquid can predict the prognosis of pregnancy.[5]

A Cochrane review in 2013 demonstrated that there was no significant difference between progesterone supplementation and placebo in maintaining pregnancy. Nevertheless, Check JH mentioned that although progesterone deficiency occurs in one third of pregnancy, progesterone supplementation to improve PIBF value still has a role in decreasing 33% spontaneous abortion.[6-8]

Considering the impact of abortion in the physical and emotional aspects of the patient's, and to learn further the role of immune-endocrine (PIBF) in affecting the prognosis of pregnancy, we are interested in doing this study.

METHODS

This was a cross-sectional study. Subejcts were pregnant women with gestational age of ≤ 20 weeks at Prof. Dr. R.D. Kandou Hospital, and subcenter hospital in Manado during the period of October 2015 to February 2016. Samples were collected by consecutive sampling. Total population consisted of 32 patients, divided into two groups: 16 with normal pregnancies, and 16 threatened abortions that matched the inclusion and exclusion criteria.

Inclusion criteria were single pregnant women with less than 20 weeks of gestation confirmed by ultrasound. Exclusion criteria were pregnant women with history of recurrent pregnancy loss, those who were currently receiving progesterone therapy, and fetal death or blighted ovum.

Blood samples from subjects were collected, and examined with PIBF ELISA-kit in the Prokita Laboratory Manado.

Data were analyzed using T-test to investigate the significance of PIBF value in the normal early pregnancy and threatened abortion. Data were processed using Statistical Product and Service Solutions (SPSS) 22.0 for Windows.

RESULT

Subject Characteristics

Table 1. Maternal Age Frequency Distribution

		Maternal age (year)			Total
		<20	**20-34**	**≥35**	
Groups	Normal pregnancy	1	10	5	16
	Threatened abortion	1	12	3	16
Total		2	22	8	32

Interpretation: most maternal age of subjects are around 20-34 years old.

Table 2. Educational Status Frequency Distribution

		Educational status				Total
		Elementary	Junior high school	High school	Bachelor	
Groups	Normal pregnancy	2	1	9	4	16
	Threatened abortion	4	6	5	1	16
Total		6	7	14	5	32

Interpretation: most educational status of subjects are at high school.

Table 3. Gravidity Status Frequency Distribution

		Gravidity status		Total
		First	Multi	
Groups	Normal pregnancy	4	12	16
	Threatened abortion	6	10	16
Total		10	22	32

Interpretation: most gravidity status of subjects are multiple.

Table 4. Gestational Age Frequency Distribution

		Gestational age (weeks)				Total
		4-8	9-12	13-16	17-20	
Groups	Normal pregnancy	7	6	3	-	16
	Threatened abortion	1	4	2	9	16
Total		8	10	5	9	32

Interpretation: most gestational age of subjects are around 9-12 weeks.

PIBF Serum Value

Our study showed that PIBF serum value of threatened abortion was lower than normal pregnancy in all groups of gestational age; with details as follow: the first group of 4-8 weeks of gestation (16.114 vs 48.62 ng/ml); in the second group of 9-12 weeks of gestation (11.107 vs 41.909 ng/ml); in the third group of 13-16 weeks of gestation (7.372 vs 54.217 ng/ml); while in the last group of 17-20 weeks of gestation there was no subject in the normal pregnancy group to compare with the threatened abortion (12.151 ng/ml).

An overall calculation of our data showed a significant difference of PIBF serum value in normal pregnant women of ≤ 20 weeks and threatened abortion (47.153 vs 11.540 ng/ml; p value=0.000). Variable coefficient data showed that the variation of PIBF serum in both groups were above 10%, this stated that the PIBF serum were not homogenous.

DISCUSSION

Pregnancy immune-biology

Throughout history of human evolution, humankind has developed a mechanism to protect us from parasites and infections by detecting and destroying foreign organic materials that enter human's body. This mechanism is called the immune system.[9]

Fetus is a product of conception, which is a semi-allograft tissue. However, there is no rejection from the maternal immune system. This is due to the absence of MHC class I nor class II on the placental villous trophoblast, hence not inducing the NK cytotoxic activities.[10,11]

Another factor protecting the fetus is the expression of FasL, a protein membrane type II that is commonly present in the immune-compromised tissue, such as testes, cornea, trophoblast (Houston, and O'Connell, 2004).[12]

Wegmann (1993) mentioned that Th1 (pro-inflammatory) and Th2 (anti-inflammatory) balance is the key of successful pregnancy. Whereas Th2 domination is required to protect feto-placental unit from the adaptive immune response, or non-specific inflammatory response.[10,12]

Pro-inflammatory cytokines are critical in the early process of implantation to induce angiogenesis. Shortly after, it should be replaced by anti-inflammatory cytokines that shift the balance from Th1 to Th2 dominance. Szekeres-Bartho explained that TCD8+ cells in pregnant women expressed progesterone receptors since early pregnancy. Whereas, under progesterone influence, this lymphocyte cells express a mediator protein with molecular weight of 34kDa called PIBF (Progesterone-induced blocking factor).[3,5,10,12,13]

through mediator PIBF. Depletion of progesterone production in pregnancy will initiate parturition. This is the main reason of progesterone supplementation therapy as luteal support to prevent abortion.[15]

PIBF as a research variable

As mentioned above, to have an immune-endocrine effect, progesterone needs protein mediator which is PIBF, which synthesized from the activated lymphocyte of $T_{\gamma/\delta}$ ($CD8^+$).[1-3,5,7] Progesterone supplementation is meant to improve PIBF value through its binding to progesterone receptors, and not to improve progesterone level.[3-7,16]

The mechanisms of PIBF in maintaining pregnancy are through several pathways: by inhibiting peripheral NK cells cytolysis activity, by inducing Th2 cytokines domination and asymmetric antibody production (Ig G).[17-22]

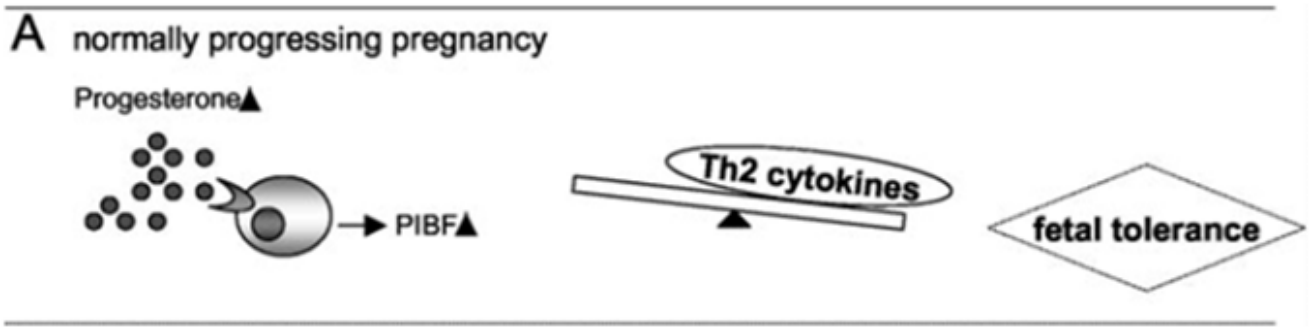

Figure 1. Normally Progessing Pregnancy. A schematic hypothesis of immune-endocrinology in pregnancy. In normal pregnancy, a sufficient concentration of progesterone leads to anti-inflammatory cytokines dominance (Th2), mediated by PIBF expression.[13]

The role of NK cells in pregnancy

NK cells dominated the leucocytes involved in the implantation process in early pregnancy. NK cells are classified as peripheral NK cells and uterine NK cells. In pregnancy, the amount of peripheral NK cells is depleted due to the minimal expression of CD16. This process is affected by the progesterone level.[14] In contrast to peripheral NK cells, uterine NK cells are dominated in pregnancy. These NK cells have a small effect of cytolysis hence needed in pregnancy.[14]

Role of progesterone in pregnancy

Progesterone is the main hormone in maintaining pregnancy. It directly affects immune system or

PIBF expression required adequate, hence PIBF value will predicts the outcome of pregnancy. This is according to the study done by B Polgar, etc which stated that PIBF in the body liquid will reflect the pathological condition of pregnancy.[1,2,5,21,23]

PIBF value can be measured through urine or serum sample, this is due to the fact that the weight molecule is 34kDa, there by small enough to be excreted by renal of PIBF, this could be detected in the urine. PIBF value measurement through urine sample is often done because it is non invasive, yet 24 hours urine sample is needed to reduce the bias.[1-3,5] Nonetheless, in this study we use serum sample, because it is considered the more consistent result due to a random one time sampling.

A study performed by Igor Hudic found difference between PIBF value in serum and urine, nonetheless it has the same predictive value where PIBF value in women with threatened abortion is lower than in normal pregnancy.[2] This is consistent with our study which found that PIBF serum value in threatened abortion is significantly lower than in normal pregnancy.

PIBF Serum Value

Previous studies have shown that PIBF has a role in maintaining pregnancy through immunological mechanism by modulating cytokine Th_1/Th_2 balance, suppressing cytotoxic and cytolysis activity of NK cells, and by increasing asymmetric antibody production.[1-3,5,7,13,19]

Our study showed that there were significant difference between PIBF serum value in normal pregnant women ≤ 20 weeks (47.153 ± 23.830) and threatened abortion (11.540 ± 4.892) (*p value*=0.000). This result is consistent with several studies which suggested that the presence of PIBF could be used as a biomarker of predicting the outcome of pregnancy or detecting pathological condition of pregnancy.[1,2,5]

Our study showed that the coefficient variation is above 10%, meaning that PIBF value has a large variation. This might be due to the factors beyond the researcher control which cause the difference of progesterone production such as inflammatory process, different fetal development, infection factor, uterine stretch, or the maternal/fetal stress.[24] Similar studies performed before also have the same conclusion where mean PIBF in threatened abortion is lower significantly compared to normal pregnancy. Despite having different value, this may due to the different study methods, reagents, processing methods, wave length used in ELISA reader, samples amount, and gestational age used in the subjects.

This study showed that PIBF could be used as a biomarker of pathologic process in pregnancy. However, due to the cross-sectional study design, it is not possible to determine the cut-off point of PIBF value required to maintain pregnancy.

CONCLUSIONS

PIBF serum value of pregnant women ≤ 20 weeks with threatened abortion were significantly lower compared to normal pregnancy. This also showed that PIBF value could be used as a biomarker of a pathologic process in pregnancy. PIBF could also be used as a rational therapy in threatened abortion. Further prospective studies are required to determine the cut-off point of PIBF value necessary to maintain pregnancy.

REFERENCES

1. Formosa M. The paradox of pregnancy: an update on the immunology of early pregnancy. Malta Med J. 2008; 20(2): 10-4.
2. Hudic I, Fatušic Z. Progesterone-induced blocking factor (PIBF) and Th_1/Th_2 cytokine in women with threatened spontaneous abortion. J Perinatal Med. 2009; 37(4): 338-42.
3. Szekeres-Bartho J, Polgar B, Kelemen K, Par G, Szereday L, editors. Progesterone-mediated immuno modulation and anti-abortive effects: the role of the progesterone induced blocking factor. Poster presentation. 10th World Congress on the Menopause; 2002.
4. Kutteh WH. Recurrent pregnancy loss. In: Carr BR, eds. Textbook of reproductive medicine. In: edition, editor. Stamford, Connecticut: Appleton & Lange; 2004.
5. Polgár B, Nagy E, Mikó É, Varga P, Szekeres-Barthó J. Urinary progesterone-induced blocking factor concentration is related to pregnancy outcome. Biol Reprod. 2004; 71(5): 1699-705.
6. Haas DM, Ramsey PS. Progestogen for preventing miscarriage. Cochrane Database Syst Rev. 2013: 10.
7. Kalinka J, Szekeres-Bartho J. The Impact of Dydrogesterone Supplementation on Hormonal Profile and Progesterone-induced Blocking Factor Concentrations in Women with Threatened Abortion. Am J Reprod Immunol. 2005; 53(4): 166-71.
8. JH C. Check JH. Debate: should progesterone supplements be used? In: Carp HJA, editor. Recurrent Pregnancy Loss Causes, Controversies and Treatment. 2: CRC Press; 2015: 123-30.
9. Bainbridge DR. Evolution of mammalian pregnancy in the presence of the maternal immune system. Reviews Reprod. 2000; 5(2): 67-74.
10. Van Nieuwenhoven ALV, Heineman MJ, Faas MM. The immunology of successful pregnancy. Hum Reprod Update. 2003; 9(4): 347-57.
11. Bainbridge DR. Evolution of mammalian pregnancy in the presence of the maternal immune system. Reviews Reprod. 2000; 5(2): 67-74.
12. Schjenken JE, Paul JW, Tolosa JM, Smith R, Clifton VL. Mechanisms of maternal immune tolerance during pregnancy: INTECH Open Access Publisher; 2012.
13. Blois SM, Joachim R, Kandil J, Margni R, Tometten M, Klapp BF, et al. Depletion of $CD8^+$ cells abolishes the pregnancy protective effect of progesterone substitution with dydrogesterone in mice by altering the Th_1/Th_2 cytokine profile. J Immunol. 2004; 172(10): 5893-9.
14. Dosiou C, Giudice LC. Natural killer cells in pregnancy and recurrent pregnancy loss: endocrine and immunologic perspectives. Endocrine reviews. 2005; 26(1): 44-62.

15. Stjernholm YV. Progesterone in Human Pregnancy and Parturition: INTECH Open Access Publisher; 2012.
16. Abadi A, Baziad A, Hestiantoro A. The benefits of progesterone therapy in imminent abortion. Med J Indones. 2005; 14(4): 258-62.
17. Kozma N, Halasz M, Polgar B, Poehlmann TG, Markert UR, Palkovics T, et al. Progesterone-induced blocking factor activates STAT6 via binding to a novel IL-4 receptor. J Immunol. 2006; 176(2): 819-26.
18. Menzies FM, Henriquez FL. Immuno modulation by the female sex hormones. Open Infect Dis J. 2009; 3: 61-72.
19. Margni RA, Gentile MT, Miranda S. Asymmetric antibodies: the antigen protective arm in a Th_2-regulated immune response. Inmunol. 2002; 21(1): 29-35.
20. Speroff L, Fritz MA. Recurrent early pregnancy loss. Clin Gynecol Endocrinol Infertil. 2005: 1069-101.
21. Griebel CP, Halvorsen J, Golemon TB, Day AA. Management of spontaneous abortion. Am Fam Physician. 2005; 72(7): 1243-50.
22. Ford HB, Schust DJ. Recurrent pregnancy loss: etiology, diagnosis, and therapy. Reviews in Obstet Gynecol. 2009; 2(2): 76.
23. Salomon LJ, Rozenberg P, Szekeres-Bartho J, Malagrida L, Giudicelli Y, Ville Y. Changes in progesterone-induced-blocking-factor expression rates following mifepristone administration in termination of pregnancy at 5 to 8 weeks. J Maternal-Fetal Neonatal Med. 2005; 17(5): 353-6.
24. Mesiano S. The endocrinology of human pregnancy and fetoplacental neuroendocrine development. Yen and Jaffe's reproductive endocrinology 6th ed Philadelphia: Saunders Elsevier. 2009: 104.