Genetics of Pre-eclampsia

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Pre-eclampsia is a major cause of maternal and perinatal mortality and morbidity worldwide, but remains unclear about the underlying disease mechanisms. Pre-eclampsia is currently believed to be a two-stage disease. The first stage involves shallow cytotrophoblast invasion of maternal spiral arteriole, resulting in placental insufficiency. The hypoxic placenta release soluble factors, cytokines, and tropho-blastic debris into maternal circulation, which induce systemic endothelial damage and dysfunction. This cause the second stage of the disease: maternal syndrome. Epidemiological research has consistently demonstrated a familial predisposition to pre-eclampsia. Intensive research efforts have been made to discover susceptibility genes that will inform our understanding of the pathophysiology of pre-eclampsia and that may provide direction for therapeutic or preventative strategies. In this review, we summarize the current understanding of the role of genetic factors in the pathophysiology of pre-eclampsia and explain the molecular approach to search for genetic clues in pre-eclampsia.

Key Words: Pre-eclampsia, Genetics, Susceptibility gene

Introduction

Pre-eclampsia is a pregnancy-specific syndrome that carries a high risk of maternal and perinatal mortality and morbidity. Pre-eclampsia affects 5 to 10% of all pregnancies and is characterized by the onset of hypertension and proteinuria after 20 weeks of gestation. It can progress to HELLP (hemolysis, elevated liver enzymes, and low platelet counts) syndrome and seizures (eclampsia). Despite decades of intense research on the problem, the mechanisms of the disease onset remains unclear.

Despite extensive clinical trials, no therapeutic approaches are currently available to either treat or prevent pre-eclampsia. Anti-hypertensive drugs, cortico-steroids for lung maturation, and/or magnesium sulfate to prevent eclampsia are administered to handle (or prevent the worsening of) symptoms as a temporization strategy to allow safe delivery of a more mature fetus. However, when temporizing management, the maternal risks must be carefully weighed against the possible fetal benefits, as the risk of fatal deterioration of the health of the mother and/or fetus is high. Several pro-phylactic therapies (anti-oxidant vitamins, calcium or folic acid supplementation, aspirin) have so far not proved efficacious at preventing pre-eclampsia in healthy, nulliparous subjects, although these therapies have shown some benefits in groups[1-4]. As a conse-quence, the sole, albeit radical, way to resolve pre-eclampsia is to remove the placenta; in the case of prematurity, this results in delivery of a preterm baby.

Therefore, pre-eclampsia, with or without intrauterine growth retardation (IUGR), remains a major cause of maternal and neonatal mortality and morbidity worldwide.

A number of epidemiological studies have confirmed that preeclampsia has a maternal genetic component. Daughters of pre-eclamptic women have a higher chance of themselves developing pre-eclampsia[5]. Phenotyping patients with pre-eclampsia are vital for any genetic study. Furthermore, previous studies have provided compelling evidence that nulliparity, a family history of pre-eclampsia (sister/mother who suffered pre-eclampsia), a personal history of a previous pregnancy with pre-eclampsia, an increase in the trophoblastic mass (multiple pregnancy, molar pregnancy), paternity changes between pregnancies, age over 40 years, obesity, and some maternal chronic conditions such as diabetes, chronic hypertension, renal disease, autoimmune disease, and antiphospholipid syndrome are risk factors for pre-eclampsia[6, 7].

It has been recognized for many years that pre-eclampsia has a familial component, and the identification of susceptibility genes is one of a number of strategies designed to elucidate the underlying pathogenetic mechanisms of this condition. The objective of this review is to summarize our understanding of what role genetic factors play in the pathophysiology of pre-eclampsia and to describe the molecular approach to search for genetic clues in pre-eclampsia.

Diagnosis

Pre-eclampsia is usually diagnosed by the presence of hypertension associated with proteinuria. Hypertension is defined as a blood pressure of at least 140 mm Hg (systolic) or at least 90 mm Hg (diastolic) on at least two occasions and at least 4-6 hrs apart after the 20 weeks of gestation in women known to be normotensive beforehand[8-10]. Blood-pressure recordings to establish the diagnosis should be no more than 7 days apart[8, 9, 11]. Hypertension is regarded as severe if there are sustained rises in blood pressure to at least 160 mm Hg (systolic), at least 110 mm Hg (diastolic), or both[8, 9, 11, 12]. Proteinuria is defined as excretion of 300 mg or more of protein every 24 hrs. If 24 hrs urine samples are not available, proteinuria is defined as a protein concentration of 300 mg/L or more ($\geq$1+ on dipstick) in at least two random urine samples taken at least 4-6 hrs apart[8, 9]. The urine dipstick measurements used to establish proteinuria should be performed no more than 7 days apart[8, 9, 11].

Dependent on the amount of systemic involvement, several other symptoms, such as edema, disturbance of hemostasis, renal or liver failure, and HELLP syndrome also complicate the clinical picture. Pre-eclampsia can be early onset (pre-eclampsia starting before 34 weeks of gestation) or late onset (pre-eclampsia starting after 34 weeks of gestation), can show mild or severe symptoms (systolic blood pressure $\geq$160 mmHg or diastolic blood pressure $\geq$110 mmHg, proteinuria >5 g/24 hrs, oliguria, neurological symptoms, other clinical symptoms such as deranged liver function, thrombocytopenia <100,000 mm^3, HELLP syndrome), and can evolve into eclampsia in the most severe cases (Table 1)[13]. In addition, it can manifest as a maternal disorder only, with normal fetal growth, or intrauterine growth restric-

Table 1. Indicators of Severity of Pre-eclampsia[13]

Abnormality	Nonsevere	Severe
Diastolic blood pressure	<110 mmHg	$\geq$110 mmHg
Systolic blood pressure	<160 mmHg	$\geq$160 mmHg
Proteinuria	$\leq$2+	$\geq$3+
Headache	Absent	Present
Visual disturbances	Absent	Present
Upper abdominal pain	Absent	Present
Oliguria	Absent	Present
Convulsion (eclampsia)	Absent	Present
Serum creatinine	Normal	Elevated
Thrombocytopenia	Absent	Present
Serum transaminase elevation	Minimal	Marked
Fetal-growth restriction	Absent	Obvious
Pulmonary edema	Absent	Present

tion of the fetus (IUGR) or sudden fetal distress may occur.

The traditional criteria used to confirm a diagnosis of pre-eclampsia (new onset of both hypertension and pro-teinuria after 20 weeks' gestation) are applicable to most healthy, nulliparous women. However, the criteria men-tioned so far are not reliable in women who have either hypertension or proteinuria before 20 weeks' gestation, especially those receiving antihypertensive drugs[10, 14, 15]. Because of the physiological changes leading to raised maternal blood pressure and increased protein excretion with advanced gestation in such women, more stringent criteria should be used to diagnose pre-eclampsia in those with microvascular disease[10, 14, 15]. Consequently, markers to predict and methods to prevent pre-eclampsia in these women are probably different from those in healthy nulliparous women.

Pathophysiology

The precise origin of pre-eclampsia remains elusive, but it is believed to be multifactorial. A certainty is the central role played by the placenta[16, 17]. A long standing hy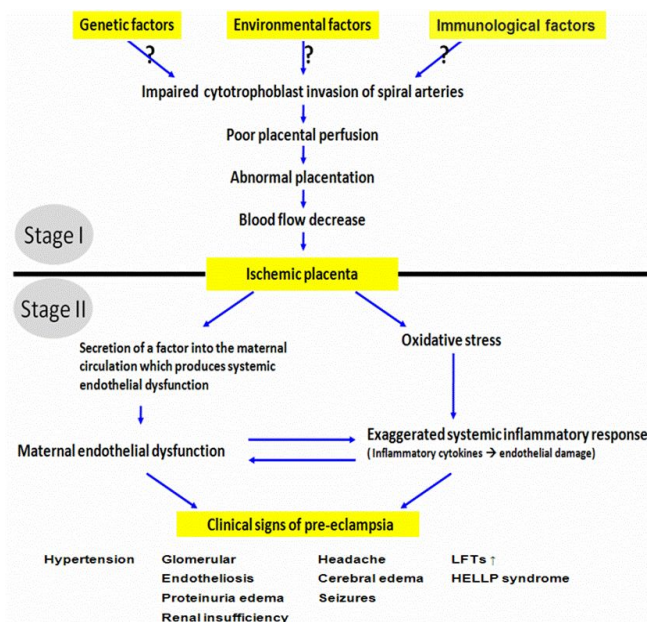pothesis has been that pre-eclampsia develops as a consequence of some kind of immune maladaptation between the mother and the fetus during the very first weeks of pregnancy, leading to the following two-stage progression (Fig. 1)[18]. In the first asymptomatic stage, local aberrant feto-maternal immune interactions within the uterine wall lead to impaired tissue and arterial invasion by trophoblast cells (Fig. 2)[19]. This results in failed transformation of the uterine spiral arteries and subsequently poor placental perfusion. Chronic hypoxia or alternate periods of hypoxia/re-oxygenation within the intervillous space are expected to trigger tissue oxidative stress and increase placental apoptosis and
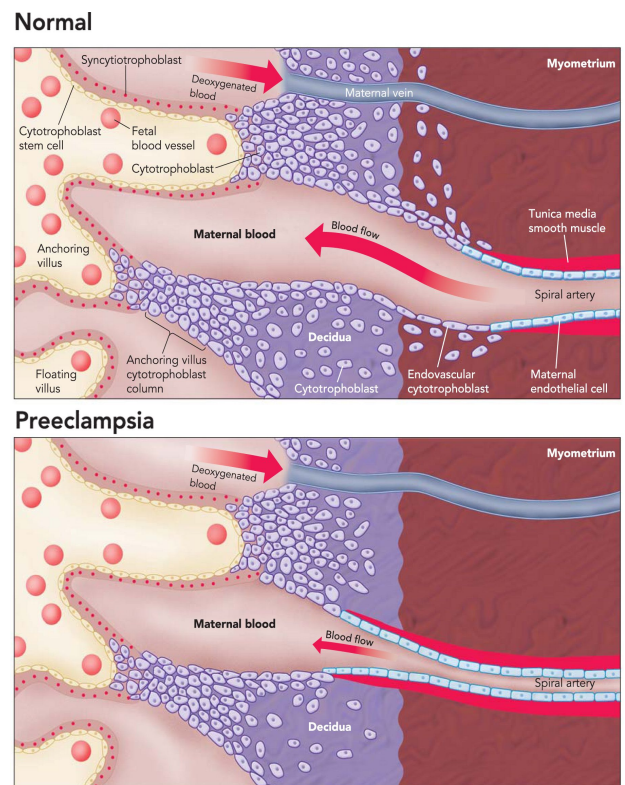

Fig. 2. Abnormal placentation in pre-eclampsia[19]. In nor-mal placental development, invasive cytotrophoblasts of fetal origin invade the maternal spiral arteries, transforming them from small-caliber resistance vessels to high-caliber capacitance vessels capable of providing placental per-fusion adequate to sustain the growing fetus. During the process of vascular invasion, the cytotrophoblasts differen-tiate from an epithelial phenotype to an endothelial pheno-type, a process referred to as "pseudovasculogenesis" or "vascular mimicry" (top). In preeclampsia, cytotrophoblasts fail to adopt an invasive endothelial phenotype. Instead, invasion of the spiral arteries is shallow, and they remain small caliber, resistance vessels (bottom).

Fig. 1. Pathophysiological mechanisms in pre-eclampsia[18].

necrosis[20, 21]. In the second stage, the clinical disorder arises, when the maternal vascular and immune systems can no longer handle the high levels of shedding of placentally produced debris and the aberrant expression of pro-inflammatory, anti-angiogenic and angiogenic factors, leading to systemic endothelial cell dysfunction and an exaggerated inflammatory response[22-24]. Recently, this hypothesis has been challenged[25]. It was proposed instead that an intrinsic failure of trophoblast differentiation at different time points of ontogeny may lead to either a mild disorder with late-onset appearance, or IUGR, with or without maternal symptoms. However, the origin of pre-eclampsia might not be restricted to an alteration in trophoblast differentiation, but may also in some cases depend on underlying maternal constitutional factors such as genetic factors, obesity, dysfunctional maternal clearance, or a dysfunctional maternal inflammatory system[26].

Candidate genes

Various candidate gene studies have been performed to identify associations between pre-eclampsia and genetic polymorphisms in specific genes between cases and controls. Candidate genes are genes with documented biological actions in pathways known to be active in pre-eclampsia that are polymorphic. Single nucleotide polymorphisms (SNPs) are the markers most commonly used to determine genetic associations. Recent initiatives such as the HapMap project (http://www.hapmap.org/) and the establishment of the dbSNP database (http://www.ncbi. nlm.nih.gov/projects/SNP/) have resulted in the identification of multiple SNPs in all human genes.

In association studies, the frequency of alleles and genotypes (homozygous and heterozygous) in the disease group and a healthy population are compared. The most advanced type of association study is a case-control study, where subjects are matched for age and ethnicity. Association studies involving unrelated subjects allow the detection of common variants or common alleles of small effect. In these types of studies, it is also possible to determine those genes that have a weak influence on disease risk. Currently, more than 60 candidate genes related to pre-eclampsia development have been identified, and several groups of candidate genes have been distinguished based on the pathophysiology of pre-eclampsia. The genes that have been most frequently investigated are those involved in blood pressure regulation (angiotensinogen, angiotensin-converting enzyme, and angiotensin receptors), inherited thrombophilias (coagulation factor V Leiden variant, prothrombin, and methylene tetrahydrofolate reductase), vasodilatation regulating genes (endothelial nitric oxide synthase, eNOS), and the gene encoding the cytokine tumor necrosis factor alpha (TNF α) (Table 2)[27-69]. However, our studies based on examinations in Korean population have suggested that the contribution of genetic polymorphism of *β-adrenoceptor*, *EDN1, ICAM-1, INHA, STOX1,* and *TNF* genes to the occurrence of pre-eclampsia is few. It seems that pre-eclampsia is a disease with polygenic inheritance patterns, influenced by environmental factors, gene-gene and gene-environmental interactions[70-75].

It must be conceded that over a decade of molecular genetic research has failed to identify a single universally accepted susceptibility gene for pre-eclampsia. There has been a lack of reproducibility of results, as has been the experience with many complex disorders. Possible explanations have been discussed widely[76], and studies of pre-eclampsia share some of these generic problems. Progress in this field will require attention to both study design and the choice of candidate genes.

Meta-analyses

It has been argued that the stringent criteria used in association studies may be impractical and that more insight might be gained by combining the results of

**Table 2.** Candidate Gene Studies in Pre-eclampsia

Gene name	Gene symbol	References	Gene name	Gene symbol	References
Thrombophilia			Interleukin 1β	IL1B	48
Factor V Leiden	F5	27-29	Interleukin 1 receptor antagonist	IL1RN	49
Prothrombin 20210	F2	29,30	Interlukin 10	IL10	50
Methylene tetrahydofolate reductase	MTHFR	27-29,31,32	T-lymphocyte-associated protein 4	CTLA4	51
Cystathione β-synthase	CBS	28	TNF-receptor supergamilt member 6	FAS	52
Plasminogen activator inhibitor 1	SERPINE1	29,32	Oxidative stress		
β-Fibrinoten	FGB	33	Microsomal epoxide hydrolase	EPHX1	53
Platelet glycoprotein IIIa	ITGB3	30	Glutathione S-transferase pi	GSTP1	41,54
Thrombomodulin	THBD	34	Glutathione S-transferase mu 1	GSTM1	54,55
Factor VII	F7	33	Glutathione S-transferase theta I	GSTT1	54,55
Platelet collafen receptor α2β1	ITGA2	29	Myeloperoxidase	MPO	55
Factor XIII A-subunit	F13A1	35	Manganese superoxidase dismutase	SOD2	55
Haemodtnamics			Cytochrome IAI	CYPIAI	54,55
Angiotensinoten	AGT	32,36	Haptoglobin	HP	56
Renin	REN	37	p22^{phox}	CYBA	57
Angiotesin-converting enzyme	ACE	32,36	Lipid metabolism		
AT1 receptor	AGTR1	32,36	Lipoprotein lipase	LPL	58
AT2 receptor	AGTR2	38	Apolipoptrotein E	APOE	59
Epithelial sodium channel	SCNN1B	39	Peroxisome-proliferator-activated receptor γ	PRARG	60
Endothelial function			Cholesteryl ester transfer protein	CETP	61
Enos	NOS3	40	β3-Adrenergicreceptor	ADRB3	62
Endothelin 1	EDN1	41	Endocrine		
Dimethylarginne dimethylaminohydrolase 1	DDAH1	42	Oestrogen receptor	ESR1	63
Dimethylarginne dimethylaminohydrolase 2	DDAH2	42	Oestrogen receptor	ESR2	64
G-protein β3	GNB3	43	Androgen receptor	AR	65
Cytokines			Inhibin α	INHA	66
TNFα	TNF	32,44	Angiogenesis		
TGF β1	TGF	45	VEGF	VEGF	67
IGF II	IGF2	46	Matrix metallopeptidase 1	MMP1	68
Interleukin 1α	IL1A	47	Catechol-methyltransferase	COMT	69

several smaller studies in a Meta-analyses. One recent Meta-analyses failed to find any evidence for an increased risk of pre-eclampsia associated with the *MTHFR* 677C>T variant [pooled odds ratio, 1.01 (95% CI, 0.79-1.29)][77]; an earlier Meta-analyses suggested that 677C>T may be associated with severe pre-eclampsia only [diastolic blood pressure ≥110 mm Hg; odds ratio, 1.41 (95% CI, 1.03-1.73)][78]. Meta-analyses of prothrombin 20210G>A did not support a role for this polymorphism in pre-eclampsia [odds ratio, 1.37 (95% CI, 0.72-2.57)][77]. Factor V Leiden was associated with an approx. 2-fold increase in risk of pre-eclampsia

in three separate meta-analyses, although many of the same studies were incorporated in these particular meta-analyses[77, 79, 80]. Some important general points have emerged from these meta-analyses. There was considerable variation in the recruitment protocols for case-control studies and in the phenotypic profile of affected women. Furthermore, there was statistical evidence of heterogeneity between the results of studies; in particular, large studies and studies published within the last few years tended to yield negative results, whereas the majority of positive results came from earlier and smaller studies.

Genome-wide linkage analysis

Genome-wide linkage analysis is the study of genetic variations across the entire human genome. Genome-wide screening has the potential to detect genetic loci that contribute to susceptibility to pre-eclampsia and identify susceptibility loci for development of this disease. Linkage analysis has proved its worth in identifying the molecular basis of many Mendelian disorders, but has been less successful in guiding the search for susceptibility genes for complex disorders. This may be due to underlying genetic heterogeneity (susceptibility loci may differ between affected pedigrees) or to the relatively low genetic risk of disease conferred by individual genes in a complex mode of inheritance. Ascertainment of multicase families presents difficulties in studies of pre-eclampsia, as there is no known male phenotype, and susceptibility in females is apparent only during pregnancy. Nevertheless, research groups from Iceland, Australia, Finland, and Netherlands have undertaken genome-wide linkage screens which have yielded encouraging results (Fig. 3)[81]. The studies in
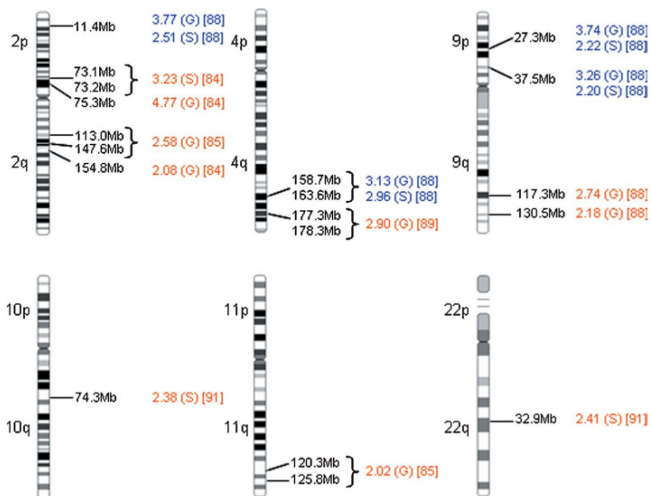
Fig. 3. Chromosomal loci linked to pre-eclampsia[81]. Chromosomal position is given according to NCBI (National Center for Biotechnology Information) Build 35.1. LOD scores (red) or non-parametric linkage scores (blue) are shown; the highest scores provide the strongest evidence of linkage. The results of linkage analysis using both strict diagnostic criteria (S, pre-eclampsia/eclampsia only) and general diagnostic criteria (G, includes pregnancy-induced hypertension) are shown.

Australia indicated the *PREG1* gene as a contributor of pre-eclampsia development[82]. In a genome-wide linkage analysis of Icelandic families representing 343 affected women, a significant locus was found on chromosome 2 (2p13)[83], while in another genome-wide linkage study of pre-eclampsia, evidence was found for a candidate region on 4q[84]. In a large Norwegian population (case-control study, 1139 cases, 2269 controls), 71 SNPs within candidate genes in the region 2q22-23 were genotyped. The gene encoding activin receptor 2 (ACVR2A) lies within this region. Activin is a possible autocrine/paracrine regulator of the human placenta[85]. The type II activin receptor has a molecular weight of 85 kD, and three are two receptor isoforms (ACVR2A and ACVR2B) that have tissue- and temporal-specific differences. The available evidence suggests that ACVR2A may play a role in the etiology of pre-eclampsia[86, 87].

Epigenetic markers and gene-gene interactions have also been investigated in genome-wide association studies to elucidate the pathophysiological processes involved in pre-eclampsia. It has been proposed that SERPIN (serine protease inhibitor) proteins could contribute to the development of pre-eclampsia. *SERPIN* genes encode more than 36 proteins with various functions ranging from protease inhibitors, storage proteins, carrier proteins, to hormone precursors without an inhibitory function. SERPIN family proteins are involved in the coagulation and fibrinolysis cascade, inflammatory processes, complement activation, and phagocytosis. Alterations in promoter methylation (hypomethylation or total methylation) of the genes encoding SERPIN proteins could play role in transcriptional regulation (alterations in CpG methylation can influence the binding of transcription factors) and activity of *SERPIN* genes.

Much attention has been paid to the gene coding for SERPINA3 (alpha-1-antichymotrypsin), which is thought to be involved in pre-eclampsia[88]. Recently, *STOX1* (storkhead box 1) missense mutations have been suggested to predispose to pre-eclampsia in Dutch families[89]. *STOX1* is a transcriptional factor that is encoded by

a gene on chromosome 10 (10q22). This factor is placentally expressed and controls polyploidization of extravillous trophoblasts. Four *STOX1* transcripts are expressed in the early placenta (including extravillous trophoblasts), and alterations in *STOX1* activity have been implicated in pre-eclampsia susceptibility. Several recent studies, however, have contested the generality of the involvement of this gene[90]. Further investigation into the function of *STOX1* is therefore required to determine if this gene is indeed involved in the pathophysiological cascades that lead to the onset of pre-eclampsia.

It should be noted that there is little overlap between the loci identified in different populations in these anaysis, possibly because of genetic differences between populations, highlighting the importance of carrying out genetic studies in different parts of the world.

Conclusion

Genetic screening could potentially be the initial step in pre-eclampsia prevention/treatment and could increase our understanding of the pathophysiological mechanisms underlying pre-eclampsia. Where should genetic studies of pre-eclampsia be directed? Recent genome-wide linkage analysis of families affected by pre-eclampsia has yielded encouraging results. In contrast, the credibility of candidate gene studies has been undermined by conflicting and inconclusive results. It is clear that large studies that have adequate statistical power to detect small genetic effects are needed to reliably identify or exclude susceptibility genes. This invites a multicentre collaborative approach between clinicians and geneticists to develop common recruitment protocols for the establishment of large DNA resource databases that can be used by statisticians to conduct meaningful meta-analyses. The results of pathophysiological and genetic studies have provided some insights into the nature of pre-eclampsia, but an understanding of the fundamental causes of pre-eclampsia remains tantalizingly elusive.

국문초록

자간전증은 전세계적으로 모성 및 주산기 사망과 이환의 주된 원인이나 아직까지 병인기전은 명확하게 규명되지 않은 실정이다. 자간전증은 일반적으로 두 단계 질환으로 알려져 있으며, 그 임상의 첫 단계는 모체의 나선동맥의 얕은 세포영양아층 침투에 의한 태반 부전이 발생한다. 태반 부전에 의한 허혈성 태반이 모체의 순환 혈류 내로 용해성 인자와 싸이토카인, 영양막 조직파편을 유리하면, 전신적인 내피세포 손상 및 기능 부전을 야기하고, 이로 인하여 자간전증 이차 단계인 모체 증후군이 나타난다. 역학적 연구에서 자간전증에 대한 유전적 소인이 일관되게 증명되었다. 집중적 연구 노력에 의한 감수성 유전자 발견은 자간전증의 병태생리를 이해하는데 있어서 유용한 정보를 줄 것이며 자간전증의 치료 및 예방 방법에 대한 방향을 제시할 것이다. 본 주제에서는 자간전증의 병태생리에 있어서 유전적 요인의 역할에 대한 최신 이해를 요약하고 자간전증의 유전적 실마리를 찾기 위한 분자적 접근에 대해 설명하고자 한다.

References

1) Wen SW, Chen XK, Rodger M, White RR, Yang Q, Smith GN, et al. Folic acid supplementation in early second trimester and the risk of preeclampsia. Am J Obstet Gynecol 2008;198:45.e1-7.
2) Bucher HC, Guyatt GH, Cook RJ, Hatala R, Cook DJ, Lang JD, et al. Effect of calcium supplementation on pregnancy-induced hypertension and preeclampsia: a Meta-analyses of randomized controlled trials. JAMA 1996;275:1113-7.
3) Coomarasamy A, Honest H, Papaioannou S, Gee H, Khan KS. Aspirin for prevention of preeclampsia in women with historical risk factors: a systematic review. Obstet Gynecol 2003;101:1319-32.
4) Rumiris D, Purwosunu Y, Wibowo N, Farina A, Sekizawa A. Lower rate of preeclampsia after antioxidant supplementation in pregnant women with low antioxidant status. Hypertens Pregnancy 2006;25:241-53.
5) Chesley LC, Annitto JE, Cosgrove RA. The familial factor in toxemia of pregnancy. Obstet Gynecol 1968; 32:303-11.
6) Duckitt K, Harrington D. Risk factors for pre-eclampsia at antenatal booking: systematic review of controlled studies. BMJ 2005;330:565.

7) Sibai BM, Ewell M, Levine RJ, Klebanoff MA, Esterlitz J, Catalano PM, et al. Risk factors associated with preeclampsia in healthy nulliparous women. Am J Obstet Gynecol 1997;177:1003-10.

8) Report of the National High Blood Pressure Education Program. Working group report on high blood ppressure in pregnancy. Am J Obstet Gynecol 2000;183:S1-22.

9) Sibai BM. Diagnosis and management of gestational hypertension and preeclampsia. Obstet Gynecol 2003; 102:181-92.

10) Brown MA, Hague WM, Higgins J, Lowe S, McCowan L, Oats J, et al. The detection, investigation and management of hypertension in pregnancy: full consensus statement. Aust N Z J Obstet Gynaecol 2000;40:139-55.

11) Hauth JC, Ewell MG, Levine RJ, Esterlitz JR, Sibai B, Curet LB, et al. Pregnancy outcomes in healthy nulliparas who developed hypertension. Obstet Gynecol 2000;95:24-8.

12) Buchbinder A, Sibai BM, Caritis S, Macpherson C, Hauth J, Lindheimer MD, et al. Adverse perinatal outcomes are significantly higher in severe gestational hypertension than in mild preeclampsia. Am J Obstet Gynecol 2002;186:66-71.

13) Cunningham FG, Leveno KJ, Bloom SL, Hauth JC, Rouse DJ, Spong CY. Pregnancy Hypertension. Williams Obstetrics. 23rd ed. McGraw-Hill: New York, 2009; 9526.

14) Caritis S, Sibai B, Hauth J, Lindheimer MD, Klebanoff M, Thom E, et al. Low-dose aspirin to prevent preeclampsia in women at high risk. N Engl J Med 1998; 338:701-5.

15) Sibai BM. Chronic hypertension in pregnancy. Obstet Gynecol 2002;100:369-77.

16) Hahn S, Gupta AK, Troeger C, Rusterholz C, Holzgreve W. Disturbances in placental immunology: ready for therapeutic interventions? Semin Immunopathol 2006; 27:477-93.

17) Huppertz B. The feto-maternal interface: setting the stage for potential immune interactions. Semin Immunopathol 2007;29:83-94.

18) Young BC, Levine RJ, Karumanchi SA. Pathogenesis of preeclampsia. Annu Rev Pathol 2010;5:173−92.

19) Wang A, Rana S, Karumanchi SA. Preeclampsia: the role of angiogenic factors in its pathogenesis. Physiology 2009;24:147-58.

20) Hung TH, Skepper JN, Charnock-Jones DS, Burton GJ. Hypoxia-reoxygenation: a potent inducer of apoptotic changes in the human placenta and possible etiological factor in preeclampsia. Circ Res 2002;90:1274-81.

21) Soleymanlou N, Jurisica I, Nevo O, Ietta F, Zhang X, Zamudio S, et al. Molecular evidence of placental hypoxia in preeclampsia. J Clin Endocrinol Metab 2005;90:4299-308.

22) Redman CW, Sargent IL. Latest advances in understanding preeclampsia. Science 2005;308:1592-4.

23) Maynard SE, Min JY, Merchan J, Lim KH, Li J, Mondal S, et al. Excess placental soluble fms-like tyrosine kinase 1 (sFlt1) may contribute to endothelial dysfunction, hypertension, and proteinuria in preeclampsia. J Clin Invest 2003;111:649-58.

24) Roberts JM, Taylor RN, Musci TJ, Rodgers GM, Hubel CA, McLaughlin MK. Preeclampsia: an endothelial cell disorder. Am J Obstet Gynecol 1989;161:1200-4.

25) Huppertz B. Placental origins of preeclampsia: challenging the current hypothesis. Hypertension 2008; 51:970-5.

26) Roberts JM, Hubel CA. The two stage model of preeclampsia: variations on the theme. Placenta 2009;30 Suppl A:S32-7.

27) O'Shaughnessy KM, Fu B, Ferraro F, Lewis I, Downing S, Morris NH. Factor V Leiden and thermolabile methylenetetrahydrofolate reductase gene variants in an East Anglian preeclampsia cohort. Hypertension 1999;33:1338-41.

28) Kim YJ, Williamson RA, Murray JC, Andrews J, Pietscher JJ, Peraud PJ, et al. Genetic susceptibility to preeclampsia: roles of cytosineto-thymine substitution at nucleotide 677 of the gene for methylenetetrahydrofolate reductase, 68-base pair insertion at nucleotide 844 of the gene for cystathionine beta-synthase, and factor V Leiden mutation. Am J Obstet Gynecol 2001; 184:1211-7.

29) Morrison ER, Miedzybrodzka ZH, Campbell DM, Haites NE, Wilson BJ, Watson MS, et al. Prothrombotic genotypes are not associated with pre-eclampsia and gestational hypertension: results from a large population-based study and systematic review. Thromb Haemost 2002;87:779-85.

30) O'Shaughnessy KM, Fu B, Downing S, Morris NH. Thrombophilic polymorphisms in pre-eclampsia: altered frequency of the functional 98C>T polymorphism of glycoprotein IIIa. J Med Genet 2001;38:775-7.

31) Pegoraro RJ, Chikosi A, Rom L, Roberts C, Moodley J. Methylenetetrahydrofolate reductase gene polymorphisms in black South Africans and the association with preeclampsia. Acta Obstet Gynecol Scand 2004; 83:449-54.

32) Lévesque S, Moutquin JM, Lindsay C, Roy MC, Rousseau F. Implication of an AGT haplotype in a multigene association study with pregnancy hypertension. Hy-

pertension 2004;43:71-8.

33) Laasanen J, Hiltunen M, Punnonen K, Mannermaa A, Heinonen S. Fibrinogen and factor VII promoter polymorphisms in women with preeclampsia. Obstet Gynecol 2002;100:317-20.

34) Borg AJ, Higgins JR, Brennecke SP, Moses EK. Thrombomodulin Ala455Val dimorphism is not associated with pre-eclampsia in Australian and New Zealand women. Gynecol Obstet Invest 2002;54:43-5.

35) Clark P, Freeman DJ, Streja E, Sattar N, Walker ID, Greer IA. The G-to-T point mutation in codon 34 of the factor XIII gene and the risk of pre-eclampsia. Blood Coagul Fibrinolysis 2003;14:155-7.

36) Roberts CB, Rom L, Moodley J, Pegoraro RJ. Hypertension-related gene polymorphisms in pre-eclampsia, eclampsia and gestational hypertension in Black South African women. J Hypertens 2004;22:945-8.

37) Maruyama A, Nakayama T, Furuya K, Mizutani Y, Yamamoto T. Association study between the human Renin gene and preeclampsia. Hypertens Pregnancy 2005;24:39-48.

38) Plummer S, Tower C, Alonso P, Morgan L, Baker P, Broughton-Pipkin F, et al. Haplotypes of the angiotensin II receptor genes AGTR1 and AGTR2 in women with normotensive pregnancy and women with preeclampsia. Hum Mutat 2004;24:14-20.

39) Pegoraro RJ, Roberts CB, Rom L, Moodley J. T594M mutation of the epithelial sodium channel beta-subunit gene in pre-eclampsia and eclampsia in Black South African women. BJOG 2004;111:1012-3.

40) Serrano NC, Casas JP, Díaz LA, Páez C, Mesa CM, Cifuentes R, et al. Endothelial NO synthase genotype and risk of preeclampsia: a multicenter case-control study. Hypertension 2004;44:702-7.

41) Barden AE, Herbison CE, Beilin LJ, Michael CA, Walters BN, Van Bockxmeer FM. Association between the endothelin-1 gene Lys198Asn polymorphism blood pressure and plasma endothelin-1 levels in normal and pre-eclamptic pregnancy. J Hypertens 2001;19:1775-82.

42) Akbar F, Heinonen S, Pirskanen M, Uimari P, Tuomainen TP, Salonen JT. Haplotypic association of DDAH1 with susceptibility to pre-eclampsia. Mol Hum Reprod 2005;11:73-7.

43) Jansen MW, Bertina RM, Vos HL, Bloemenkamp KW, Helmerhorst FM, Wladimiroff JW, et al. C825T polymorphism in the human G protein beta 3 subunit gene and preeclampsia; a case control study. Hypertens Pregnancy 2004;23:211-8.

44) Lachmeijer AM, Crusius JB, Pals G, Dekker GA, Arngrímsson R, ten Kate LP. Polymorphisms in the tumor necrosis factor and lymphotoxin-alpha gene region and preeclampsia. Obstet Gynecol 2001;98:612-9.

45) Kim SY, Lim JH, Park SY, Yang JH, Kim MY, Kim MH, et al. Transforming growth factor-beta 1 gene polymorphisms in Korean patients with pre-eclampsia. Am J Reprod Immunol 2010;63:291-8.

46) Bermingham J, Jenkins D, McCarthy T, O'Brien M. Genetic analysis of insulin-like growth factor II and HLA-G in pre-eclampsia. Biochem Soc Trans 2000;28:215-9.

47) Haggerty CL, Ferrell RE, Hubel CA, Markovic N, Harger G, Ness RB. Association between allelic variants in cytokine genes and preeclampsia. Am J Obstet Gynecol 2005;193:209-15.

48) Lachmeijer AM, Nosti-Escanilla MP, Bastiaans EB, Pals G, Sandkuijl LA, Kostense PJ, et al. Linkage and association studies of IL1B and IL1RN gene polymorphisms in preeclampsia. Hypertens Pregnancy 2002;21:23-38.

49) Faisel F, Romppanen EL, Hiltunen M, Helisalmi S, Punnonen K, Salonen J, et al. Polymorphism in the interleukin 1 receptor antagonist gene in women with preeclampsia. J Reprod Immunol 2003;60:61-70.

50) de Groot CJ, Jansen MW, Bertina RM, Schonkeren JJ, Helmerhorst FM, Huizinga TW. Interleukin 10-2849AA genotype protects against pre-eclampsia. Genes Immun 2004;5:313-4.

51) Samsami Dehaghani A, Doroudchi M, Kalantari T, Pezeshki AM, Ghaderi A. Heterozygosity in CTLA-4 gene and severe preeclampsia. Int J Gynaecol Obstet 2005;88:19-24.

52) Sziller I, Nguyen D, Halmos A, Hupuczi P, Papp Z, Witkin SS. An A > G polymorphism at position -670 in the Fas (TNFRSF6) gene in pregnant women with pre-eclampsia and intrauterine growth restriction. Mol Hum Reprod 2005;11:207-10.

53) Zusterzeel PL, Peters WH, Visser W, Hermsen KJ, Roelofs HM, Steegers EA. A polymorphism in the gene for microsomal epoxide hydrolase is associated with pre-eclampsia. J Med Genet 2001;38:234-7.

54) Zusterzeel PL, Visser W, Peters WH, Merkus HW, Nelen WL, Steegers EA. Polymorphism in the glutathione S-transferase P1 gene and risk for preeclampsia. Obstet Gynecol 2000;96:50-4.

55) Kim YJ, Park HS, Park MH, Suh SH, Pang MG. Oxidative stress-related gene polymorphism and the risk of preeclampsia. Eur J Obstet Gynecol Reprod Biol 2005;119:42-6.

56) Raijmakers MT, Roes EM, te Morsche RH, Steegers EA, Peters WH. Haptoglobin and its association with

the HELLP syndrome. J Med Genet 2003;40:214-6.

57) Raijmakers MT, Roes EM, Steegers EA, Peters WH. The C242T-polymorphism of the NADPH/NADH oxidase gene p22phox subunit is not associated with pre-eclampsia. J Hum Hypertens 2002;16:423-5.

58) Kim YJ, Williamson RA, Chen K, Smith JL, Murray JC, Merrill DC. Lipoprotein lipase gene mutations and the genetic susceptibility of preeclampsia. Hypertension 2001;38:992-6.

59) Chikosi AB, Moodley J, Pegoraro RJ, Lanning PA, Rom L. Apolipoprotein E polymorphism in South African Zulu women with preeclampsia. Hypertens Pregnancy 2000;19:309-14.

60) Laasanen J, Heinonen S, Hiltunen M, Mannermaa A, Laakso M. Polymorphism in the peroxisome prolife-rator-activated receptor-gamma gene in women with preeclampsia. Early Hum Dev 2002;69:77-82.

61) Belo L, Gaffney D, Caslake M, Santos-Silva A, Pereira-Leite L, Quintanilha A, et al. Apolipoprotein E and cholesteryl ester transfer protein polymorphisms in normal and preeclamptic pregnancies. Eur J Obstet Gynecol Reprod Biol 2004;112:9-15.

62) Malina AN, Laivuori HM, Agatisa PK, Collura LA, Crombleholme WR, Sims CJ, et al. The Trp64Arg polymorphism of the beta3-adrenergic receptor is not increased in women with preeclampsia. Am J Obstet Gynecol 2004;190:779-83.

63) Malamitsi-Puchner A, Tziotis J, Evangelopoulos D, Fountas L, Vlachos G, Creatsas G, et al. Gene analysis of the N-terminal region of the estrogen receptor alpha in preeclampsia. Steroids 2001;66:695-700.

64) Maruyama A, Nakayama T, Sato N, Mizutani Y, Furuya K, Yamamoto T. Association study using single nu-cleotide polymorphisms in the estrogen receptor beta (ESR2) gene for preeclampsia. Hypertens Res 2004; 27:903-9.

65) Lim JH, Kim SY, Lee SW, Park SY, Han JY, Chung JH, et al. Association between genetic polymorphisms in androgen receptor gene and the risk of preeclampsia in Korean women. J Assist Reprod Genet 2011;28: 85-90.

66) Ciarmela P, Florio P, Battistini S, Grasso D, Amato T, Boschi S, et al. Mutational analysis of the inhibin alpha gene in preeclamptic women. J Endocrinol Invest 2005;28:30-3.

67) Papazoglou D, Galazios G, Koukourakis MI, Panago-poulos I, Kontomanolis EN, Papatheodorou K, et al. Vascular endothelial growth factor gene polymorphisms and pre-eclampsia. Mol Hum Reprod 2004;10:321-4.

68) Jurajda M, Kanková K, Muzik J, Unzeitig V, Drábková M, Izakovicová-Hollá L, et al. Lack of an association

of a single nucleotide polymorphism in the promoter of the matrix metalloproteinase-1 gene in Czech women with pregnancy-induced hypertension. Gynecol Obstet Invest 2001;52:124-7.

69) Lim JH, Kim SY, Kim dJ, Park SY, Han HW, Han JY, et al. Genetic polymorphism of catechol-O-methyl-transferase and cytochrome P450c17α in preeclampsia. Pharmacogenet Genomics 2010;20:605-10.

70) Lim JH, Kim SY, Park SY, Yang JH, Han JY, Hong DS, et al. The association between polymorphisms of β-adrenoceptors and preeclampsia. J Genet Med 2007; 4:160-6.

71) Kim SY, Park SY, Lim JH, Yang JH, Kim MY, Park HY, et al. No association between endothelin-1 gene polymorphisms and preeclampsia in Korean population. J Genet Med 2008;5:34-40.

72) Lim JH, Park SY, Kim SY, Lee MH, Yang JH, Kim MY, et al. Intercellular adhesion molecule-1 gene poly-morphism (K469E) in Korean preeclamptic women. J Genet Med 2008;5:105-10.

73) Kim SY, Lim JH, Yang JH, Kim MY, Kim MH, Park SY, et al. The 769G>A variant of the inhibin-alpha gene in Korean patients with preeclampsia. J Endocrinol Invest 2008;31:700-3.

74) Kim SY, Park SY, Lim JH, Yang JH, Kim MY, Park HY, et al. The Y153H variant of the STOX1 gene in Korean patients with preeclampsia. J Genet Med 2009;6:56-61.

75) Lim JH, Kim SY, Park SY, Han HW, Yang JH, Kim MY, et al. Tumor necrosis factor-alpha gene polymorphism (C-850T) in Korean patients with preeclampsia. J Genet Med 2009;6:155-60.

76) Colhoun HM, McKeigue PM, Davey Smith G. Problems of reporting genetic associations with complex out-comes. Lancet 2003;361:865-72.

77) Lin J, August P. Genetic thrombophilias and pree-clampsia: a Meta-analyses. Obstet Gynecol 2005;105: 182-92.

78) Kosmas IP, Tatsioni A, Ioannidis JP. Association of C677T polymorphism in the methylenetetrahydrofolate reductase gene with hypertension in pregnancy and pre-eclampsia: a Meta-analyses. J Hypertens 2004; 22:1655-62.

79) Kosmas IP, Tatsioni A, Ioannidis JP. Association of Leiden mutation in factor V gene with hypertension in pregnancy and pre-eclampsia: a Meta-analyses. J Hypertens 2003;21:1221-8.

80) Dudding TE, Attia J. The association between adverse pregnancy outcomes and maternal factor V Leiden genotype: a Meta-analyses. Thromb Haemost 2004; 91:700-11.

81) Chappell S, Morgan L. Searching for genetic clues to the causes of pre-eclampsia. Clin Sci 2006;110:443-58.

82) Makino Y, Shibata K, Makino I, Kangawa K, Kawarabayashi T. Alteration of the adrenomedullin receptor components gene expression associated with the blood pressure in pregnancy-induced hypertension. J Clin Endocrinol Metab 2001;86:5079-82.

83) Arngrímsson R, Sigurardóttir S, Frigge ML, Bjarnadóttir RI, Jónsson T, Stefánsson H, et al. A genome-wide scan reveals a maternal susceptibility locus for pre-eclampsia on chromosome 2p13. Hum Mol Genet 1999;8:1799-805.

84) Harrison GA, Humphrey KE, Jones N, Badenhop R, Guo G, Elakis G, et al. A genomewide linkage study of preeclampsia/eclampsia reveals evidence for a candidate region on 4q. Am J Hum Genet 1997;60:1158-67.

85) Fitzpatrick E, Göring HH, Liu H, Borg A, Forrest S, Cooper DW, et al. Fine mapping and SNP analysis of positional candidates at the preeclampsia susceptibility locus (PREG1) on chromosome 2. Hum Biol 2004;76:849-62.

86) Peng C, Huang TH, Jeung EB, Donaldson CJ, Vale WW, Leung PC. Expression of the type II activin receptor gene in the human placenta. Endocrinology 1993;133:3046-9.

87) Roten LT, Johnson MP, Forsmo S, Fitzpatrick E, Dyer TD, Brennecke SP, et al. Association between the candidate susceptibility gene ACVR2A on chromosome 2q22 and pre-eclampsia in a large Norwegian population-based study (the HUNT study). Eur J Hum Genet 2009;17:250-7.

88) Fitzpatrick E, Johnson MP, Dyer TD, Forrest S, Elliott K, Blangero J, et al. Genetic association of the activin A receptor gene (ACVR2A) and pre-eclampsia. Mol Hum Reprod 2009;15:195-204.

89) van Dijk M, Mulders J, Poutsma A, Könst AA, Lachmeijer AM, Dekker GA, et al. Maternal segregation of the Dutch preeclampsia locus at 10q22 with a new member of the winged helix gene family. Nat Genet 2005;37:514-9.

90) Iglesias-Platas I, Monk D, Jebbink J, Buimer M, Boer K, van der Post J, et al. STOX1 is not imprinted and is not likely to be involved in preeclampsia. Nat Genet 2007;39:279-80.