Biomedical Research 2018; 29 (3): 530-535 *ISSN 0970-938X*
www.biomedres.info

Relationship of polymorphism of adhesion molecules *VCAM-1* and *ICAM-1* with preeclampsia.

Ya Chen, Ying Zhang, Yunxia Cao*

Department of Obstetrics and Gynecology, the First Affiliated Hospital of Anhui Medical University, Hefei, PR China

Abstract

Objective: To investigate relationship of polymorphism of adhesion molecules *VCAM-1* and *ICAM-1* with preeclampsia.

Method: The present study included 252 patients with PE in our hospital during 2014 February to 2016 December and 200 healthy pregnant women were used as control. Allelic discrimination of the rs5498 polymorphisms of the *ICAM-1* gene and rs3181092 of the *VCAM-1* gene was assessed using the TaqMan assay. Data was analysed using SPSS 18.0.

Results: In PE patients, both ratios of AA and AA+AG genotypes of *VCAM-1* were significantly higher than those in the control group, P<0.05. Comparison of genotypes between PE patients of early-onset and late-onset showed that in late-onset PE patients, ratio of AA genotype in *VCAM-1* was significantly higher than that in the early-onset group, P<0.05. Similarly, ratio of genotype AA in severe PE patients was significantly higher than that in mild PE patients, P<0.05. However, in all comparisons, distribution of rs5498 polymorphism of *ICAM-1* showed no significant difference in different groups.

Conclusion: Rs3181092 polymorphism of *VCAM-1* was associated with occurrence of PE, especially for late-onset and severe PE patients. However whether rs5498 polymorphism of *ICAM-1*was associated with PE needed more investigation.

Keywords: Polymorphism, *VCAM-1*, *ICAM-1*, Preeclampsia.

Accepted on November 15, 2017

Introduction

Preeclampsia (PE), a severe complication of pregnancy and one of the leading causes for mortality and morbidity in mothers and fetuses, is characterized by new onset of proteinuria and hypertension after 20 w of gestation and affects about 2%~10% pregnant women all over the world [1-3].

It is thought that PE has been associated with insufficient trophoblast invasion of maternal spiral arteries, placental ischemia, impaired placental perfusion, activation of thrombosis, and widespread endothelial cell dysfunction [4-6]. However, the causes for these pathophysiological mechanisms are still unclear. It is considered that genetic factors play an important role in etiology of PE with an estimated heritability of 54% for PE [7].

Studies show that Intercellular Adhesion Molecule-1 (*ICAM-1*), a transmembrane glycoprotein which is expressed in endothelial cells and leukocytes in the immune system, is associated with disorders of female reproductive system such as endometriosis, ovarian stimulation syndrome, and preeclampsia [8,9]. The Vascular Cell Adhesion Molecule-1 (*VCAM-1*), an immunoglobulin like transmembrane protein, is also involved in many inflammatory related diseases [10,11]. It has been proven that polymorphisms in the *VCAM-1* promoter can affect the expression of *VCAM-1* [12]. Studies also showed that polymorphisms of *ICAM-1* and *VCAM-1* genes have been implicated as genetic risk factor in several autoimmune diseases [13,14]. However, to our best knowledge, few studies focused on the relationship of polymorphisms of *ICAM-1* and PE, and no study has been reported on polymorphisms of *VCAM-1* in PE patients.

In the present study, we aimed to investigate polymorphism of adhesion molecules *VCAM-1* and *ICAM-1* in preeclampsia and demonstrate relationship of polymorphism of *VCAM-1* and *ICAM-1* with PE. The present study would firstly demonstrate relationship of polymorphism of adhesion molecules *VCAM-1* with preeclampsia, and might provide a new target and better understanding for study and treatment of PE.

Method and Materials

Patients

The present study included 252 patients with PE in our hospital during 2014 February to 2016 December. The diagnosis of PE was confirmed according to clinical findings including increased blood pressure (≥ 140 mmHg systolic or ≥ 90 mmHg diastolic on 2 or more measurements at least 6 h apart) and proteinuria ≥ 0.3 g/24 h or ≥ +1 on a urine dipstick after 20 w of gestation [7]. Patients were further divided into early-onset

and late-onset subgroups according to gestational age of disease onset. Early onset PE was defied as preeclampsia that developed before 34 w of gestation, and late onset PE was defined as which developed at or after 34 w of gestation. Severe PE was defined as severe hypertension (SBP>150 mmHg or DBP>100 mmHg) or severe proteinuria (>2.0 g in a 24 h urine collection or >3+ by dipstick test) [4]. Patients with the following characteristics were excluded: infected patients; patients with chronic hypertension or patients with gestational hypertension but without proteinuria; patients with multiple pregnancies; patients with autoimmune, metabolic, or cardiovascular diseases and patients with other severe systemic diseases. Data of 200 health controls was collected from healthy pregnant women who went to our hospital at the same period. This study was approved by Ethic committee of the First Affiliated Hospital of Anhui Medical University.

Determination of the genotype

Peripheral blood was collected and DNA was extracted as described elsewhere [15]. Allelic discrimination of the rs5498 polymorphisms of the *ICAM-1* gene and rs3181092 of the *VCAM-1* gene was assessed with the ABI StepOne^{TM} Real-Time PCR System (Applied Biosystems, Foster City, CA, USA), and analysed with SDS vers. 3.0 software (Applied Biosystems) using the TaqMan assay [15]. Primer sequences used in the study were as follows: *ICAM-1* rs5498, forward CCATCGGGGAATCAGTGACT, reverse, ATGACTGCGGCTGCTACCA; *VCAM-1* rs3181092, forward GGAGATGTTTCACGAAGTTTGT, reverse, AAGGGTCACATTTTGAGTATCC.

Statistical analysis

The measurement data was expressed by mean ± SD. Independent continuous variables were compared using the t-test and categorical data such as differences in allelic and genotypic frequencies of different groups were compared using the chi square test or Fisher exact test. Hardy-Weinberg equilibrium was determined by chi square test. Cross-products (Odds ratio (OR)) with a 95% confidence interval (95% CI) was also recorded. A P-value was less than 0.05 it was considered to be statistically significant. All analyses were made using SPSS 18.0.

Results

Clinical basic information for all participants

The present study included 252 patients with PE 200 health controls. Data of maternal age, gestation age, delivery weeks, birth weight, blood pressure, proteinuria and family history of hypertension were collected. As shown in Table 1, except for blood pressure, proteinuria and family history of hypertension, other characteristics showed no significant difference between the patients and the control. However in patients of PE, the mean blood pressure, amount of proteinuria and ratio of family history of hypertension were all significantly higher than those of the control group, P>0.05.

Comparison of genotypes between PE patients and the control

As shown in Table 2, genotypes for rs3181092 in *VCAM-1* and rs5498 in *ICAM-1* were analysed in all PE patients and the control group. The distribution of both *VCAM-1* and *ICAM-1* polymorphism genotypes was in Hardy-Weinberg equilibrium for cases and controls. Results showed that in PE patients, both ratios of AA and AA+AG genotypes of *VCAM-1* were significantly higher than those in the control group, P<0.05. Ratio of A was also higher in patients than in the control, however the difference was not significant. On the other hand, distribution of rs5498 polymorphism of *ICAM-1* showed no significant difference between patients and the control group. These results suggested that genotype A of *VCAM-1* rs3181092 might be associated with incidence of PE.

Comparison of genotypes between PE patients of early-onset and late-onset

Comparison of genotypes between PE patients of early-onset and late-onset was then conducted and the results were shown in Table 3. Results showed that in late-onset PE patients, ratio of AA genotype in *VCAM-1* was significantly higher than those in the early-onset group, P<0.05. Ratios of AA+AG and allele A were also higher in late-onset patients than in early-onset patients, however the difference was not significant. And similar to above, distribution of rs5498 polymorphism of *ICAM-1* showed no significant difference between the two groups.

Comparison of genotypes between severe and mild PE patients

Difference of genotypes between severe and mild PE patients was at last analysed. As shown in Table 4, ratio of genotype AA in severe PE patients was significantly higher than that in mild PE patients, P<0.05. The difference of ratios of AA+AG and allele A was not significant. What's more, genotype of *ICAM-1* rs5498 showed no significant difference.

Table 1. Basic clinical information for patients and the control.

	Patients, n=252, (%)	Control, n=200, (%)	P value	OR (95% CI)
Maternal age, year	29.5 ± 5.4	28.3 ± 7.2	0.693	
Gestation age, day	8.1 ± 1.6	8.4 ± 1.8	0.785	

Birth weight, kg	2.6 ± 0.5	2.9 ± 0.2	0.326	
Systolic blood pressure	169.3 ± 21.6	112.8 ± 13.7	0.000	
Diastolic blood pressure	110.4 ± 11.3	74.9 ± 15.4	0.000	
Proteinuria	3.6 ± 0.6	0.3 ± 0.1	0.005	
Delivery mode				
Vaginal delivery	181 (71.8)	139 (69.5)	0.721	1.117 (0.608~2.054)
Caesarean delivery	71 (28.2)	61 (30.5)		
Family history of hypertension	41 (16.3)	3 (1.5)	0.000	12.788 (2.342~69.827)

Table 2. *Comparison of genotypes for PE patients and the control.*

Variable	Patients, n=252, (%)	Control, n=200, (%)	P value	OR (95% CI)
VCAM-1 rs3181092				
Genotype				
GG	25 (9.9)	40 (20.0)		Ref=1
AG	148 (58.7)	112 (56.0)	0.078	2.118 (0.910~4.930)
AA	79 (31.3)	48(24.0)	0.038	2.635 (1.041~6.670)
AG+AA	227 (90.1)	160 (80.0)	0.045	2.275 (1.003~5.161)
Allele				
G	198 (39.3)	192 (48.0)		Ref=1
A	306 (60.7)	208 (52.0)	0.215	1.426 (0.813~2.499)
ICAM-1 rs5498				
AA	109 (43.3)	85 (42.5)		Ref=1
AG	113 (44.8)	91 (45.5)	0.910	0.966 (0.535~1.745)
GG	30 (11.9)	24 (12.0)	0.953	0.973 (0.393~2.410)
AG+GG	143 (56.7)	115 (57.5)	0.909	0.968 (0.553~1.695)
Allele				
A	331 (65.7)	261 (65.3)		Ref=1
G	173 (34.3)	139 (34.7)	1.000	0.982 (0.548~1.760)

Table 3. *Comparison of genotypes between PE patients of early-onset and late-onset.*

Variable	Early-onset, n=79, (%)	Late-onset, n=173, (%)	P value	OR (95% CI)
VCAM-1 rs3181092				
Genotype				
GG	10 (12.7)	15 (8.7)		Ref=1
AG	55 (69.6)	93 (48.0)	0.989	0.993 (0.389~2.537)
AA	14 (17.7)	65 (43.3)	0.012	0.280 (0.101~0.780)
AG+AA	69 (87.3)	158 (91.3)	0.360	0.655 (0.263~1.629)
Allele				
G	75 (47.5)	123 (35.5)		Ref=1

A	83 (52.5)	223 (64.5)	0.085	0.608 (0.345~1.073)
ICAM-1 rs5498				
AA	31 (39.2)	78 (45.1)		Ref=1
AG	42 (53.2)	71 (41.0)	0.183	1.493 (0.827~2.695)
GG	6 (7.6)	24 (13.9)	0.353	0.629 (0.236~1.680)
AG+GG	48 (60.8)	95 (54.9)	0.398	1.274 (0.726~2.236)
Allele				
A	104 (65.8)	227 (65.6)		Ref=1
G	54 (34.2)	119 (34..4)	0.976	0.991 (0.553~1.777)

Table 4. *Comparison of genotypes between severe and mild PE patients.*

Variable	Mild, n=166, (%)	Severe, n=86, (%)	P value	OR (95% CI)
VCAM-1 rs3181092				
Genotype				
GG	20 (12.0)	5 (5.8)		Ref=1
AG	101 (60.8)	47 (54.7)	0.244	0.537 (0.187~1.545)
AA	45 (27.1)	34 (39.5)	0.045	0.332 (0.110~1.001)
AG+AA				
Allele				
G	141 (42.5)	57 (33.1)		Ref=1
A	191 (57.5)	115 (66.9)	0.170	0.669 (0.377~1.190)
ICAM-1 rs5498				
AA	72 (43.4)	37 (43.0)		Ref=1
AG	73 (44.0)	40 (46.5)	0.830	0.938 (0.520~1.691)
GG	21 (12.6)	9 (10.5)	0.713	1.189 (0.473~2.987)
AG+GG	94 (56.6)	49 (57.0)	0.954	0.984 (0.562~1.722)
Allele				
A	217 (65.4)	114 (66.3)		Ref=1
G	115 (34.6)	58 (33.7)	0.893	1.041 (0.580~1.867)

Discussion

As one of the leading causes for mortality and morbidity in mothers and fetuses, Preeclampsia (PE) remains to be a worldwide healthy problem [16]. Though detailed mechanisms for occurrence of PE are still unclear, it is thought that genetic factors are crucial in development of PE. Intercellular Adhesion Molecule-1 (*ICAM-1*) and Vascular Cell Adhesion Molecule-1 (*VCAM-1*) are adhesion molecules which have been proven to be associated with many diseases including gynecological diseases like endometriosis [17] or PE [18]. However to our best knowledge, few studies focused on polymorphism of *VCAM-1* and *ICAM-1* in preeclampsia.

In the present study, for the first time we demonstrated that rs3181092 polymorphism of *VCAM-1* was associated with occurrence of PE, however whether rs5498 polymorphism of *ICAM-1*was associated with PE needed more investigation.

Studies showed that the single nucleotide G/A polymorphism in the sixth exon of the *ICAM-1* gene (K469E, rs5498) could result in an amino acid substitution from glutamic acid (E) to lysine (K) in immunoglobulin-like domain 5 of the *ICAM-1* protein which is important for the activity of the *ICAM-1* protein in many diseases. Lin et al demonstrated that *ICAM-1* single-nucleotide polymorphisms were associated with oral cancer susceptibility and clinicopathologic development [15]. Popovic et al. showed that polymorphism rs5498 of the

ICAM-1 gene affected the progression of carotid atherosclerosis in patients with type 2 diabetes mellitus [19]. Studies also showed that K469E polymorphism of the *ICAM-1* gene was associated with retinopathy in type 2 diabetes [20]. Recently, Tabatabai et al. studied genotype of *ICAM-1* gene K469E polymorphism in 192 PE patients and found that K469E polymorphism was associated with severe preeclampsia; however in comparison of PE patients and the control, K469E polymorphism showed no significant difference [21]. However in the present study, we found that rs5498 polymorphism of *ICAM-1* not only showed no significant difference in patients and the control, but also showed no significant difference in comparison for patients of early-onset and late-onset, as well as comparison for patients of mild and severe PE. This difference with other studies might be due to the different study population and sample size, which needs more clinical evidences to confirm.

On the other hand, we found that rs3181092 polymorphism of *VCAM-1* was associated with occurrence of PE. There are very few studies focusing on this mutant target. A genetic study showed that rs3181092 polymorphism of *VCAM-1* might be associated with risk of chronic lymphatic leukemia [22]. There is also a study demonstrating that rs3181092 in *VCAM-1* was associated with carotid plaque [23]. However up to now, no studies show relationship of *VCAM-1*and preeclampsia, we are the first to demonstrate that AA genotype for rs3181092 polymorphism of *VCAM-1* may be associated with occurrence of PE.

In conclusion, the present demonstrated that rs3181092 polymorphism of *VCAM-1* was associated with occurrence of PE; ratio of AA genotype of rs3181092 of *VCAM-1* was significant higher in PE patients, especially for late-onset and severe PE patients. However whether rs5498 polymorphism of *ICAM-1* was associated with PE needed more investigation. The present study would firstly demonstrate relationship of polymorphism of adhesion molecules *VCAM-1* with preeclampsia, and might provide a new target and better understanding for study and treatment of PE.

References

1. Sibai BDG, Kupferminc M. Pre-eclampsia. Lancet 2005; 365: 785-99.
2. Ronald TB. Williams Obstetrics. Journal of the American Medical Association 2010; 304: 474-475.
3. Chen X GT, Liao Z, Chen S, Xiao J. Foxp3 (−/att) polymorphism contributes to the susceptibility of preeclampsia. Plos One 2013; 8: 59696.
4. Sowmya SRA, Nallari P, Jyothy A, Venkateshwari A. Role of il-6 -174(g/c) promoter polymorphism in the etiology of early-onset preeclampsia. Inflamm Res 2015; 64: 433-439.
5. Warrington JP, George EM, Palei AC, Spradley FT, Granger JP. Recent advances in the understanding of the pathophysiology of preeclampsia. Hypertension 2013; 62: 666-673.
6. Laresgoiti-Servitje E. A leading role for the immune system in the pathophysiology of preeclampsia. J Leuk Biol 2013; 94: 247-257.
7. Wan JP, Wang H, Li CZ, Zhao H, You L, Shi DH. The common single-nucleotide polymorphism rs2681472 is associated with early-onset preeclampsia in Northern Han Chinese women. Reprod Sci 2014; 21: 1423-1427.
8. Kim KH, Park JK, Choi YW, Kim YH, Lee EN, Lee JR. Hexane extract of aged black garlic reduces cell proliferation and attenuates the expression of ICAM-1 and VCAM1 in TNF-alpha-activated human endometrial stromal cells. Int J Mol Med 2013; 32: 67-78.
9. Kwon HSSIS, Hwang HS, Park YW. Intercellular adhesion molecule-1 k469e polymorphism: is it associated with preeclampsia in the Korean population? J Reprod Med 2012; 57: 148-152.
10. Afford SC, Humphreys EH, Reid DT, Russell CL, Banz VM, Oo Y. Vascular cell adhesion molecule 1 expression by biliary epithelium promotes persistence of inflammation by inhibiting effector T-cell apoptosis. Hepatology 2014; 59: 1932-1943.
11. Amran AA, Zakaria Z, Othman F, Das S, Al-Mekhlafi HM, Nordin NA. Changes in the vascular cell adhesion molecule-1, intercellular adhesion molecule-1 and c-reactive protein following administration of aqueous extract of piper sarmentosum on experimental rabbits fed with cholesterol diet. Lip Health Dis 2011; 10: 2.
12. Dadgar PFKM, Noorbakhsh F, Talebi S, Vodjgani M. Investigating the effect of rs3783605 single-nucleotide polymorphism on the activity of vcam-1 promoter in human umbilical vein endohelial cells. Iran J Allergy Asthma Immunol 2015; 1: 179-187.
13. Balkan M, Akbas H, Penbegul N, Rustemoglu A, Yucel I, Yildiz I. Possible lack of association between E469K polymorphism of ICAM-1 and non-obstructive azoospermia in south-east Turkey. Biotechnol Equip 2016; 31: 143-147.
14. Bielinski SJ, Pankow JS, Li N, Hsu FC, Adar SD, Jenny NS. ICAM1 and VCAM1 polymorphisms, coronary artery calcium, and circulating levels of soluble ICAM-1: the multi-ethnic study of atherosclerosis (MESA). Atherosclerosis 2008; 201: 339-344.
15. Lin CW, Chuang CY, Tang CH, Chang JL, Lee LM, Lee WJ. Combined effects of icam-1 single-nucleotide polymorphisms and environmental carcinogens on oral cancer susceptibility and clinicopathologic development. PLoS One 2013; 8: 72940.
16. Jahantigh D, Mousavi M, Forghani F, Javan MR, Movahedinia S, Rezaei M. Association between maternal circulating IL-27 levels and preeclampsia. Cytokine 2017.
17. Schutt AKAKA, Slackdavis JK, Stovall DW. Vcam-1 on peritoneum and α4β1 integrin in endometrium and their implications in endometriosis. Int J Gynecol Pathol Off J Int Soc Gynecol Patholog 2015; 34: 85.
18. Farzadnia MAH, Hasan-Zade M, Rahimi HR. A comparative study of serum level of vascular cell adhesion

molecule-1 (svcam-1), intercellular adhesion molecule-1(icam-1) and high sensitive c-reactive protein (hs-crp) in normal and pre-eclamptic pregnancies. Iran J Bas Med Sci 2013; 16: 689-693.

19. Sarecka-Hujar B, Zak I, Krauze J. Interactions between rs5498 polymorphism in the ICAM1 gene and traditional risk factors influence susceptibility to coronary artery disease. Clin Exp Med 2009; 9: 117-124.
20. Fan WY, Liu NP. Meta-analysis of association between K469E polymorphism of the ICAM-1 gene and retinopathy in type 2 diabetes. Int J Ophthalmol 2015; 8: 603-607.
21. Tabatabai E, Salimi S, Mohammadoo-Khorasani M, Yaghmaei M, Mokhtari M, Farajian Mashhadi F. KE and EE genotypes of ICAM-1 gene K469E polymorphism is associated with severe preeclampsia. Dis Mark 2014; 2014: 124941.
22. Enjuanes A, Benavente Y, Bosch F, Martin-Guerrero I, Colomer D, Perez-Alvarez S. Genetic variants in apoptosis and immunoregulation-related genes are associated with risk of chronic lymphocytic leukemia. Cancer Res 2008; 68: 10178-10186.
23. Gardener H, Beecham A, Cabral D, Yanuck D, Slifer S, Wang L. Carotid plaque and candidate genes related to inflammation and endothelial function in Hispanics from northern Manhattan. Stroke 2011; 42: 889-896.

*Correspondence to

Yunxia Cao

Department of Obstetrics and Gynecology

The First Affiliated Hospital of Anhui Medical University

PR China