Association of anticardiolipin IgM antibody with preeclampsia

Sharmin Khanam, Parveen Fatima, Begum Nasrin and Md. Mozammel Hoque

Abstract

The objective of this study was to evaluate association of anticardiolipin IgM antibody with preeclampsia among the 70 pregnant women having preeclampsia from March 2016 to February 2017. Equal number of age matched normotensive healthy pregnant women was selected as control. Demographic, obstetric and relevant laboratory data were collected and compared between the two groups. The level of anticardiolipin IgM antibody was significantly higher in preeclamptic women than in normal pregnant (mean 8.7 vs 5.8 U/mL, p<0.001). Even in categorical distribution when the value was considered as positive by assessing a cut-off, cases had three times more chance to have positive level of anticardiolipin IgM antibody than controls (OR=3.3, 95% CI=1.1-9.6, p=0.046). Pearson's correlation test revealed that both systolic and diastolic blood pressure had a positive correlation with the level of anticardiolipin IgM antibody (p<0.05). In conclusion, this study shows that anticardiolipin IgM antibody was associated with preeclampsia. The level of this anticardiolipin IgM antibody is directly proportional to the severity of preeclampsia and both systolic and diastolic blood pressure.

Introduction

Preeclampsia constitutes a multifactorial disease that involves genetic, metabolic, immunological and environmental factors and includes the presence of endothelial dysfunction that develops before the onset of clinical symptoms.[1][-6] Despite of availability of modern technology and over a decade of intense study, the cause of preeclampsia still not well-explained and remains a major cause of maternal and perinatal mortality and morbidity.[7-8] Antiphospholipid antibodies are a heterogeneous group of immunoglobulin (IgG, IgM, IgA) which have great affinity for several combinations of phospholipids, phospholipid-binding proteins or both. Platelets and endothelial cells are their major targets.[9-10] It can damage phospholipids that present in all endothelial cell surfaces. Damaged endothelial cells do not function normally which can cause thrombotic complications.[11-13] A relationship between antiphospholipid antibodies and preeclampsia was first suspected because of the high rate of preeclampsia among women with antiphospholipid syndrome.[14-15] Deregulated immunity at the feto-maternal interface in the beginning of pregnancy leads to the poor placentation and subsequently to the onset of clinical symptoms of preeclampsia.[16]

The most commonly used tests to detect antiphospholipid syndrome include lupus anticoagulant, anticardiolipin antibodies, and anti-β2-glycoprotein 1 antibodies. Among them, detection of anticardiolipin IgM antibody is popularly used to detect antiphospholipid syndrome. The relationship between anticardiolipin antibody and preeclampsia is still controversial. The aim of this study was to detect the levels of anticardiolipin IgM antibody in serum of pregnant women having preeclampsia and to evaluate its relation with preeclampsia by comparing the level with the normal pregnant women.

Materials and Methods

This was conducted on 70 pregnant women clinically diagnosed as preeclampsia during their second half of pregnancy (20 to 40 weeks) from March 2016 to February 2017. Similar number of age matched apparently healthy normotensive pregnant women were selected as control. However, pregnant women aged <20 or >38 years, multiple pregnancy, chronic hypertension, gestational hypertension, liver disease, cardiac disease, or connective tissue disorder were excluded from this study. Women having sign of active infection, previous history of repeated pregnancy loss, previous history of preeclampsia, or any thrombotic disorder were excluded. Participation of this study was totally voluntary and written informed consent was taken from all participants

Table I

Socio-demographic and clinical characteristics

Variable	Pre-eclampsia (n=70)	Normal pregnant (n=70)	p
Age (year)	26.5 ± 5.4	26.9 ± 5.2	0.703
Body mass index (kg/m²)	26.8 ± 1.7	25.7 ± 2.8	0.009
Parity: Nullipara	41	32	0.175
Gestational age (week)	33.0 ± 2.9	32.1 ± 4.0	0.150
Anticardiolipin IgM antibody (U/mL)	8.7 ± 3.9	5.8 ± 2.6	<0.001
Data are expressed as mean ± SD			

Table II

Anticardiolipin IgM antibody concentration

Anticardiolipin IgM antibody	Pre-eclampsia (n=70)	Normal pregnant (n=70)	OR	95% CI	p value
Positive (≥10 U/mL)	14	5	3.25	1.10-9.58	0.048
Negative (<10 U/mL	56	65	0.31	0.10-0.91	0.048

micro-centrifuged tube at -20°C until the analysis was done. Reference value of anticardiolipin IgM antibody level was considered negative when it was <10 Unit/mL and positive when ≥10 Unit/mL. Anticardiolipin IgM antibody (IgM) was determined by Immunometric Enzyme Immunoassay

Statistical analysis

All data were analyzed by using statistical package for social sciences (SPSS), windows (version 20.0; USA) and Epi info (version 1.0.3, USD, Stone Mountain, GA). Mean comparison between two groups were done by student's t-test. To understand the relationship between two variables Pearson's correlation coefficient (r) was done. Probability of <0.05 was considered as statistically significant. Strength of association was determined by estimating odds ratios (OR) and their 95% confidence intervals (CI).

Results

The mean (± SD) age of the study subject was 26.5 ± 5.4 years among the cases and 26.9 ± 5.2 in the controls. Though the nulliparous were predominant (59%) among the cases, it was comparable between the two groups (p>0.05). However, mean body mass index (BMI) was significantly higher among the cases compare to controls (Table I). The mean level of anticardiolipin IgM antibody was significantly higher in preeclamptic group compared to control (8.7 vs 5.8 U/mL, p<0.001). The values were categorized according to the cut off 10 unit/mL as positive, and it has been revealed that in preeclampsia about three times higher chance of having positive anticardiolipin IgM antibody level compared to control (OR=3.25, 95% CI=1.1-9.6, p=0.048) (Table II). Among the 70 preeclampsia cases, 56% had mild preeclampsia and 44% had severe preeclampsia. The mean anticardiolipin IgM antibody was higher in severe preeclamptic group compared to mild preeclampsia group (7.5 vs 10.4 U/mL, p<0.001). Pearson's correlation coefficient test was also done to evaluate the correlation between anticardiolipin IgM antibody level and the systolic and diastolic blood pressure. It was observed that there was positive correlation between the levels of anticardiolipin IgM antibody and degree of rise of both systolic and diastolic blood pressure (r=0.367; p=0.002 in systolic blood pressure and r=0.241; p=0.044 in diastolic blood pressure) (Figure 1).

before enrollment. Following the inclusion and exclusion criteria, samples were collected by purposive sampling. Pre-tested case record form was used to capture relevant demographic, clinical and laboratory data. Physical examination, anthropometric measurements (height, weight) and obstetric examination were performed as per the hospital's standard practice and recorded on the case record form. Blood pressue was measured following standard procedure after 10 minutes' rest. Korotkoff phase-I (first beat heard) and phase-V (disappearance of sound) were used for systolic and diastolic blood pressure with sphygmomanometer. In this study, proteinuria was defined as the urinary excretion of ≥1+ dipstick reading. The participants with blood pressure ≥140/90 mm Hg on two occasions were evaluated for the presence of urinary protein by dipstick method to establish the diagnosis of preeclampsia. When proteinuria found ≥1+ in collected urine sample then the diagnosis of preeclampsia was established and was selected as case. The participants who were found normotensive and had no proteinuria were selected as control.

After selecting cases and controls, with all aseptic precaution 5 mL antecubital vein venous blood sample was collected from each subject for measurement of anticardiolipin IgM antibody. Blood was immediately transferred into a clean dry test tube. Then the sample was sent to laboratory with different code and centrifugation was done at 3,000 rpm for 10 min. The separated serum was carefully drawn by micropipette and was stored in

Discussion

In this study, it was observed that 20% of pregnant women with preeclamsia was associated with a positive level of anticardiolipin antibody, whereas it was 7% among the normal pregnancy. The mean
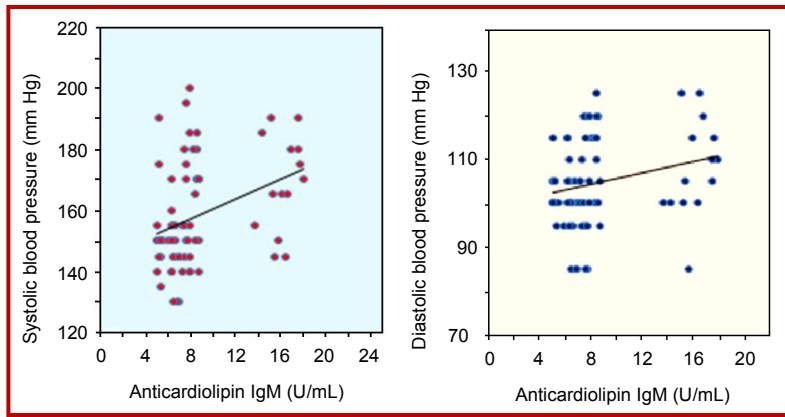
Figure 1: Scatter diagram showing a significant positive correlation of anticardi-olipin IgM antibody with systolic (r=0.367; p=0.002) and diastolic (r=0.241; p= 0.044) blood pressure in preeclampsia group

serum level of anticardiolipin antibody was also significantly higher (8.8 vs 5.8 U/mL, p<0.001) among the cases compared to control. The role of anticardiolipin antibody as a risk factor for preeclampsia in women with no evidence of autoimmune diseases has been the focus of many studies for the past decades, mainly in cohort and case-control studies.[17-18]

Although there are not much previous evidences that suggested a positive relation of anti-cardiolipin antibody with preeclampsia, some earlier studies showed the similar relation which is consistent with these findings.[18-20] Pereira et al. (2015) showed that anticardiolipin IgM antibody was detected in 13.9% women with preeclampsia and the level was significantly higher in preeclampsia group compar-ed to control.[19] Ferrer-Oliveras et al. (2012) showed similar findings. There was significant differences of anticardiolipin IgM antibody between the pre-eclampsia group and the controls (8.1 vs 1.2%, p=0.041).[21] A study by Kestlerova et al. (2012) found there was a significantly higher incidence of anticardiolipin IgM antibody in the preeclampsia group, as compared to the controls (10 vs 4%, p<0.05).[22] Study results done by Briones-Garduno et al. (2003) recommended that immunologic mechanism that induces synthesis of anticardiolipin IgM antibody during the acute state of disease, accounting for vascular changes and prothombotic state responsible for maternal and neonatal compli-cations.[17]

Allen et al. (1996) found elevated levels of IgG or IgM antibodies to cardiolipin in 11% of women diagnosed with preeclampsia in the third trimester compared to only 3% positive in controls and suggested that antiphospholipid antibodies may play a pathogenic role in some women with pre-eclampsia.[23] Branch et al. (1989) suggest that anti-phospholipid antibodies are found in a substantial proportion of cases of early-onset severe pre-eclampsia and have important clinical implications.

They suggest that patient with early onset severe preeclampsia be screened for antiphospholipid antibodies; if positive these women should be considered for prophylactic anticoagulation thera-py.[24] Yasuda et al. (1995) reported that rates of preeclampsia (11.7%) in the anticardiolipin antibody-positive group were significantly higher than those in the negative group (p<0.001; RR 6.2, 95% CI 2.4-16.0) and concluded that anticardiolipin antibody is associated with adverse pregnancy outcome like preeclampsia.[25] However, systematic review and meta-analysis revealed that there was heterogeneity among the studies; out of nine studies, four studies demonstrated that anticardiolipin antibody increa-sed the chance of preeclampsia but five studies reported there was no statistically significant associations.[26] This heterogeneity might occur due to the difference in their case selection, clinical, demographic or epidemiological variances. In this study, there was a strong association of the severity of preeclampsia with anticardiolipin IgM antibody level. Severe preeclampsia groups showed signifi-cant higher level of antibody than mild pre-eclampsia group. Pearson's correlation test revealed that both systolic and diastolic blood pressure had a positive correlation with the level of anticardiolipin IgM antibody (p<0.05). Previous studies also reported such correlation of anticardiolipin anti-body level in severe preeclampsia than mild pre-eclampsia.[20, 25] There are different clinical and laboratory tools to assess the severity of pre-eclampsia. Previously proteinurea was thought to be an indicator of severity of preeclampsia but recently American College of Obstetricians and Gynecology has recommended proteinurea as diagnostic tool only, not as a tool for severity of preeclampsia.[27] So, in this study proteinureain was not considered as the severity scale of preeclampsia to see the relation anticardiolipin antibody with severity of preeclampsia. Rather here blood pressure was considered as the most common indicator of severity of preeclampsia. The study result has indicated that both systolic and diastolic pressure has a positive correlation with level of anticardiolipin antibody. Pearson's correlation test reveled level of anticardiolipin IgM antibody increased with per unit change of systolic or diastolic pressure.

Conclusion

Anticardiolipin IgM antibody was raised among the patients suffering from preeclampsia. There was significant positive correlation between anticardio-lipin IgM antibody and severity of preeclampsia.

Ethical Issue

This study protocol was approved by the Institutional

Review Board, Bangabandhu Sheikh Mujib Medical University.

References

1. Sibai B, Dekker G, Kupferminc M. Pre-eclampsia. Lancet 2005; 365: 785-99.

2. Moffett A, Hiby SE. How does the maternal immune system contribute to the development of pre-eclampsia? Placenta 2007; 28 SupplA: S51-S66.

3. Duckitt K, Harrington D. Risk factors for pre-eclampsia at antenatal booking: Systematic review of controlled studies. BMJ. 2005; 330: 565.

4. Hiby SE, Walker JJ, O'Shaughnessy KM, Redman CW, Carrington M, Trowsdale J, Moffett A. Combi-nations of maternal KIR and fetal HLA-C genes influence the risk of preeclampsia and reproductive success. J Exp Med. 2004; 200: 957-65.

5. Johnson MP, Fitzpatrick E, Dyer TD, Jowett JB, Brennecke SP, Blangero J, Moses EK. Identification of two novel quantitative trait loci for pre-eclampsia susceptibility on chromosomes 5q and 13q using a variance components-based linkage approach. Mol Human Reprod. 2006; 13: 61-67.

6. Kaufmann P, Black S, Huppertz B. Endovascular trophoblast invasion: Implications for the patho-genesis of intrauterine growth retardation and preeclampsia. Biol Reprod. 2003; 69: 1-7.

7. Redman CW, Sargent IL. Latest advances in under-standing preeclampsia. Science 2005; 308: 1592-94.

8. Solomon CG, Seely EW. Preeclampsia: Searching for the cause. N Engl J Med. 2004; 350: 641-42.

9. Levine JS, Branch DW, Rauch J. The antiphospho-lipid syndrome. N Engl J Med. 2002; 346: 752-63.

10. Petri M. Epidemiology of the antiphospholipid antibody syndrome. J Autoimmun. 2000; 15: 145-51.

11. Dreyfus M, Hedelin G, Kutnahorsky R, Lehmann M, Viville B, Langer B, Fleury A, M'Barek M, Treisser A, Wiesel ML, Pasquali JL. Antiphospho-lipid antibodies and preeclampsia: A case-control study. Obstet Gynecol. 2001; 97: 29-34.

12. Infante-Rivard C, David M, Gauthier R, Rivard GE. Lupus anticoagulants, anticardiolipin antibodies, and fetal loss: A case-control study. N Engl J Med. 1991; 325: 1063-66.

13. Yamamoto T, Takahashi Y, Geshi Y, Sasamori Y, Mori H. Anti-phospholipid antibodies in pre-eclampsia and their binding ability for placental villous lipid fractions. J Obstet Gynaecol Res. 1996; 22: 275-83.

14. Heilmann L, von Tempelhoff GF, Pollow K. Anti-phospholipid syndrome in obstetrics. Clin Appl Thromb Hemost. 2003; 9: 143-50.

15. Branch DW, Silver RM, Blackwell JL, Reading JC, Scott JR. Outcome of treated pregnancies in women with antiphospholipid syndrome: An update of the Utah experience. Obstet Gynecol. 1992; 80: 614-20.

16. Redman CWG, Sargent IL. Review article: Immuno -logy of pre-eclampsia. Am J Reprod Immunol. 2010; 63: 534-43.

17. Briones-Garduño J, Díaz dL-PM, Barrios-Prieto E, Salazar-Exaire J. IgM antiphospholipical anti-bodies in preeclampsia-eclampsia. Cirugia y Cirujanos. 2003; 71: 449-54.

18. Branch DW, Porter TF, Rittenhouse L, Caritis S, Sibai B, Hogg B, Lindheimer MD, Klebanoff M, MacPherson C, VanDorsten JP, Landon M. Antiphospholipid antibodies in women at risk for preeclampsia. Am J Obstet Gynecol. 2001; 184: 825-34.

19. Pereira KN, Beck ST, Konopka CK, Segala Z, Moresco RN, Silva JE. Level of reactivity of IgM anticardiolipin antibodies in preeclampsia: A likely early serologic marker. J Brasileiro de Patologia e Medicina Lab. 2015; 51: 13-16.

20. Mello G, Parretti E, Marozio L, Pizzi C, Lojacono A, Frusca T, Facchinetti F, Benedetto C. Thrombo-philia is significantly associated with severe pre-eclampsia: Results of a large-scale, case-controlled study. Hypertension 2005; 46: 1270-74.

21. Ferrer-Oliveras R, Llurba E, Cabero-Roura L, Alijotas-Reig J. Prevalence and clinical usefulness of antiphospholipid and anticofactor antibodies in different Spanish preeclampsia subsets. Lupus 2012; 21: 257-63.

22. Kestlerova A, Feyereisl J, Frisova V, Měchurová A, Šůla K, Zima T, Běláček J, Madar J. Immunological and biochemical markers in preeclampsia. J Reprod Immunol. 2012; 96: 90-94.

23. Allen JY, Tapia-Santiago C, Kutteh WH. Anti-phospholipid antibodies in patients with pre-eclampsia. Am J Reprod Immunol. 1996; 36: 81-85.

24. Branch DW, Andres R, Digre KB, Rote NS, Scott JR. The association of antiphospholipid antibodies with severe preeclampsia. Obstet Gynecol. 1989; 73: 541-45.

25. Yasuda M, Takakuwa K, Tokunaga A, Tanaka K. Prospective studies of the association between anticardiolipin antibody and outcome of preg-nancy. Obstet Gynecol. 1995; 86: 555-59.

26. do Prado AD, Piovesan DM, Staub HL, Horta BL. Association of anticardiolipin antibodies with pre-eclampsia: A systematic review and meta-analysis. Obstet Gynecol. 2010; 116: 1433-43.

27. ACOG. Hypertension in pregnancy. American College of Obstetricians and Gynecology, 2013.