The American Journal of Chinese Medicine, Vol. 33, No. 3, 501–505

Institute for Advanced Research in Asian Science and Medicine

Novel Approach of Molecular Genetic Understanding of Iridology: Relationship Between Iris Constitution and Angiotensin Converting Enzyme Gene Polymorphism

Jae-Young Um,[*] Nyeon-Hyoung An,[‡] Gui-Bi Yang,[§] Geon-Mok Lee,[§] Ju-Jang Cho,[§] Jae-Woon Cho,[§] Woo-Jun Hwang,[§] Han-Jung Chae,[†] Hyung-Ryong Kim,[¶] Seung-Heon Hong[‡] and Hyung-Min Kim[*]

[*]*Department of Pharmacology*, *College of Oriental Medicine*
Kyung Hee University, *Seoul*, *130-701*, *South Korea*
[†]*Department of Pharmacology and Cardiovascular Research Institute*, *School of Medicine*
Chonbuk National University, *South Korea*
[‡]*College of Pharmacy*, [§]*Professional Graduate School of Oriental Medicine*
[¶]*Department of Dental Pharmacology and Nanoscience and Technology Research Institute*
School of Dentistry, *Wonkwang University*, *Iksan*, *Jeonbuk*, *570-749*, *South Korea*

Abstract: Iridology is the study of the iris of the eye to detect the conditions of the body and its organs, genetic strengths and weaknesses, etc. Although iridology is not widely used as a scientific tool for healthcare professionals to get to the source of people's health conditions, it has been used as a supplementary source to help the diagnosis of medical conditions by noting irregularities of the pigmentation in the iris among some Korean Oriental medical doctors. Angiotensin converting enzyme (ACE) gene polymorphism is one of the most well studied genetic markers of vascular disease. We investigated the relationship between iridological constitution and ACE polymorphism in hypertensives. We classified 87 hypertensives and 79 controls according to iris constitution and determined the ACE genotype of each individual. DD genotype was more prevalent in patients with a neurogenic constitution than in controls. This finding supports the hypothesis that D allele is a candidate gene for hypertension and demonstrates the association among ACE genotype, Korean hypertensives and iris constitution.

Keywords: Angiotensin Converting Enzyme (ACE); Gene; Polymorphism; Iris Constitution.

Correspondence to: Dr. Hyung-Min Kim, Department of Pharmacology, College of Oriental Medicine, Kyung Hee University, 1 Hoegi-dong, Dongdaemun-gu, Seoul, 130-701, Korea. Tel: (+82) 2-961-9448, Fax: (+82) 2-968-1085, E-mail: hmkim@khu.ac.kr

Introduction

Iridology is the study of the iris of the eye to detect the conditions of the body and its organs, genetic strengths and weaknesses, etc. Iridology, developed more than 100 years ago, assumes that all body organs are represented on the surface of the iris via intricate neural connections (Sharan, 1989) and that dysfunction of most organs is marked on the iris, usually as a pigmentary change. The right half of the body is represented in the right iris, the left half in the left iris. Each iris is divided into 60 sectors, and each segment is related to an inner organ or a body function. Although iridology is a popular alternative theory and is received favorably by patients, there is little evidence to support the efficacy of its practice. Up to date, more than 80 publications on the subject of iridology have been reported. However, most of the papers were review articles, comments, and descriptions of the technique. We hypothesized that the predisposition to certain diseases indicated by particular iris constitutions may be due to genetic factors. We then evaluated the diagnostic validity of iridology in terms of genetic factors.

The angiotensin-converting enzyme (ACE) genotype is one of the most well studied genetic markers of cardiovascular risk. Numerous studies have attempted to relate the ACE/DD polymorphism with myocardial infarction and/or coronary artery disease, leading to conflicting results. However, a recent meta-analysis conducted on 15 studies published before 1995 (3394 cases of myocardial infarction and 5479 control subjects) demonstrated a mean odds ratio for myocardial infarction for the DD versus the ID/II genotypes of 1.26. The relative risk appeared to be variable according to ethnicity, and higher in the Japanese population (Samani *et al*., 1996; Carluccio *et al*., 2001). In the previous studies we reported that ACE polymorphism is not a risk factor for the development of cerebral infarction in the Korean population (Um *et al*., 2001), and that the ACE polymorphism was not associated with a major branch of Korean Traditional Medicine, Sasang constitution (Um *et al*., 2003). Also a recent study has shown that ACE/DD homozygosity, in comparison with the other ACE genotypes, was associated with increases in the incidence of hypertension in the Caucasian population (Staessen *et al*., 2001). Therefore, we investigated the association between ACE polymorphism and hypertension with regard to iris constitution.

Materials and Methods

Patients

From January 1999 to July 2003, 87 hypertensive patients aged 28 to 62 years were enrolled from the Oriental Medical Hospital, Wonkwang University, Jeonju, Korea. Hypertension was defined as having a systolic blood pressure of ≥ 140 mmHg or a diastolic blood pressure of ≥ 90 mmHg. The control groups consisted of 79 healthy volunteers, with normal blood pressure. Control subjects were matched to hypertensive subjects, according to age and gender. Determination of the subjects' iris constitutions, including patients and controls, was performed by Professor Woo-Jun Hwang of the Professional Graduate School of Oriental Medicine, Wonkwang University. An automatic iris analysis system,

the Bexel Irina, was used for the assessments of the irises. All hypertensive patients and controls were Korean and gave their informed consent before participating in this study, which was approved by the ethics committee of the hospital.

ACE Genotyping

The blood was stored at –20°C until it was ready to be extracted. The genomic DNA was extracted using inorganic procedure. The ACE polymorphism was detected by PCR amplification. The reaction was run with a sense primer, ACE1: 5′-CATCCTTTCTCCCATTTCTC-3′; an antisense primer, ACE3: 5′-TGGGATTACAGG-CGTGATACAG-3′; and a primer for inserted region (286 bp), ACE2: 5′-ATTTCAGAGCTGGAATAAAATT-3′. These primers allow for the detection of an 86 bp fragment in the absence of the insertion (DD) and two fragments of 64 and 490 bp in the presence of the insertion (II). The 490, 64 and 86 bp fragments yielded by PCR amplification were identified as heterozygous (ID) (Evans *et al*., 1994).

Statistical Analysis

Comparisons of the genotype and allele frequencies of the ACE genotypes between groups were carried out using the Pearson chi-square test. All statistical analyses were performed using SPSS v9.00 (SPSS Inc.) statistical analysis software. A $p < 0.05$ was considered statistically significant.

Results and Discussion

Table 1 shows the characteristics of the present subjects. We classified 87 hypertensive patients and 79 controls according to their iris constitutions, and determined their ACE genotypes. The distribution of ACE genotypes in hypertensive patients were as follows: II, 27 (31.0%); ID, 42 (48.3%); DD, 18 (20.7%). It was significantly different from the distribution in controls: II, 35 (44.3%); ID, 39 (49.4%); DD, 5 (6.3%) (Table 2). DD genotype was more prevalent in hypertensive patients than in controls ($\chi^2 = 8.125$, $df = 2$, $p < 0.05$). The distribution of iris constitutions in the hypertensive patients was significantly different from the distribution in the control group ($\chi^2 = 40.244$, $df = 3$, $p < 0.001$) (Table 3). Especially, the frequency of DD genotype was significantly higher in hypertensive patients, stratified as a neurogenic constitution, than in controls (32.1% versus 7.7%) (Table 4). Furthermore, the frequency of DD genotype was significantly higher in neurogenic (32.1%) and cardio-renal connective tissue weakness constitution (24.3%) than in abdominal connective tissue weakness constitution (0%) (neurogenic versus abdominal connective tissue weakness: $\chi^2 = 14.757$, $df = 2$, $p < 0.005$; cardio-renal connective tissue weakness versus abdominal connective tissue weakness: $\chi^2 = 9.170$, $df = 2$, $p < 0.05$) (Table 4).

Table 1. Characteristics of Hypertensives and Controls

Characteristics	Patients	Controls
n	87	79
Age (year)	57.8 ± 15.4	56.1 ± 9.8
Male (%)	55	55

Values are means ± SD.

Table 2. Distribution of ACE Genotypes in Hypertensives and Controls

	Genotypes			Statistics*
	II	ID	DD	
Control, n (%) (n = 79)	35 (44.3)	39 (49.4)	5 (6.3)	$\chi^2 = 8.125$
Patients, n (%) (n = 87)	27 (31.0)	42 (48.3)	18 (20.7)	$p < 0.05$

*Statistical tests by Pearson χ^2-test (two-sided).

Table 3. Distribution of Iris Constitution

	Iris Constitution				Statistics*
	Neurogenic	Abdominal Connective Tissue Weakness	Cardio-renal Connective Tissue Weakness	The Others	
Control, n (%) (n = 79)	13 (16.5)	31 (39.2)	13 (16.5)	22 (27.8)	$\chi^2 = 40.244$
Patients, n (%) (n = 87)	28 (32.2)	22 (25.3)	37 (42.5)	0(0)	$p < 0.01$

*Statistical tests by χ^2-test (two-sided).

Table 4. Relationship Between Iris Constitution and ACE Genotype in Hypertensives

	Iris Constitution				Statistics	
	Ceurogenic n (%)	Abdominal Connective Tissue Weakness, n (%)	Cardio-renal Connective Tissue Weakness, n (%)	The Others n (%)		
Controls						
II	1 (7.7)	19 (61.3)	3 (23.1)	12 (54.5)	$\chi^2 = 10.612$ $p^* < 0.005$	$\chi^2 = 7.406$ $p^\dagger < 0.05$
ID	11 (84.6)	11 (35.5)	7 (53.8)	10 (45.5)		
DD	1 (7.7)	1 (3.2)	3(23.1)	0 (0)		
Patients						
II	4 (14.3)	13 (59.1)	10 (27.0)	0 (0)	$\chi^2 = 14.757$ $p^* < 0.005$	$\chi^2 = 9.170$ $p^\dagger < 0.05$
ID	15 (53.6)	9(40.9)	18 (48.6)	0 (0)		
DD	9 (32.1)	0 (0)	9 (24.3)	0 (0)		

Statistical tests by χ^2-test (two-sided). *Neurogenic versus abdominal connective tissue weakness. †Cardio-renal connective tissue weakness versus abdominal connective tissue weakness.

Although iridology has been criticized as an unfounded diagnostic tool, many iridologists are presently practicing in many areas. In Germany, 80% of *Heilpraktiker* (non-medically qualified health practitioners) practice iridology (Ernst, 2000). In this study, we investigated the ACE genotypes of hypertensive patients classified by their iris constitutions. As a result, 74.7% of hypertensive patients were neurogenic or cardio-renal connective tissue weakness type. Also, the frequencies of DD genotype were significantly higher in hypertensive patients than in controls. These results are consistent with the reports that DD genotype was associated with hypertension (Staessen *et al*., 2001). Therefore, our results support that D allele is a candidate gene for hypertension, and suggest an apparent relationship between ACE genotype and iris constitutions, as well as the novel possibility of molecular genetic understanding of iridology.

Acknowledgments

This work was supported by Wonkwang University in 2004.

References

Carluccio, M., M. Soccio and R. De Caterina. Aspects of gene polymorphisms in cardiovascular disease: the renin-angiotensin system. *Eur. J. Clin. Invest.* 31: 476–488, 2001.

Ernst, E. Iridology: not useful and potentially harmful. *Arch. Ophthalmol*. 118: 120–121, 2000.

Evans, A.E., O. Poirier, F. Kee, L. Lecerf, E. McCrum, T. Falconer, J. Crane, D.F. O'Rourke and F. Cambien. Polymorphisms of the angiotensin-converting enzyme gene in subjects who die from coronary heart disease. *Q. J. Med*. 87: 211–214, 1994.

Samani, N.J., J.R. Thompson, L. O'Toole, K. Channer and K.L. Woods. A meta-analysis of the association of the deletion allele of the angiotensin-converting enzyme gene with myocardial infarction. *Circulation* 94: 708–712, 1996.

Sharan, F. Iridology: *A Complete Guide to Diagnosing Through the Iris and to Related Forms of Treatment*. Thorsons Publications, Ltd., England, 1989.

Staessen, J.A., J.G. Wang, E. Brand, C. Barlassina, W.H. Birkenhager, S.M. Herrmann, R. Fagard, L. Tizzoni and G. Bianchi. The deletion/insertion polymorphism of the angiotensin converting enzyme gene and cardiovascular-renal risk. *J. Hypertens*. 19: 1349–1358, 2001.

Um, J.Y., J.C. Joo, K.Y. Kim, N.H. An, K.M. Lee and H.M. Kim. Angiotensin converting enzyme gene polymorphism and traditional classification in Koreans with cerebral infarction. *Hereditas* 138(3): 166–171, 2003.

Um, J.Y., H. Kim, T.J. Choi, C.S. Jin, S.T. Park, K.C. Lee, H.S. Rhee, K.M. Lee, Y.M. Lee, H.M. Kim, N.H. An and J.J. Kim. Polymorphism of the angiotensin-converting enzyme gene in patients with cerebral infarction in Koreans. *J. Mol. Neurosci*. 17: 279–283, 2001.