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Angiotensin I-Converting Enzyme Insertion-Related Genotypes and Allele are Associated With Higher Susceptibility of Endometriosis and Leiomyoma

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ABSTRACT Endometriosis and leiomyoma display features similar to malignancy, requiring neovascularization to proliferation and growth. Altered vascular-related genes might be related to the development of endometriosis and leiomyoma. Polymorphisms of the angiotensin-converting enzyme (ACE) insertion/deletion (I/D) genes have been linked with some vascular diseases. This study investigates whether ACE I/D gene polymorphisms could be used as markers of susceptibility in endometriosis and leiomyoma. Women were divided into three groups: (1) endometriosis (n = 125); (2) leiomyoma (n = 120); (3) normal controls (n = 128). Genomic DNA was obtained from peripheral leukocyte. ACE I/D gene polymorphisms in intron 16 were amplified by polymerase chain reaction and restriction fragment length polymorphism (PCR-RFLP) Genotypes and allelic frequencies in both groups were compared. We observed the genotype distribution and allele frequency of ACE I/D gene polymorphisms in both groups were significantly different. Proportions of ACE*I homozygote/heterozygote/D homozygote in both groups were: (1) 50.4/24/25.6%; (2) 25/23.33/51.67%; (3) 10.2/29.7/60.1%. Proportions of I/D alleles in each group were: (1) 62.4/37.6%; (2) 36.7/63.3%; (3) 25/75%. We concluded that ACE*I/D gene polymorphisms are associated with endometriosis and leiomyoma susceptibilities. ACE*I-related genotypes and allele are strongly related to the occurrence of endometriosis and moderately related to the occurrence of leiomyoma. *Mol. Reprod. Dev. 74: 808–814, 2007.*
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Key Words: angiotensin I-converting enzyme, ACE; endometriosis; leiomyoma; polymorphism

INTRODUCTION

Endometriosis, a polygenic/multifactorial disease, is related with the complex interactions between hormone and cytokines activation, immunoinflammatory process, and genetic factors (Vigano et al., 1998). Endometriosis displays some features of malignancy, including local invasion and aggressive spread to distant organs. Similar to tumor metastases, endometriotic implants require neovascularization to become established, grow, and invade tissues. The neovascular processes are prominent in the endometriosis tissues. Leiomyoma, the most common benign uterine neoplasma, is occurred in around one-fourth of the women during their lifetimes (Cramer, 1992). Leiomyoma is related with the complex mechanism of auto- and paracrine interaction or the effect of sex-steroid hormone action on cells (El-Badry et al., 1991).

Cardiovascular genes play a role in the regulation and growth of tumor. Altered vascular-related genes might be related with the development of endometriosis or leiomyoma. The renin-angiotensin system (RAS) regulates blood pressure through its effects on vascular tone, renal hemodynamics, and fluid–electrolyte balance (Fornage et al., 1998). Renin converts angiotensinogen to angiotensin I. Angiotensin I-converting enzyme (ACE) cleaves angiotensin I to angiotensin II, which is the key component in RAS (Berge and Berg, 1994). ACE regulates the systemic circulation through angiotensin II formation and kinin metabolism. The ACE (encoded by the gene *DCP1*) and *RAS* genes are related with the regulatory pathway in cardiovascular disease (Zhu et al., 2001); while the *ACE* gene is

ACE I-related genotypes and allele are associated with higher susceptibilities of endometriosis and myoma.

Yao-Yuan Hsieh and Cheng-Chun Lee have equal contributions for this survey.

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implicated as a risk factor for coronary artery disease and myocardial infarction (Zhu et al., 2001).

Angiogenesis and vascular remodeling play critical roles in the growth, invasion, and regression of endometriosis or leiomyoma (Donnez et al., 1998). It has been demonstrated that the presence of angiotensin receptors in endometrial tissue (Braileanu et al., 2002). Angiotensin II in endometrial stromal cells was mediated via angiotensin I receptors (Braileanu et al., 2002). Angiotensin II could increase the intracellular calcium concentration by interaction with angiotensin receptor in endometrial stromal cells (Braileanu et al., 2002). Vasopressin also stimulates phospholipase C activity in endometrial explants (Braileanu et al., 2001). These findings suggested the underlying contributions of ACE upon the development of endometriosis and leiomyoma.

Heritable genetic factors may contribute to the initiation and progression of endometriosis (Treloar et al., 1999). Gene polymorphisms are useful tools in the study of multifactorial disorders (Anderson et al., 1994). Molecular geneticists are developing the third-generation human genome map with single-nucleotide polymorphisms (SNPs). The analyses of SNPs can be implemented to analyze the mechanisms of complex genetic disorders. Numerous chronic disorders, such as endometriosis, leiomyoma, osteoporosis, hypertension, diabetes, and asthma, have been attributed to genetic susceptibility.

Most studies on ACE gene polymorphisms were focused on their associations with cardiovascular diseases, serum ACE level, and blood pressure (Zhu et al., 2001). Few investigators demonstrated their roles in gynecological diseases, such as endometriosis and leiomyoma. Recently, a 287-bp Alu insertion/deletion (I/D) restriction fragment length polymorphism (RFLP) in intron 16 of the human *ACE* gene has been largely surveyed in individual diseases. However, no investigator demonstrates their association with endometriosis or leiomyoma. Herein, we tried to evaluate the associations of ACE I/D polymorphisms with endometriosis and leiomyoma. To the best of our knowledge, this is the first survey in this field.

MATERIALS AND METHODS

Premenopausal Taiwanese women with surgically diagnosed severe endometriosis, leiomyoma, and normal individuals without endometriosis and leiomyoma were included. The nonendometriosis or nonleiomyoma statuses were confirmed after detail ultrasonography examination. All patients were divided into three groups: (1) severe endometriosis (n = 125, according to revised American Fertility Society classification); (2) leiomyoma (n = 120); (3) normal controls (n = 128). The studies were approved by the ethical committee and institutional review board of the China Medical University Hospital. Informed consents were signed by all women who donated their blood.

All women accepted the peripheral blood sampling for genotype analyses. The ACE I/D polymorphism (intron 17) were determined according to previous described condition (Abbud et al., 1998). Genomic DNA was isolated from peripheral blood using Genomaker DNA extractor kit (Blossom, Taipei, Taiwan). About 50 ng of genomic DNA was mixed with 20 pmol of polymerase chain reaction (PCR) primer in a total volume of 25 μl containing 10 mM Tris-Hcl, pH 8.3, 50 mM Kcl, 1.5 mM $MgCl_2$, 0.2 mM each deoxyribonucleotide triphosphate, and 1 unit of Amplitaq DNA polymerase (Perkin Elmer Applied Biosystems, Foster City, CA). For the ACE I/D polymorphism, a 300-bp fragment of ACE was amplified by PCR. The sequences of the primers were as following (from 5′ to 3′ end): forward, CTGGAGACCACTCCCATCCTTTCT; reverse, GATGTGGCCATCACATTCGTCAGAT. The SNP information for the genes involved was obtained through internet (http://www.ncbi.nlm.nih.gov/LocusLink/).

The PCR amplification was performed in a programmable thermal cycler GenAmp PCR system 2400 (Perkin Elmer Applied Biosystems). The PCR conditions were set as follows: one cycle at 94°C for 5 min, 35 cycles at 94°C for 30 sec, 58°C for 45 sec, 72°C for 45 sec, and final cycle of extension at 72°C for 10 min. After complete PCR processes, the products included a 490-bp fragment (with 300-bp insertion) for "I" (Insertion) allele and a 190-bp fragment for "D" (Deletion) allele. Thus, each sample revealed one of three electrophoresis patterns: a 490-bp band corresponding to I/I genotype, a 190-bp band corresponding to D/D genotype, or a combination of 490- and 190-bp bands corresponding to I/D genotype.

PCR products (5 μl) were loaded into 3% agarose gel containing ethidium bromide for electrophoresis. Genotypes and allelic frequencies for ACE I/D gene polymorphisms in both groups were compared. Correlations between the ACE I/D genotype and endometriosis/leiomyoma was evaluated. Allelic frequencies are expressed as a percentage of the total number of alleles. The SAS system (version 8.1, SAS Institute, Inc., Cary, NC) with χ^2 test were utilized for statistical analyses. A P-value of <0.05 was considered statistically significant.

RESULTS

Genotype distribution and allele frequency of ACE I/D gene polymorphisms in three groups were significantly different (Table 1). Higher percentages of I-related genotype (I homozygote, I/D heterozygote) presented in the endometriosis/leiomyoma population compared to normal controls. Proportions of insertion homozygote/heterozygote/deletion homozygote for ACE in three groups were: (1) 50.4/24/25.6%; (2) 25/23.33/51.67%; (3) 10.2/29.7/60.1%, respectively (P-value < 0.0001, Table 1). Allele frequencies for ACE I/D polymorphism between tumor groups and normal controls were also significantly different (Table 2). Proportions of I/D alleles in both groups were: (1) 62.4/37.6%; (2) 36.7/63.3%; (3) 20.4/79.6%, respectively (P-value < 0.0001, Table 2).

Most common genotype and allele for ACE I/D gene polymorphisms in endometriosis groups were I-related genotype and allele. The I and D distributions were comparable in leiomyoma group. In contrast, the

TABLE 1. Distributions of ACE I/D Gene Polymorphisms in Women With Endometriosis, Leiomyoma, and Normal Controls

Genotype	Endometriosis (n = 125) (%)*,***	Leiomyoma (n = 120) (%)**,***	Normal controls (n = 128) (%)*,**
Insertion/insertion	63 (50.4)	30 (25)	13 (10.2)
Insertion/deletion	30 (24)	28 (23.33)	38 (29.7)
Deletion/deletion	32 (25.6)	62 (51.67)	77 (60.1)

P-value was calculated by χ^2 test.
**P*-value < 0.000001.
***P*-value = 0.008.
****P*-value = 0.000024.

D-related genotypes were strikingly higher in normal populations (Tables 1 and 2). These findings indicated that ACE*I-related genotype and allele were strongly associated with higher susceptibility of endometriosis as well as moderately correlated with the susceptibility of leiomyoma.

DISCUSSION

Endometriosis is a common gynecological tumor, but its etiology remains unclear. The prevalence of endometriosis is around 10% in general population (Goldman and Cramer, 1989) and as high as 30–40% in infertile women (Strathy et al., 1982). Numerous hormones and cytokines are associated with endometriosis formation, including estrogen, progesterone (Irahara et al., 2001), insulin-like growth factor (Boehm et al., 1990), epidermal growth factor, fibroblast growth factor (Di Lieto et al., 1997), etc. The endometrium, which has prominent vessels and blood flow, is one of the few adult tissues that exhibit regular intervals of rapid growth and abruption. Therefore, angiogenesis is an important component of the growth and function of these tissues. Endometriosis is a disease of endometrium tissues shedding outside the uterus during menstruation. These explants require a rich blood supply, which enables their survivals and growths. Therefore, the activation of angiogenesis might be a key factor in the pathogenesis of endometriosis (Inan et al., 2003).

Leiomyoma is the most common tumor in women, but the related etiopathogenesis is also obscure. Angiogenesis and vascular-related factors are involved in the pathogenesis and growth of leiomyomas. Uterine tissue contains numerous growth factors, which modified vascular expression and blood supply (Di Lieto et al., 2005). Growth factor could preferentially promote the angiogenesis of leiomyoma cells compared with myometrial cells (Strawn et al., 1995). Leiomyoma contains abnormal vascualization than myometrium does, which may be related to the genesis and progression of these tumors (Boehm et al., 1990). The developments of both endometriosis and leiomyoma are associated with exposure to ovarian sex steroids and increased require of vascular supply for their growth. These observations suggest that angiogenic factors may be related with the development of endometriosis/leiomyoma. However, the vascular-related genes contributing to endometriosis and leiomyoma remain largely unknown.

Endometriosis and leiomyoma development and progression were proposed to be multigenic models. The *ACE* gene might be linked to other important genes in the process of progression. However, the pathophysiological role of ACE upon endometriosis or leiomyoma is not fully understood. The implications of ACE polymorphisms on endometriosis or leiomyoma remain to be established. It has been suggested that the RAS is involved in the regulation of cell proliferation in the endometrium (Li et al., 2000; Dinh et al., 2001; Schauser et al., 2001; Baudin, 2002). Some steroid hormones might influence the regulation of RAS (Bachman et al., 1991; Harrap et al., 1993). Estrogens increase the hepatic synthesis of renin, which influence the conversion of angiotensinogen to angiotensin I as well as the following catalyses of ACE (Harrap et al., 1993). Moreover, one of angiotensin II receptors (subtype 2) is abundant in the uterus tissue (Dinh et al., 2001; Baudin, 2002). Therefore, it is logical to suspect that RAS might be involved in endometriosis or leiomyoma.

ACE catalyses the conversion of angiotensin I to the physiologically active peptide angiotensin II, which controls fluid–electrolyte balance and systemic blood

TABLE 2. Allelic Frequencies for ACE I/D Polymorphisms in Women With Endometriosis, Leiomyoma, and Normal Controls

Allele frequencies	Endometriosis (n = 250) (%)*,***	Leiomyoma (n = 240) (%)**,***	Normal controls (n = 256) (%)*,**
Insertion	156 (62.4)	88 (36.7)	64 (25)
Deletion	94 (37.6)	152 (63.3)	192 (75)

P-values were calculated by χ^2 test.
**P*-value < 0.00001.
***P*-value = 0.0048.
****P*-value = 0.0000001.

pressure. ACE activity is related with angiogenesis. ACE inhibition by perindopril improves myocardial angiogenesis (Toblli et al., 2004). Angiotensin II, a key regulator of blood pressure and body fluid homeostasis, exerts mitogenic effects on endothelial cells. Angiotensin II has been shown to have possible mitogenic and angiogenic effects upon tumor cell lines (Koh et al., 2003). Angiotensin II is also a humoral regulator of peripheral angiogenesis (Walther et al., 2003). Because ACE catalyzed the angiotensin II formation, the ACE activity is positively correlated with angiotensin II production. Furthermore, ACE inhibitors were found to significantly inhibit tumor growth and angiogenesis along with suppression of the vascular endothelial growth factor (VEGF) level (Yasumatsu et al., 2004).

Recently, it has been reported that ACE polymorphisms may be determinants in the development of human cancers (Haiman et al., 2003; Koh et al., 2003; Medeiros et al., 2004). The long-term use of ACE may protect against cancer (Lever et al., 1998). The ACE inhibitor (e.g., captopril) has been shown to inhibit proliferation of tumor cells and to reduce tumor growth (Reddy et al., 1995; Volpert et al., 1996; Small et al., 1997; Hii et al., 1998). Angiotensin II might have an important role in carcinogenesis, and the antiangiogenic activity is partly mediated by angiotensin II and ACE inhibition. ACE and angiotensin II inhibitors might be considered as useful anti-tumor agents. Combined these presentations, it is logical to expect that ACE might appear to be an angiogenic and tumorogenesis effects upon endometriosis and leiomyoma.

The *ACE* gene, which is located on chromosome 17q23, contains some gene polymorphisms and candidate markers for hypertension and related diseases (Doria et al., 1994). The ACE gene polymorphism located on the intron 17 of *ACE* gene. *ACE* gene mediates interaction effects of the fibrinolytic and RASs on plasma levels of plasminogen activator inhibitor 1 (PAI-1) (Moore et al., 2002). It might contain linkage disequilibrium with other important gene variant. ACE gene polymorphisms are related to numerous diseases, including coronary heart disease, hypertension, ventricular hypertrophy (Alvarez et al., 2000; Henskens et al., 2003; Sethi et al., 2003), systemic lupus erythematosus, nephritis (Parsa et al., 2002), carotid artery wall thickness (Sayed-Tabatabaei et al., 2003), post-transplant erythrocytosis (Yildiz et al., 2003), diabetic nephropathy (Chang et al., 2003), Alzheimer's disease (Kehoe et al., 2003), ischemic cerebrovascular disease (Um et al., 2003), dementia (Choi et al., 2003), segmental glomerulosclerosis (Dixit et al., 2002), cystic fibrosis (Arkwright et al., 2003), sarcoidosis (Schurmann et al., 2001), etc.

Most studies have associated the presence (insertion, I) or absence (deletion, D) of a 287-bp Alu repeat element in intron 16 with the levels of circulating enzyme or cardiovascular pathophysiologies. ACE I/D polymorphism affects the uteroplacental and umbilical flows as well as the recurrence of an adverse pregnancy outcome in women with pre-eclampsia (Mello et al., 2003). Individuals with the DD genotype show a significantly increased left-ventricular mass in response to physical training, compared to the II or ID genotype as well as the lowest plasma ACE levels (Alvarez et al., 2000). ACE I/D genotype DD might be a promoter to clinical manifestation of sarcoidosis. A significant association was observed between the presence of ACE*D genotype/allele and an elevation of serum ACE activity (Martinez et al., 2000).

Despite many epidemiological studies suggest that the ACE*D allele confers increased susceptibility to cardiovascular disease; however, other reports have found no such association or even a beneficial effect (Rieder et al., 1999). ACE polymorphism may be associated with the development of endometrial carcinoma (Freitas-Silva et al., 2004). The presence of I allele (genotypes ID and II) is significantly associated to an earlier age of onset of endometrial carcinoma (Freitas-Silva et al., 2004). In contrast, the D-related genotype (DD, ID) exhibited higher risk of breast cancer (Koh et al., 2003) and advanced progression of prostate cancer (Medeiros et al., 2004). Furthermore, some investigators have demonstrated the nonassociations between the ACE gene polymorphisms with individual diseases, including hypertension (Harrap et al., 1993; Berge and Berg, 1994; Mondorf et al., 1998), myocardial infarction (Foy et al., 1997), left ventricular hypertrophy (West et al., 1997), pregnancy outcomes, pregnancy-induced hypertension (Tamura et al., 1996), nephronophthisis (Omran et al., 1999), and sarcoidosis (Schurmann et al., 2001).

There is controversial and inconsistent reports about the ACE I/D distributions in individual diseases among different races (Table 3). The D allele has a frequency of approximately 0.53 in Caucasians (Cambien, 1994) in

TABLE 3. Distributions of ACE Insertion/Deletion (I/D) Alleles for Individual Diseases

Literatures	I/D (%) for individual diseases	I/D (%) for normal controls
Zee et al. (1992)	56/44 (hypertension)	41/59
Yamamoto et al. (1997)		64/36 (children)
Isbir et al. (1999)	62.3/37.7 (coronary artery disease)	49.3/50.7
Nakai et al. (1994)	42/58 (coronary artery disease)	58/42
Moriyama et al. (1995)	81/19 (noneffective patients with proteinuria after ACE inhibitors therapy)	53/47 (effective patients with proteinuria after ACE inhibitors therapy)
Matsubara et al. (2002)	67.5/22.5 (hypertension)	

Significant difference.
Nonsignificant difference between systolic/diastolic pressure.

contrasted to a 0.42 in Asians (Hiramori, 1994). These discrepancies might be due to different illness classifications, racial or disease variations. In fact, different ethnic groups might influence the ACE gene distributions (McKenzie et al., 2001). Ethnic variation plays a major role in genetic regulation of serum ACE activity and ACE gene polymorphism to cardiovascular disease (Bloem et al., 1996). Furthermore, the gender-specific influence of these gene polymorphisms should be concerned in the relative surveys.

In our previous survey, we demonstrated the correlations of ACE A2350G and A-2240T gene polymorphisms in endometriosis (Hsieh et al., 2005). We observed that ACE 2350*G and ACE-240*T-related genotypes and alleles are associated with higher susceptibilities of endometriosis. In this study, we further observed that the genotype distributions and allele frequencies for ACE I/D polymorphisms were significantly different between the individuals with and without endometriosis/leiomyoma. The ACE*I-related genotype and allele are strongly related to the occurrence of endometriosis, compared to being moderately correlated with the occurrence of leiomyoma. Our results firstly indicated the ACE gene polymorphisms might predispose to endometriosis or leiomyoma developments. This finding is also compatible with some previous reports (Table 3), who suggested that the ACE*I allele might be associated with higher susceptibility of individual diseases. Therefore, our data strongly suggests that the ACE gene polymorphisms might substantially contribute to the pathogenesis of endometriosis or leiomyoma. It also suggests that RAS might be involved in the pathogenesis of these diseases.

The mechanisms of SNPs upon individual disease remain uncertain. Despite the SNPs do not alter the transcript levels, some investigator demonstrated the disequilibrium effects of certain genotypes might influence the related 3-dimensional structure and efficiency of the transcripts (Shintani et al., 1999; Kennon et al., 2004; Shirasawa et al., 2004). Presumably, the distinct biological condition caused by ACE is among the numerous contributions, which influence the endometriosis developments. These contributions include genetic, dietary, and environmental regulating hormonal and nonhormonal conditions. Furthermore, the ACE polymorphisms might be in linkage disequilibrium with other unidentified functional polymorphisms, which cooperatively influences the endometriosis susceptibility.

In conclusion, associations of ACE*I/D gene polymorphisms with endometriosis and leiomyoma exist. ACE*I related genotype and alleles increase the susceptibility to endometriosis as well as leiomyoma. The ACE gene polymorphisms likely contribute to the pathogenesis of these gynecological diseases. Although the real role and mechanism of ACE gene polymorphism upon these disorders has not yet been clarified, this polymorphism deserves more attention to realize its importance to endometriosis/leiomyoma development. Furthermore, this study could be extended to determine whether the RAS and its related gene polymorphisms also affect the endometriosis or leiomyoma formation. After the clarification of its role upon endometriosis and leiomyoma, ACE gene polymorphism may become a useful marker to predict the future development of these diseases and to permit early therapeutic intervention in women at high risk for endometriosis or leiomyoma.

REFERENCES

Abbud ZA, Wilson AC, Cosgrove NM, Kostis JB. 1998. Angiotensin-converting enzyme gene polymorphism in systemic hypertension. Am J Cardiol 81:244–246.

Alvarez R, Terrados N, Ortolano R, Iglesias-Cubero G, Reguero JR, Batalla A, Cortina A, Fernandez-Garcia B, Rodriguez C, Braga S, Alvarez V, Coto E. 2000. Genetic variation in the renin-angiotensin system and athletic performance. Eur J Appl Physiol 82:117–120.

Anderson TI, Heimdal KR, Skrede M, Tveit K, Berg K, Borresen AL. 1994. Oestrogen receptore (ESR) polymorphisms and breast cancer susceptibility. Hum Genet 94:665–670.

Arkwright PD, Pravica V, Geraghty PJ, Super M, Webb AK, Schwarz M, Hutchinson IV. 2003. End-organ dysfunction in cystic fibrosis: Association with angiotensin I converting enzyme and cytokine gene polymorphisms. Am J Respir Crit Care Med 167:384–389.

Bachman J, Feldmer M, Ganten U, Stock G, Ganten D. 1991. Sexual dimorphism of blood pressure: Possible role of the renin angiotensin system. J Steroid Biochem Mol Biol 40:511–515.

Baudin B. 2002. Angiotensin II receptor polymorphisms in hypertension. Pharmacogenomics considerations. Pharmacogenomics 3: 65–73.

Berge KE, Berg K. 1994. No effect of insertion/deletion polymorphism at the ACE locus on normal blood pressure level or variability. Clin Genet 45:169–174.

Bloem LJ, Manatunga AK, Pratt JH. 1996. Racial difference in the relationship of an angiotensin I-converting enzyme gene polymorphism to serum angiotensin I-converting enzyme activity. Hypertension 27:62–66.

Boehm KD, Daimon M, Gorodeski IG, Sheean LA, Utian WH, Ilan J. 1990. Expression of the insulin-like and platelet-derived growth factor genes in human uterine tissues. Mol Reprod Dev 27:93–101.

Braileanu GT, Simasko SM, Hu J, Assiri A, Mirando MA. 2001. Effects of arginine- and lysine-vasopressin on phospholipase C activity, intracellular calcium concentration and prostaglandin F2alpha secretion in pig endometrial cells. Reproduction 121:605–612.

Braileanu GT, Simasko SM, Speth RC, Daubert DC, Hu J, Mirando MA. 2002. Angiotensin II increases intracellular calcium concentration in pig endometrial stromal cells through type 1 angiotensin receptors, but does not stimulate phospholipase C activity or prostaglandin F2alpha secretion. Reprod Fertil Dev 14:199–205.

Cambien F. 1994. The angiotensin-converting enzyme (ACE) genetic polymorphism: Its relationship with plasma ACE level and myocardial infarction. Clin Genet 46:94–101.

Chang HR, Cheng CH, Shu KH, Chen CH, Lian JD, Wu MY. 2003. Study of the polymorphism of angiotensinogen, anigiotensin-converting enzyme and angiotensin receptor in type II diabetes with end-stage renal disease in Taiwan. J Chin Med Assoc 66:51–56.

Choi YH, Kim JH, Kim DK, Kim JW, Kim DK, Lee MS, Kim CH, Park SC. 2003. Distributions of ACE and APOE polymorphisms and their relations with dementia status in Korean centenarians. J Gerontol A Biol Sci Med Sci 58:227–231.

Cramer DW. 1992. Epidemiology of myomas. Semin Reprod Endocrinol 10:320–324.

Di Lieto A, De Rosa G, Albano G, Staibano S, Campanile M, Pontillo M, Gallo F, Micalef R, Paladini A. 1997. Influence of goserelin depot therapy on epidermal growth factor receptor expression in pelvic endometriosis. Gynecol Endocrinol 11:17–20.

Di Lieto A, De Falco M, Pollio F, Mansueto G, Salvatore G, Somma P, Ciociola F, De Rosa G, Staibano S. 2005. Clinical response, vascular change, and angiogenesis in gonadotropin-releasing hormone analogue-treated women with uterine myomas. J Soc Gynecol Investig 12:123–128.

Dinh D, Frauman AG, Johnston CI, Fabiani ME. 2001. Angiotensin receptors: Distribution, signaling and function. Clin Sci 100: 481–92.

Dixit M, Mansur A, Dixit N, Gilman J, Santarina L, Glicklich D. 2002. The role of ACE gene polymorphism in rapidity of progression of focal segmental glomerulosclerosis. J Postgrad Med 48:266–269.

Donnez J, Smoes P, Gillerot S, Casanas-Roux F, Nisolle M. 1998. Vascular endothelial growth factor (VEGF) in endometriosis. Hum Reprod 13:1686–1690.

Doria A, Warram JH, Rich SS, Krolewski AS. 1994. Angiotensin I-converting enzyme (ACE): Estimation of DNA haplotypes in unrelated individuals using denaturing gradient gel blots. Hum Genet 94:117–123.

El-Badry OM, Helman LJ, Chatten J, Steinberg SM, Evans AE, Israel MA. 1991. Insulin-like growth factor II-mediated proliferation of human neuroblastoma. J Clin Invest 87:648–657.

Fornage M, Amos CI, Kardia S, Sing CF, Turner ST, Boerwinkle E. 1998. Variation in the region of the angiotensin-converting enzyme gene influences interindividual differences in blood pressure levels in young white males. Circulation 97:1773–1779.

Foy CA, Rice GI, Ossei-Gerning N, Mansfield MW, Grant PJ. 1997. Angiotensin-converting enzyme (ACE) gene polymorphisms in patients characterised by coronary angiography. Hum Genet 100: 420–425.

Freitas-Silva M, Pereira D, Coelho C, Bicho M, Lopes C, Medeiros R. 2004. Angiotensin I-converting enzyme gene insertion/deletion polymorphism and endometrial human cancer in normotensive and hypertensive women. Cancer Genet Cytogenet 155:42–46.

Goldman MB, Cramer DW. 1989. Current concepts in endometriosis. Prog Clin Biol Res 323:17–23.

Haiman CA, Henderson SO, Bretsky P, Kolonel LN, Henderson BE. 2003. Genetic variation in angiotensin I-converting enzyme (ACE) and breast cancer risk: The multiethnic cohort. Cancer Res 63:6984–6987.

Harrap SB, Davidson HR, Connor JM, Soubrier F, Corvol P, Fraser R, Foy CJ, Watt GC. 1993. The angiotensin I converting enzyme gene and predisposition to high blood pressure. Hypertension 21:455–460.

Henskens LH, Spiering W, Stoffers HE, Soomers FL, Vlietinck RF, de Leeuw PW, Kroon AA. 2003. Effects of ACE I/D and AT1R-A1166C polymorphisms on blood pressure in a healthy normotensive primary care population: First results of the Hippocates study. J Hypertens 21:81–86.

Hii SI, Nicol DL, Gotley DC, Thompson LC, Green MK, Jonsson JR. 1998. Captopril inhibits tumour growth in a xenograft model of human renal cell carcinoma. Br J Cancer 77:880–883.

Hiramori K. 1994. Treatment of acute myocardial infarction. Nippon Naika Gakkai Zasshi 83:455–459.

Hsieh YY, Chang CC, Tsai FJ, Hsu CM, Lin CC, Tsai CH. 2005. Angiotensin I-converting enzyme ACE 2350*G and ACE-240*T-related genotypes and alleles are associated with higher susceptibility to endometriosis. Mol Hum Reprod 11:11–14.

Inan S, Kuscu NK, Vatansever S, Ozbilgin K, Koyuncu F, Sayhan S. 2003. Increased vascular surface density in ovarian endometriosis. Gynecol Endocrinol 17:143–150.

Irahara M, Uemura H, Yasui T, Kinoshita H, Yamada M, Tezuka M, Kiyokawa M, Kamada M, Aono T. 2001. Efficacy of every-other-day administration of conjugated equine estrogen and medroxyprogesterone acetate on gonadotropin-releasing hormone agonists treatment in women with endometriosis. Gynecol Obstet Invest 52: 217–222.

Isbir T, Yilmaz H, Agachan B, Aydin M, Isbir CS. 1999. Association between angiotensin-converting enzyme gene polymorphism and coronary artery disease. IUBMB Life 48:205–207.

Kehoe PG, Katzov H, Feuk L, Bennet AM, Johansson B, Wiman B, de Faire U, Cairns NJ, Wilcock GK, Brookes AJ, et al. 2003. Haplotypes extending across ACE are associated with Alzheimer's disease. Hum Mol Genet 12:859–867.

Kennon B, Ingram MC, Friel EC, Anderson NH, MacKenzie SM, Davies E, Shakerdi L, Wallace AM, Fraser R, Connell JM. 2004. Aldosterone synthase gene variation and adrenocortical response to sodium status, angiotensin II and ACTH in normal male subjects. Clin Endocrinol (Oxf) 61:174–181.

Koh WP, Yuan JM, Sun CL, van den Berg D, Seow A, Lee HP, Yu MC. 2003. Angiotensin I-converting enzyme (ACE) gene polymorphism and breast cancer risk among Chinese women in Singapore. Cancer Res 63:573–578.

Lever AF, Hole DJ, Gillis CR, McCallum IR, McInnes GT, MacKinnon PL, Meredith PA, Murray LS, Reid JL, Robertson JW. 1998. Do inhibitors of angiotensin-I-converting enzyme protect against risk of cancer? Lancet 352:179–184.

Li C, Ansari R, Yu Z, Shah D. 2000. Definitive molecular evidence of reninangiotensin system in human uterine decidual cells. Hypertension 36:159–164.

Martinez E, Puras A, Escribano J, Sanchis C, Carrion L, Artigao M, Divison JA, Masso J, Vidal A, Fernandez JA. 2000. Angiotensin-converting enzyme (ACE) gene polymorphisms, serum ACE activity and blood pressure in a Spanish-Mediterranean population. J Hum Hypertens 14:131–135.

Matsubara M, Suzuki M, Fujiwara T, Kikuya M, Metoki H, Michimata M, Araki T, Kazama I, Satoh T, Hashimoto J, Hozawa A, Ohkubo T, Tsuji I, Katsuya T, Higaki J, Ogihara T, Satoh H, Imai Y. 2002. Angiotensin-converting enzyme I/D polymorphism and hypertension: The Ohasama study. J Hypertens 20:1121–1126.

McKenzie CA, Abecasis GR, Keavney B, Forrester T, Ratcliffe PJ, Julier C, Connell JM, Bennett F, McFarlane-Anderson N, Lathrop GM, et al. 2001. Trans-ethnic fine mapping of a quantitative trait locus for circulating angiotensin I-converting enzyme (ACE). Hum Mol Genet 10:1077–1084.

Medeiros R, Vasconcelos A, Costa S, Pinto D, Lobo F, Morais A, Oliveira J, Lopes C. 2004. Linkage of angiotensin I-converting enzyme gene insertion/deletion polymorphism to the progression of human prostate cancer. J Pathol 202:330–335.

Mello G, Parretti E, Gensini F, Sticchi E, Mecacci F, Scarselli G, Genuardi M, Abbate R, Fatini C. 2003. Maternal-fetal flow, negative events, and preeclampsia: Role of ACE I/D polymorphism. Hypertension 41:932–937.

Mondorf UF, Russ A, Wiesemann A, Herrero M, Oremek G, Lenz T. 1998. Contribution of angiotensin I converting enzyme gene polymorphism and angiotensinogen gene polymorphism to blood pressure regulation in essential hypertension. Am J Hypertens 11:174–183.

Moore JH, Lamb JM, Brown NJ, Vaughan DE. 2002. A comparison of combinatorial partitioning and linear regression for the detection of epistatic effects of the ACE I/D and PAI-1 4G/5G polymorphisms on plasma PAI-1 levels. Clin Genet 62:74–79.

Moriyama T, Kitamura H, Ochi S, Izumi M, Yokoyama K, Yamauchi A, Ueda N, Kamada T, Imai E. 1995. Association of angiotensin I-converting enzyme gene polymorphism with susceptibility to antiproteinuric effect of angiotensin I-converting enzyme inhibitors in patients with proteinuria. J Am Soc Nephrol 6:1676–1678.

Nakai K, Itoh C, Miura Y, Hotta K, Musha T, Itoh T, Miyakawa T, Iwasaki R, Hiramori K. 1994. Deletion polymorphism of the angiotensin I-converting enzyme gene is associated with serum ACE concentration and increased risk for CAD in the Japanese. Circulation 90:2199–2202.

Omran H, Haffner K, Vollmer M, Pigulla J, Wagner G. Caridi G. Hildebrandt F. 1999. Exclusion of the candidate genes ACE and Bcl-2 for six families with nephronophthisis not linked to the NPH1 locus. Nephrol Dial Transplant 14:2328–2331.

Parsa A, Peden E, Lum RF, Seligman VA, Olson JL, Li H, Seldin MF, Criswell LA. 2002. Association of angiotensin-converting enzyme polymorphisms with systemic lupus erythematosus and nephritis: Analysis of 644 SLE families. Genes Immun 1:S42–S46.

Reddy MK, Baskaran K, Molteni A. 1995. Inhibitors of angiotensin-converting enzyme modulate mitosis and gene expression in pancreatic cancer cells. Proc Soc Exp Biol Med 210:221–226.

Rieder MJ, Taylor SL, Clark AG, Nickerson DA. 1999. Sequence variation in the human angiotensin converting enzyme. Nat Genet 22:59–62.

Sayed-Tabatabaei FA, Houwing-Duistermaat JJ, van Duijn CM, Witteman JC. 2003. Angiotensin-converting enzyme gene polymorphism and carotid artery wall thickness: A meta-analysis. Stroke 34:1634–1639.

Schauser KH, Nielsen AH, Dantzer V, Poulsen K. 2001. Angiotensin-converting enzyme activity in the bovine uteroplacental unit changes in relation to the cycle and pregnancy. Placenta 22:852–862.

Schurmann M, Reichel P, Muller-Myhsok B, Dieringer T, Wurm K, Schlaak M, Muller-Quernheim J, Schwinger E. 2001. Angiotensin-converting enzyme (ACE) gene polymorphisms and familial occurrence of sarcoidosis. J Intern Med 249:77–83.

Sethi AA, Nordestgaard BG, Tybjaerg-Hansen A. 2003. Angiotensinogen gene polymorphism, plasma angiotensinogen, and risk of hypertension and ischemic heart disease: A meta-analysis. Arterioscler Thromb Vasc Biol 23:1269–1275.

Shintani S, Matsuo K, Crohin CC, McBride J, Tsuji T, Donoff RB, Posner M, Todd R, Wong DT. 1999. Intragenic mutation analysis of the human epidermal growth factor receptor (*EGFR*) gene in malignant human oral keratinocytes. Cancer Res 59:4142–4147.

Shirasawa S, Harada H, Furugaki K, Akamizu T, Ishikawa N, Ito K, Ito K, Tamai H, Kuma K, Kubota S, et al. 2004. SNPs in the promoter of a B cell-specific antisense transcript, SAS-ZFAT, determine susceptibility to autoimmune thyroid disease. Hum Mol Genet 13:2221–2231.

Small W, Molteni A, Kim YT, Taylor JM, Chen Z, Ward WF. 1997. Captopril modulates hormone receptor concentration and inhibits proliferation of human mammary ductal carcinoma cells in culture. Breast Cancer Res Treat 44:217–224.

Strathy JH, Molgaard CA, Coulam CB, Melton LJ III. 1982. Endometriosis and infertility: A laparoscopic study of endometriosis among fertile and infertile women. Fertil Steril 38:667–672.

Strawn EY Jr, Novy MJ, Burry KA, Bethea CL. 1995. Insulin-like growth factor I promotes leiomyoma cell growth in vitro. Am J Obstet Gynecol 172:1837–1844.

Tamura T, Johanning GL, Goldenberg RL, Johnston KE, DuBard MB. 1996. Effect of angiotensin-converting enzyme gene polymorphism on pregnancy outcome, enzyme activity, and zinc concentration. Obstet Gynecol 88:497–502.

Toblli JE, Cao G, DeRosa G, Di Gennaro F, Forcada P. 2004. Angiotensin-converting enzyme inhibition and angiogenesis in myocardium of obese Zucker rats. Am J Hypertens 17:172–180.

Treloar SA, O'Connor DT, O'Connor VM, Martin NG. 1999. Genetic influences on endometriosis in an Australian twin sample. Fertil Steril 71:701–710.

Um JY, An NH, Kim SH, Lee KM, Kim YS, Jang H, Cho KH, Moon BS, Kim HM. 2003. Genetic susceptibility to ischemic cerebrovascular disease in Koreans. J Mol Neurosci 20:31–38.

Vigano P, Gaffuri B, Somigliana E, Busacca M, Di Blasio AM, Vignali M. 1998. Expression of intercellular adhesion molecule (ICAM)-1 mRNA and protein is enhanced in endometriosis versus endometrial stromal cells in culture. Mol Hum Reprod 4:1150–1156.

Volpert OV, Ward WF, Lingen MW, Chesler L, Solt DB, Johnson MD, Molteni A, Polverini PJ, Bouck NP. 1996. Captopril inhibits angiogenesis and slows the growth of experimental tumors in rats. J Clin Invest 98:671–679.

Walther T, Menrad A, Orzechowski HD, Siemeister G, Paul M, Schirner M. 2003. Differential regulation of in vivo angiogenesis by angiotensin II receptors. FASEB J 17:2061–2067.

West MJ, Summers KM, Wong KK, Burstow DJ. 1997. Renin-angiotensin system gene polymorphisms and left ventricular hypertrophy. The case against an association. Adv Exp Med Biol 432:117–122.

Yamamoto K, Kataoka S, Hashimoto N, Kakihara T, Tanaka A, Kawasaki T, kikuchi T, Takahashi H, Uchiyama M. 1997. Serum level and gene polymorphism of angiotensin I converting enzyme in Japanese children. Acta Paediatr Jpn 39:1–5.

Yasumatsu R, Nakashima T, Masuda M, Ito A, Kuratomi Y, Nakagawa T, Komune S. 2004. Effects of the angiotensin-I converting enzyme inhibitor perindopril on tumor growth and angiogenesis in head and neck squamous cell carcinoma cells. J Cancer Res Clin Oncol 130: 567–573.

Yildiz A, Yazici H, Cine N, Kazancioglu R, Akkaya V, Sever MS, Ark E, Erginel-Unaltuna N. 2003. Angiotensin converting enzyme gene polymorphism and development of post-transplant erythrocytosis. J Nephrol 16:399–403.

Zee RY, Lou YK, Griffiths LR, Morris BJ. 1992. Association of a polymorphism of the angiotensin I-converting enzyme gene with essential hypertension. Biochem Biophys Res Commun 184:9–15.

Zhu X, Bouzekri N, Southam L, Cooper RS, Adeyemo A, McKenzie CA, Luke A, Chen G, Elston RC, Ward R. 2001. Linkage and association analysis of angiotensin I-converting enzyme (ACE)-gene polymorphisms with ACE concentration and blood pressure. Am J Hum Genet 68:1139–1148.