行政院國家科學委員會補助專題研究計畫成果報告

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※　血管張力素元與張力素轉化 °基因多態型　※
※　　與冠狀動脈整型術後再狹窄的關連　　※
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計畫類別： o個別型計畫　　á整合型計畫

計畫編號： NSC 89－2314－B－002－423

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計畫主持人： 江福田教授

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一、中文摘要

腎素─張力素系統(RAS)一直在心血管疾病的致病機轉上扮演重要的角色。因此該系統基因之多態型可能與冠狀動脈氣球整型術後之再狹窄有關連。但最近之研究，結論不一致，故吾等擬全面探討該系統基因之多態型與再狹窄之關係。自 1996 至 1999 年，吾等收集 285 例冠狀動脈心臟病人，他們皆接受過成功的氣球整型術與一次追蹤的冠狀動脈攝影(平均追蹤時間為 42.4 個月)。再狹窄之定義為在前一次氣球擴張術後管腔口徑堵塞在 50%以上。吾等以 PCR 定序與 PCR-RFLP 之技術做基因型之鑑定，共包括血管張力素元基因-6、-20、-158、-217、+174 與+235 之變異，張力素轉化 ◦ D/I 型與張力素 II 第一型受器(Angiotensin II type I receptor) +1166 之多態型。結果吾等發現，張力素轉化 ◦ D 型對偶基因在單變項或多變項分析皆與再狹窄有密切關連(D/D 型有 72.5%、D/I 型 54.4%、I/I 型 47.5%再狹窄，P 值趨向 <0.01)。此一效果受到張力素元 M174 型對偶基因之干擾(干擾檢定 P 值 = 0.03)。結論：本研究之結果顯示張力素 D 型對偶基因在冠動脈氣球整型術後再狹窄扮演重要角色。但張力素元基因 M174 型可調節之，此現象似乎可以說明從前研究結論之分歧。

關鍵詞：腎素─張力素系統、多態型、整型術、再狹窄

Abstract

The renin-angiotensin system (RAS) has been implicated in the pathogenesis of cardiovascular diseases. This suggests that DNA polymorphisms of component genes in RAS may modulate the disease process including post-percutaneous transluminal coronary angioplasty (PTCA) restenosis. Due to conflicting results in previous studies, we think systemic analysis of predisposing alleles in RAS will improve our understanding of the mechanisms.

We investigated the influence of eight gene polymorphisms involved at different steps of enzymatic cascade in RAS on post-PTCA restenosis simultaneously. This prospective study included 285 Chinese patients who underwent a successful angioplasty procedure and follow-up angiography (mean 42.4 ± 14.5 months) if angina recurred with positive non-invasive stress tests. Restenosis is defined as a more than 50% diameter stenosis at the previously treated vessel site. Among these genotypes, only ACE D allele was significantly associated with post-PTCA restenosis in univariate (D/D 72.5%, D/I 54.4%, I/I 47.5%, p value test for trend < 0.01) and multivariate (p value test for trend < 0.01) analysis. One novel finding in our study is that the effect of ACE D allele on restenosis is reversed by AGT M174 allele (p value to test for the interaction term = 0.03).

Our results indicate ACE D allele increases the risk of post-PTCA restenosis

which is modified by AGT M174 allele in Chinese patients. The gene-gene interactions have increasing importance in the post-genome era and may explain why the conflicting results in previous studies.

Keywords： renin-angiotensin system, polymorphism, angioplasty, restenosis.

二、緣由與目的

Percutaneous transluminal coronary angioplasty (PTCA) is a standard treatment for coronary artery disease. However, post-PTCA restenosis is the major limitation. Even in the stenting era, restenosis is still a challenge to the interventionists. Vascular remodeling in post-PTCA restenosis is disclosed in clinical studies but the exact molecular and cellular mechanisms are still unknown. Renin-angiotensin system (RAS) has been extensively studied as an important mediator of cardiovascular disease since its role in hypertension conducted first by Hilbert et al. in 1991 (1). The major components of RAS included angiotensinogen (AGT), renin, angiotensin-converting enzyme (ACE), angiotensin II and angiotensin II receptors. Binding of the final product of this enzymatic cascade angiotensin II to its receptor resulted in vasoconstriction, and aldosterone and catecholamine release (2). In addition, angiotensin II has other biologic effects that influence vascular endothelium and smooth muscle cells, and cardiac fibroblasts (3-5). These may play a role in the pathophisiology of coronary artery disease, myocardial infarction, and restenosis after percutaneous coronary interventions. The prominent role of RAS in cardiovascular regulation suggests that DNA polymorphisms of component genes may modulate cardiovascular disease process (6-13). However, the conflicting results were attained in such studies so far (14,15). Therefore, we think simultaneous analysis of several predisposing alleles in RAS should provide more information about the pathophisiology of atherosclerosis and subsequent interventions and to adapt therapeutic management if possible (16,17).

The present study is designed to investigate the relation between DNA polymorphisms of RAS including AGT T174M and M235T gene polymorphisms, AGT5' upstream core promoter region G-6A, A-20C, G-152A, G-217A gene polymorphisms, ACE gene insertion/deletion (I/D) polymorphism and angiotensin II type 1 receptor (AT1R) A1166C gene polymorphism, and risk of restenosis after balloon coronary angioplasty (PTCA) and to evaluate the potential interactions with these polymorphisms in Chinese.

三、結果與討論

We investigated the influence of eight gene polymorphisms involved at different steps of enzymatic cascade in RAS on post-PTCA restenosis simultaneously. This prospective study included 285 Chinese patients who underwent a successful angioplasty procedure and follow-up angiography (mean 42.4 ± 14.5 months) if angina recurred with positive non-invasive stress tests. Among these genotypes, only ACE D allele was significantly associated with post-PTCA restenosis in univariate (D/D 72.5%, D/I 54.4%, I/I 47.5%, p value test for trend < 0.01) and multivariate (p value test for trend < 0.01) analysis. One novel finding in our study is that the effect of ACE D allele on restenosis is reversed by AGT M174 allele (p value to test for the interaction term = 0.03).

Our results indicate ACE D allele increases the risk of post-PTCA restenosis which is modified by AGT M174 allele in Chinese patients. The gene-gene interactions have increasing importance in the post-genome era and may explain why the conflicting results in previous studies.

Post-PTCA restenosis is the consequence of repair and growth after vessel

injury by balloon catheter. The process consists of neointimal hyperplasia, smooth muscle cell proliferation and excessive connective tissue accumulation due to responses to various cytokines. There is evidence suggests that RAS and polymorphisms of component genes are involved in the process (23,24). Because the mechanisms of restenosis process are polygenic and multifactorial, it is important to analyze the predisposing alleles in global views including gene-gene interactions. However, rare studies were approached with this strategy (14,25,26). Therefore, eight gene polymorphisms involved at different steps of enzymatic cascade in RAS were analyzed in this study. To our knowledge, this is the most systematic approach to post-PTCA restenosis with relation to gene polymorphisms of RAS.

The genotype frequencies of AGT-T174M, AGT5'-A-20C and ACE I/D gene polymorphisms are similar to other populations (6,10,11,27,28). The frequencies of AGT-T235 and AGT5'-A(-6) homozygotes are higher than that reported for Caucasian populations (6,7,28) but similar to that reported for Japanese or other Chinese populations (27,29,30). The frequency of AT1R-C1166 allele is higher in Caucasian populations and similar to Japanese populations (31, 32). AGT5'-G-152A and G-217A are noted distinctly in our populations and their functional studies are ongoing. In this study, we disclosed ACE D allele increased the risk of restenosis after PTCA in Chinese patients. Based on previous study by Rigat et al (33), ACE D allele displayed a positive association with the corresponding plasma concentration of ACE. This may resulted in increased Angiotensin II generation, which stimulates the release of several cytokines such as platelet-derived growth factor and fibroblast-derived growth factor that are potent stimulants for smooth muscle cell proliferation and growth. The notion was also supported by the findings that ACE inhibitors were effective in retarding, preventing, or reversing restenosis (34). Another interesting finding in this study is the effect of ACE D allele on restenosis is modified by AGT-M174 allele. Approximately 19% (25/285) of individuals possess AGT-M174 allele in our population. Their trend for restenosis with increased ACE D allele is reversed by AGT-M174 allele. Although no significant phenotype with AGT-M174 allele was noted before, it may acts as a regulator through the unknown mechanism. The gene-gene interactions were ever described previously between ACE D and AT1R-C1166 homozygotes (13,35). They found an increased risk for CAD among individuals with D/D + C/C genotypes. However, a number of statistical tests used in this study should be considered, and further confirmation of this relationship and its mechanism are definitely required.In the post-genome era, the gene-gene interactions become a potential problem because more and more genes are identified and development of microarray results in global views of gene expressions. Understanding these interactions can help us to clarify the genotype-phenotype associations, and may also explain why the conflicting results were obtained in the previous studies.

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