Türk Göğüs Kalp Damar Cerrahisi Dergisi
Turkish Journal of Thoracic and Cardiovascular Surgery

Original Article / Orijinal Makale

Association between deletion polymorphism of angiotensin converting enzyme gene and pulmonary hypertension in pulmonary thromboembolism

Pulmoner tromboembolide pulmoner hipertansiyon ile anjiyotensin dönüştürücü enzim geninin delesyonel polimorfizmi arasındaki bağlantı

Şahin Bozok,[1] Gökhan İlhan,[1] Buğra Destan,[1] Hakan Karamustafa,[1] Sedat Ozan Karakişi,[1] Mustafa Çetin,[2] Nebiye Tüfekçi,[1] Cemal Aslan,[1] Mustafa Emir[1]

Departments of [1]Cardiovascular Surgery, [2]Cardiology, Rize University Faculty of Medicine, Rize Training and Research Hospital, Rize, Turkey

Background: This study aims to identify the association between pulmonary arterial pressure and insertion/deletion (I/D) polymorphism of ACE gene in the patients with pulmonary thromboembolism (PTE) due to various reasons.

Methods: A total of 48 patients (23 females, 25 males; mean age 60±13 years; range 42 to 78 years) were included in the study. Patients with PTE were classified according to carrying of ACE D allele. Group 1 consisted of patients with wild type, while group 2 consisted of patients with ACE D allele carrier.

Results: Tweny-eight patients (58%) had ACE ID (heterozygous) genotype, while six (13%) had ACE DD (homozygous) genotype. The remaining 14 (29%) had no deletion allele of ACE gene. The mean systolic pulmonary arterial pressure (sPAP) was 45.7±17 mmHg in patients with ID genotype, 70.1±20 mmHg in those with DD genotype, and 32.5±9 mmHg in those with II genotype. The comparison of the patients who carried ACE D allele with those who did not demonstrated that the former group had significantly higher levels of sPAP (32.5±8.8 versus 50.8±20 mmHg, p=0.017). It was found that carrying of ACE D allele (Exp(B): 7.331, p=0.032) was found to be independent predictor of pulmonary hypertension in patients with PTE.

Conclusion: In conclusion, we believe that the risk for the development of pulmonary hypertension is higher especially in PTE cases with deletion polymorphism of ACE gene. Therefore, evaluation of the ACE gene in these patients will contribute to shed light into the etiology and prognosis of the disease.

Key words: ACE D allele; ACE I/D gene polymorphism; pulmonary hypertension; pulmonary thromboembolism.

Amaç: Bu çalışmada, değişik nedenlere bağlı pulmoner tromboemboli (PTE) gelişmiş ACE geninin insersiyon/delesyon (I/D) polimorfizmi olan hastalarda bu durum ile pulmoner arteryel basınç arasındaki ilişki araştırıldı.

Çalışma planı: Çalışmaya toplam 48 hasta (23 kadın, 25 erkek; ort. yaş 60±13 yıl; dağılım 42-78 yıl) dahil edildi. Pulmoner tromboembolili hastalar ACE D allel taşıyıp taşımamalarına göre sınıflandırıldı. Grup 1 vahşi tip taşıyıcısı hastalarda oluşurken, grup 2 ACE D alleli taşıyıcısı hastalardan oluşturuldu.

Bulgular: Hastaların 28'inde (%58) ACE ID (heterozigot) genotip, altısında (%13) ACE DD (homozigot) genotip mevcut idi. Kalan 14'ünde (%29) ACE genine ait delesyon alleli yok idi. Ortalama sistolik pulmoner arter basıncı (sPAP) ID genotipli hastalarda 45.7±17 mmHg, DD genotipli hastalarda 70.1±20 mmHg ve genotip II hastalarda 32.5±9 mmHg idi. ACE D allel taşıyan hastalarla taşımayanlar arasında yapılan karşılaştırmada, ACE D allel taşıyıcılığı belirgin olarak daha yüksek sPAP görüldü (32.5±8.8 karşın 50.8±20 mmHg, p=0.017). ACE D allelini taşımak (Exp(B): 7.331, p=0.032) PTE'si olan hastalarda pulmoner hipertansiyonun bağımsız göstergesi olarak bulundu.

Sonuç: Sonuç olarak, ACE geninin delesyon polimorfizmi olan PTE'li olgularda pulmoner hipertansiyonun gelişme riskinin daha yüksek olduğuna inanıyoruz. Bu nedenle, ACE geninin bu hastalarda değerlendirilmesi, hastalığın etyoloji ve prognozunu aydınlatmaya katkıda bulunacaktır.

Anahtar sözcükler: ACE D alleli; ACE I/D gen polimorfizmi; pulmoner hipertansiyon; pulmoner tromboemboli.

Received: November 17, 2011 *Accepted:* January 12, 2012

Correspondence: Şahin Bozok, M.D. Rize Üniversitesi Tıp Fakültesi, Rize Eğitim ve Araştırma Hastanesi, Kalp ve Damar Cerrahisi Anabilim Dalı, 53100 Rize, Turkey. Tel: +90 464 - 217 03 65 e-mail: sahinboz@yahoo.com

Pulmonary thromboembolism (PTE) is a disorder that leads to serious health issues. Among the general population, its frequency is 0.2-0.3%, and about 30% of the cases result in death. Mortality from this disease can be reduced to as low as 2-8% with early diagnosis and timely treatment.[1] For this reason, prompt management of the disease is of utmost importance.

Even though PTE may be encountered in every specialty of medicine, its diagnosis may be missed for several reasons, including the variability of clinical symptoms, resemblance to other diseases, disregard of risk factors, time needed for a precise diagnosis, and failure to consider PTE among the differential diagnoses. Despite advances in its diagnosis and treatment, the clinical approach to patients still bears some difficulties. Fibrinogen, d-dimer, and arterial blood gas measurements may help in the diagnosis of PTE; however, a more conclusive diagnosis can be reached by elaborate methods like spiral computed tomography (CT) and ventilation-perfusion scintigraphy.[2] Pulmonary angiography is the gold standard in diagnosis, yet it is seldom used due to its adverse effects and invasive aspect.[3,4] Computed tomography has a sensitivity rate of 90% and a specificity rate of 96% for the detection of central and lobar pulmonary thromboembolism. However, it may be inadequate to identify segmental or subsegmental emboli, which are better evaluated using CT angiography.[4]

Alongside some hereditary factors disrupting the coagulation balance, potential etiologic factors of PTE include several entities such as immobilization, major surgical procedures, and malignancies.[5] Another reported risk factor for the development of venous emboli is the angiotensin-converting enzyme (ACE) insertion/deletion (I/D) gene polymorphism.[6] The ACE decreases bradykinin, an important mediator for the release of the tissue plasminogen activator (t-PA), which impedes fibrinolysis and increases the risk of thrombosis. The ACE gene located on chromosome 17q23 has an I/D polymorphism. This gives rise to three genotypes: DD, ID, and II.[7,8]

Pulmonary thromboembolism may lead to a number of complications with various severities. These include pneumonia, hemoptysis, pleural effusion, syncope, right-sided heart failure, and cardiogenic shock.[9,10] Pulmonary hypertension (PHT) is a significant factor which complicates the clinical picture and decreases a patient's quality of life.

In this study, we aimed to investigate the association between the ACE I/D gene polymorphism and the development of PHT following PTE.

PATIENTS AND METHODS

Patient population and study design

A total of 80 consecutive patients admitted to the emergency unit between July 2007 and July 2011 who were hospitalized due to suspicion of PTE were prospectively considered for enrollment in our study. Twenty patients who had previously diagnosed with chronic obstructive pulmonary disease (COPD), malignancies, coronary artery disease, severe left ventricular valve diseases, or left ventricular heart failure and 12 patients diagnosed with PTE via scintigraphy and/or tomography (alternative diagnoses were confirmed) were excluded from the study. A total of 48 patients (23 females, 25 males; mean age 60±13 years; range 42 to 78 years) whose diagnoses were confirmed as PTE by ventilation-perfusion scintigraphy and/or multi-slice spiral CT according to ESC guideline recommendations[11] were enrolled in the study after obtaining informed consent. Patients with PTE were classified according to how they carried the ACE D allele. Group 1 consisted of patients with the wild type, and group 2 was composed of ACE D allele carriers. Data collection per protocol included symptoms at admission to the emergency unit, predisposing conditions, history of diabetes mellitus, hypertension, and vital signs at the time of admission to the emergency unit as well as the results of several diagnostic procedures, including blood gas analysis and other blood parameters, an electrocardiogram (ECG), transthoracic echocardiography (ECO), a lower extremity Doppler ultrasound examination, contrast-enhanced spiral CT, ventilation-perfusion scintigraphy, and duration of hospitalization. Hypertension was defined as blood pressure >140/90 mmHg on more than two occasions during office measurements or being on an antihypertensive treatment. Diabetes mellitus was defined as fasting blood sugar ≥126 mg/dL or being on an antidiabetic treatment. Rhythm and electrocardiographic findings of right ventricle loading, including S1Q3T3, right bundle branch block pattern, and T wave changes on right precordial derivations, were evaluated. Per protocol, our center was advised to perform transthoracic ECO within 24 hours of admission. Echocardiographic examinations were performed via the Vivid 7 system (GE Healthcare, Wauwatosa, Wisconsin, USA) with 2.5-5MHz probes. The ejection fraction (EF) was calculated by the modified Simpson's method. Chamber sizes were defined according to recent ESC guidelines.[12] In order to evaluate right ventricular (RV) dysfunction, the presence of RV dilatation and

systolic pulmonary artery pressure (SPAP) were evaluated on ECO. Right ventricular dimensions were evaluated according to the most recent ESC guidelines[12] using an RV dimension >3.4 cm at the basal plane or >3.8 cm at the midplane to designate RV dilation. Systolic pulmonary artery pressure was calculated as previously discussed.[12] Digital records of transthoracic echocardiographic examinations without recorded identities were evaluated offline by an expert echocardiographer blinded to the study plan. The study was performed in accordance with the Declaration of Helsinki for Human Research and was approved by the institutional review board.

Statistical analysis

Parametric data was expressed as mean ± standard deviation (SD) or median (range) and categorical data as percentages. Statistical procedures were performed using the Statistical Package for Social Sciences software version 15.0 (SPSS Inc., Chicago, Illinios, USA). Independent parameters were compared via an independent samples t-test, and the Mann-Whitney U test was used to test parametric data without binomial distribution. Multivariable logistic regression was used to evaluate independent parameters affecting high SPAP (≥35 mmHg). A p value ≤0.05 was considered significant.

Genotyping

Blood samples from the subjects were placed into ethylenediaminetetraacetic acid (EDTA) tubes and stored at -20 °C. Genomic DNA analysis of the peripheral blood sample was executed using the Invisorb Spin Stool DNA Kit (Invitek, Berlin, Germany). For multiplex amplification of the ACE gene, CVD StripAssays (ViennaLab, Labordiagnostika GmbH, Vienna, Austria) was utilized. Polymerase chain reaction (PCR) products (4 ml) obtained before hybridization were assessed for successful amplification in 1% electrophoresis. For reverse hybridization analysis executed with ProfiBlot T48 (Tecan, Switzerland), 10 ml of the PCR products with successful amplification were used. Groups were divided to three subgroup as "the wild (II) group; pure I carrier, the heterozygote (ID) group; I and D carrier, homozygote mutant (DD) group; pure D carrier" for more specify the allele effect.

RESULTS

Fourteen (29%) patients had ACE II (wild type), 28 (58%) had ACE ID (heterozygote mutant), and six (13%) had ACE DD (homozygote mutant) genotypes.

The baseline characteristics of patients with PTE were classified into two categories according to how they carried the D allele, and this information is presented in Table 1. There was no statistically significant difference between the two groups in terms of age, gender, body mass index, admission symptoms, presence of risk factors, such as hypertension, diabetes mellitus, and smoking, or predisposing conditions, for example immobilization, previous history of PTE, and surgical intervention or trauma within 14 days.

A comparison of the two subgroups of patients with PTE along with the electrocardiography and ECO parameters and laboratory findings are also summarized in Table 1. The mean systolic pulmonary artery pressure was 32.5±9 mmHg in the wild type group and 50.8±20 mmHg in the ACE D allele carrier group (p=0.017, Table 1 and Figure 1). On the other hand, in patients with PTE, the mean SPAP was 32.5±9 mmHg in the wild (II) subgroup, 45.7±17 mmHg in the heterozygote (ID) subgroup, and 70.1±20 mmHg in the homozygote mutant (DD) subgroup. The difference among the three groups was statistically significant (p<0.001, Figure 2). Also, troponin levels (0.01±001 versus 0.02±0.02 ng/mL, p=0.018) were statistically different among the two allele carriage groups in patients with PTE. However, other ECO and laboratory parameters and electrocardiography findings did not different in either of the allele carriage groups (Table 1).

The clinical and laboratory findings of patients with PTE were classified into two categories according to the presence of PHT (SPAP ≥35 mmHg). The presence of atrial fibrillation was higher in those who had PHT compared with those who did not [0 (%) versus 8 (29%), p=0.014]. Patients with PHT were also more likely to carry the ACE D allele [10 (50%) versus 24 (86%), p=0.018], and the d-dimer levels were found to be significantly higher in patients who had PHT when compared with those who did not (702±499 versus 1638±2064 ng/ml, p=0.042).

Univariate predictors of mortality were enrolled into multivariable logistic regression analysis. Carrying the D allele [Exp(B): 7.331, p=0.032] was found to be an independent predictor of PHT in patients with PTE.

DISCUSSION

Pulmonary thromboembolism may lead to right-sided ventricular dysfunction and cardiogenic shock. Moreover, it still has a grave mortality rate due to alterations it causes in clinical and hemodynamic parameters.[13] Various techniques, such as spiral CT and ventilation/perfusion scintigraphy, can be used to diagnose PTE. Although it is the gold standard in the diagnosis of PTE, pulmonary angiography is not a preferred method (unless inevitably required)

Table 1. Baseline characteristics of pulmoner tromboembolizm patients grouped into two categories according to carrying of D allele

	Wild type (n=14)			D allele carrier (n=34)			
	n	%	Mean±SD	n	%	Mean±SD	*p*
Age (age)			60±12			60±14	0.958
Female gender	6	43		17	50		0.895
Body mass index			30±8			26±4	0.183
Admission symptoms							
Dyspnea	13	93		31	91		0.848
Chest pain	8	57		27	79		0.157
Hemoptysis	2	14		4	12		0.810
Syncope	1	7		4	12		0.634
Hypertension	5	36		11	32		0.822
Diabetes mellitus	4	29		7	21		0.708
Smoking	6	43		14	41		0.915
Presence of deep vein thrombosis	3	21		5	15		0.676
Immobilization	3	21		10	29		0.728
Previous history of pulmonary embolism	1	8		4	12		0.685
Previous history of surgery or trauma	2	14		3	9		0.621
O_2 saturation (%)			87±7			81±9	0.075
Electrocardiography parameters							
Atrial fibrillation	2	14		6	18		0.776
Right bundle branch block	0	0		5	16		0.301
S1Q3T3	1	8		5	16		0.656
T wave changes	1	8		4	12		0.642
Echocardiography parameters							
Left ventricle ejection fraction (%)			58±4			56±11	0.545
Right ventricle dilatation/hypokinesia	4	29		19	56		0.160
Systolic PA pressure (mmHg)			32.5±9			50.8±20	0.017
Troponin I (ng/mL)			0.01±001			0.02±0.02	0.018
D-dimer (mg/mL)			744±505			1488±1950	0.223
Alanine transaminase (IU/L)			27±17			30±25	0.699
Aspartat transaminase (IU/L)			29±15			30±15	0.905

PA: Pulmonary artery.

due to its invasive nature and possible adverse effects.[3,4] In identification of central and lobar pulmonary thromboembolism, CT has a sensitivity of 90% and a specificity of 96%; however it may be insufficient for the diagnosis of subsegmental PTE. In this case, CT angiography is more necessary.[4] On the other hand, ECO is an efficient method for the diagnosis and follow-up of right-sided ventricular dysfunction, which is an important factor in determining the prognosis. It is also a reliable method to identify and follow precapillary or postcapillary PHT.[13] We assessed the cardiac functions of PTE patients diagnosed with thin-section thoracoabdominal CT angiography and excluded the ones with left-sided ventricular disease or mitral valve disorders. All the patients included in this study underwent a detailed ECO, and their SPAP levels were measured.

Pulmonary hypertension is defined as pulmonary artery pressure (PAP) higher than 25 mmHg at rest and 30 mmHg during exercise.[14] Pulmonary artery pressure may be measured with invasive and non-invasive techniques. Echocardiography is a non-invasive, reliable method which has a sensitivity rate of 90% and a specificity rate of 85% in the evaluation of right-sided ventricular functions.[15] All 48 patients included in this study underwent ECO for PAP assessment and were classified according to their ACE I/D polymorphism status (Table 1).

A number of possible reasons for the development of PHT were investigated; however, no direct cause was disclosed. Some of the possible etiologic factors are cardiac dysfunction, chronic obstructive lung diseases, thromboembolic events, and vascular pathologies. Another suggested factor is thrombotic gene mutation,

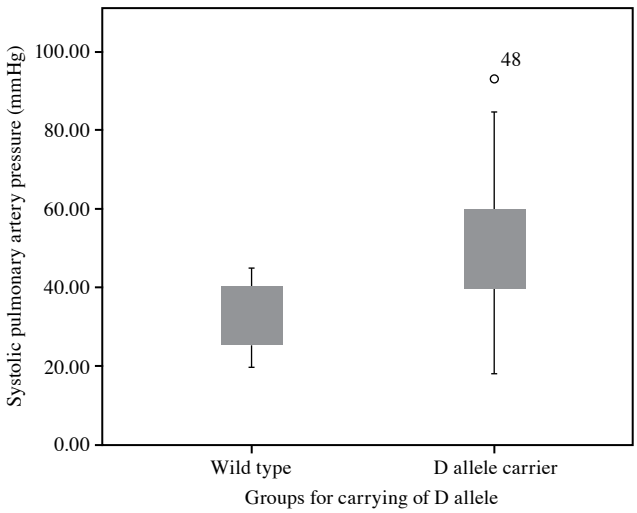

Figure 1. Comparison of systolic pulmonary artery pressure between groups for carrying of D allele.

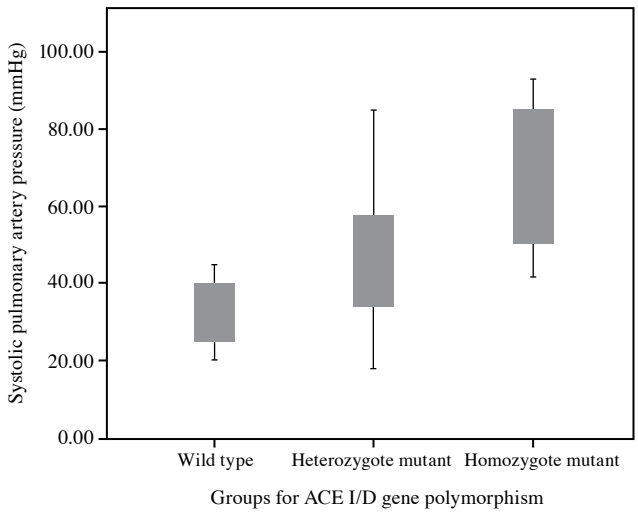

Figure 2. Comparison of systolic pulmonary artery pressure between groups for ACE I/D gene polymorphsim.

such as Factor V Leiden, which has been proven to deteriorate homeostasis and has been blamed directly or indirectly for thrombotic events like PTE.[16] The ACE gene has a prothrombotic function. The renin-angiotensin system is a complex mediator which affects blood pressure, homeostasis, cardiovascular remodeling, and vascular tone. It includes angiotensinogen, ACE, angiotensin II, and several key proteins including receptors for these items. By the action of ACE on epithelial cells, angiotensin I is converted to angiotensin II which, in turn, stimulates plasminogen activator inhibitor 1 (PAI-1) which is responsible for the down regulation of fibrinolysis. Bradykinin, an important mediator for the release of t-PA, is diminished by ACE. This hampers fibrinolysis and increases the risk of thrombosis. The ACE gene is located on chromosome 17q23, and the ACE gene I/D polymorphism results from the insertion or deletion of a repeated Alu sequence at intron 16 and leads to the formation of the DD, ID, and II genotypes. The DD genotype exhibits higher ACE activity. Studies performed on diverse ethnic groups implied that the deletion polymorphism of the ACE gene may be involved in venous thrombosis.[7,8,17] The ACE plays a significant role in vascular homeostasis through angiotensin II modeling and bradykinin inhibition. It has been reported that the ACE I/D genotype accounts for half of the phenotypic variance of serum ACE, and the ACE/DD genotype has been associated with higher levels of serum ACE in the literature. Positive associations between the DD genotype and hypertension have been reported in previous studies.[18,19] Some earlier studies suggested that the ACE gene may be associated with PHT in patients with chronic obstructive lung disease and with right-sided ventricular dysfunction.[20,21] In another study, Tanabe et al.[22] reported that the ACE D allele carrier status might be one of the prognostic factors for medically treated thrombotic PHT patients. We evaluated the patients with PHT following PTE for the ACE gene polymorphism and calculated the mean PAP levels of the groups formed according to the different genotypes (Table 1). The mean PAP level in patients with no deletion polymorphism was found to be lower than the PAP levels in those with heterozygous (ID) and homozygous (DD) deletion polymorphisms. It was especially noteworthy that a comparison of the DD genotype, which had the highest level of mean PAP, with the other groups yielded a statistical significance (Figure 2).

To conclude, PTE is a potentially grave disease that may reach high mortality rates despite advances in diagnosis and treatment. In turn, PHT is a complication that may result from PTE, and it seriously deteriorates a patient's quality of life. If PTE is diagnosed early and treated in a timely manner, these consequences may be partially avoided. Although larger and more comprehensive trials should be undertaken, we believe that our research may provide the impetus for the further studies needed to clarify the etiology of PTE.

There are two noteworthy limitations of this study. First, the phenotypic reflections, such as plasma renin activity, plasma renin concentration, and angiotensin II levels of the ACE gene polymorphisms, could not be evaluated. In addition, single gene deletions are not usually reproducible. Because of this, the study was planned as a pilot study, and the phenotypic observations will be controlled in further studies.

Declaration of conflicting interests

The authors declared no conflicts of interest with respect to the authorship and/or publication of this article.

Funding

The authors received no financial support for the research and/or authorship of this article.

REFERENCES

1. Oner F, Topu Z, Celik G, Gurkan O, Atasoy C, Kucuk O, et al. Validation of a non-invasive diagnostic algorithm in assessment of patients with high pretest clinical probability of pulmonary embolism. Turkish Thoracic Journal 2004;5:26-31.
2. Castaner E, Gallardo X, Ballesteros E, Andreu M, Pallardo Y, Mata JM, et al. CT diagnosis of chronic pulmonary thromboembolism. Radiographics 2009;29:31-50.
3. Kearon C. Diagnosis of pulmonary embolism. CMAJ 2003;168:183-94.
4. Pipavath SN, Godwin JD. Acute pulmonary thromboembolism: a historical perspective. AJR Am J Roentgenol 2008;191:639-41.
5. Tsai AW, Cushman M, Rosamond WD, Heckbert SR, Polak JF, Folsom AR. Cardiovascular risk factors and venous thromboembolism incidence: the longitudinal investigation of thromboembolism etiology. Arch Intern Med 2002;162:1182-9.
6. Wells PS, Rodger MA, Forgie MA, Langlois NJ, Armstrong L, Carson NL, et al. The ACE D/D genotype is protective against the development of idiopathic deep vein thrombosis and pulmonary embolism. Thromb Haemost 2003;90:829-34.
7. Munhoz TP, Scheibe RM, Schmitt VM. Angiotensin converting enzyme (ACE) DD genotype: relationship with venous thrombosis. Rev Bras Hematol Hemoter 2005;27:87-90.
8. Niemiec P, Zak I, Wita K. The D allele of angiotensin I-converting enzyme gene insertion/deletion polymorphism is associated with the severity of atherosclerosis. Clin Chem Lab Med 2008;46:446-52.
9. Koutkia P, Wachtel TJ. Pulmonary embolism presenting as syncope: case report and review of the literature. Heart Lung 1999;28:342-7.
10. Wolfe TR, Allen TL. Syncope as an emergency department presentation of pulmonary embolism. J Emerg Med 1998;16:27-31.
11. Torbicki A, Perrier A, Konstantinides S, Agnelli G, Galiè N, Pruszczyk P, et al. Guidelines on the diagnosis and management of acute pulmonary embolism: the Task Force for the Diagnosis and Management of Acute Pulmonary Embolism of the European Society of Cardiology (ESC). Eur Heart J 2008;29:2276-315.
12. Rudski LG, Lai WW, Afilalo J, Hua L, Handschumacher MD, Chandrasekaran K, et al. Guidelines for the echocardiographic assessment of the right heart in adults: a report from the American Society of Echocardiography endorsed by the European Association of Echocardiography, a registered branch of the European Society of Cardiology, and the Canadian Society of Echocardiography. J Am Soc Echocardiogr 2010;23:685-713.
13. Konstantinides S, Geibel A, Olschewski M, Kasper W, Hruska N, Jäckle S, et al. Importance of cardiac troponins I and T in risk stratification of patients with acute pulmonary embolism. Circulation 2002;106:1263-8.
14. Naeije R, Huez S. Right ventricular function in pulmonary hypertension: physiological concepts. Eur Heart J 2007;9(Suppl H):H5-9.
15. Coghlan JG, Davar J. How should we assess right ventricular function in 2008? Eur Heart J 2007;9(Suppl H):H22-8.
16. Barst RJ, McGoon M, Torbicki A, Sitbon O, Krowka MJ, Olschewski H, et al. Diagnosis and differential assessment of pulmonary arterial hypertension. J Am Coll Cardiol 2004;43:40S-47S.
17. Dilley A, Austin H, Hooper WC, Lally C, Ribeiro MJ, Wenger NK, et al. Relation of three genetic traits to venous thrombosis in an African-American population. Am J Epidemiol 1998;147:30-5.
18. Fuentes RM, Perola M, Nissinen A, Tuomilehto J. ACE gene and physical activity, blood pressure, and hypertension: a population study in Finland. J Appl Physiol 2002;92:2508-12.
19. Kreutz R, Hübner N, Ganten D, Lindpaintner K. Genetic linkage of the ACE gene to plasma angiotensin-converting enzyme activity but not to blood pressure. A quantitative trait locus confers identical complex phenotypes in human and rat hypertension. Circulation 1995;92:2381-4.
20. Abraham WT, Raynolds MV, Gottschall B, Badesch DB, Wynne KM, Groves BM, et al. Importance of angiotensin-converting enzyme in pulmonary hypertension. Cardiology 1995;86 Suppl 1:9-15.
21. Kanazawa H, Okamoto T, Hirata K, Yoshikawa J. Deletion polymorphisms in the angiotensin converting enzyme gene are associated with pulmonary hypertension evoked by exercise challenge in patients with chronic obstructive pulmonary disease. Am J Respir Crit Care Med 2000;162:1235-8.
22. Tanabe N, Amano S, Tatsumi K, Kominami S, Igarashi N, Shimura R, et al. Angiotensin-converting enzyme gene polymorphisms and prognosis in chronic thromboembolic pulmonary hypertension. Circ J 2006;70:1174-9.