過氧化體增生活化受體γ、基質?屬蛋白溶解酵素-9 基因多形性與缺血性中風之相關研究

Association of the peroxisome proliferator-activated receptor gamma and matrix metalloproteinases-9 gene polymorphisms with ischemic stroke

中文摘要

腦血管疾病是臺灣居民的第二位死因，僅次於癌症。其中，缺血性中風(ischemic stroke)是最主要的類型，約佔 70%。缺血性中風的發生機制是血管內的小血栓經由頸動脈流入腦血管中，阻塞住某一部份腦細胞的血流，而造成腦組織的傷害。而缺血性中風的發生與動脈粥狀硬化有極大的關聯，動脈粥狀硬化不僅可能造成血管內徑的狹窄，影響血流，其血管內膜的粥狀硬化斑塊亦可能脫落形成血栓而阻塞腦部血流。過氧化體增生活化受體γ(peroxisome proliferator-activated receptorγ, PPARγ)在動脈粥狀硬化形成過程中扮演重要的角色，包括血管的重建、脂肪斑的形成、發炎反應以及血栓的形成等等的過程。另外，由於基質金屬蛋白溶解酵素-9(matrix metalloproteinases-9, MMP-9)能夠溶解細胞外基質成分的特性，因此在動脈粥狀硬化與斑塊形成過程中，MMP-9 亦是一關鍵的因素。本研究目的為探討 PPARγ、MMP-9 基因多形性以及傳統危險因子對缺血性中風的獨立與交互作用。

研究對象包括 537 位缺血性中風患者與 537 位經年齡、性別配對的對照族群。資料收集包括利用結構式問卷經標準化流程收集基本人口學、相關危險因子與疾病史等資料，以及利用禁食八小時靜脈血測量相關血液生化值資料。基因型的判定是使用聚合酶連鎖反應(polymerase chain reaction, PCR)與限制片段長度多形性方法(restriction fragment length polymorphism, RFLP)。使用邏輯斯迴歸模式分析 PPARγ、MMP-9 基因多形性與缺血性中風的關係。

結果顯示抽菸、飲酒、高血壓、糖尿病、高血脂症、家族中風史、腹部肥胖等危險因子會增加罹患缺血性中風的風險。基因型部分的分析發現，PPARγ C-681G、C-2821T 基因多形性與缺血性中風的發生有相關性。相較於 CC 基因型，PPARγ C-681G 攜帶 G 對偶基因者有 1.44 倍的風險罹患缺血性中風；PPARγ C-2821T 攜帶 C 對偶基因者相較於 TT 基因型者有 1.35 倍的風險罹患缺血性中風。而在單套型分析部份，帶有 GCC 單套型者有 1.32 倍的風險罹患缺血性中風(單套型次序：C-681G、C-2821T、Pro12Ala)。此外，高血壓、糖尿病與 PPARγ C-681G、C-2821T 對罹患缺血性中風的危險性存在累乘協同作用的關係，且在調整年齡、性別與教育程度前後均達統計上顯著差異。並且在 C 反應蛋白大於

0.2mg/dL 的情況下，PPAR γ C-2821T 與 MMP-9 R279Q 對缺血性中風的發生存在基因與基因的交互作用。

總結以上，本篇研究發現 PPAR γ 基因多形性與缺血性中風的發生有相關性，而其風險與糖尿病、高血壓存在累乘協同效應。此外在發炎反應劇烈的條件下，PPAR γ 與 MMP-9 有顯著的基因-基因交互作用。顯見 PPAR γ 對缺血性中風發生與否的重要性。

英文摘要

Stroke is the second leading cause of death in Taiwan. Moreover, the major subtype of stroke is ischemic stroke (about 70%). Atherosclerosis is a well-known risk factor of ischemic stroke. The mechanisms of atherosclerosis progression were proved to include inflammation, cell adhesion, thrombosis, functional disturbance of lipid metabolism, and platelet function. Recent studies showed that peroxisome proliferator-activated receptor γ (PPARγ) was expressed in the endothelium, VSMCs, macrophages, T lymphocytes, and human atherosclerotic lesions. Furthermore, PPARγ was reported to involve in almost every stage of atherosclerosis development and progression. The other important risk factor is matrix metalloproteinases-9 (MMP-9). MMP-9 has been found in the migration of vascular smooth muscle cells and the degradation of the extracellular matrix of atheromatous plaque. As the fibrous cap is getting thin, the plaque becomes increasingly unstable and prone to rupture. This progress is possibe to stimulate plaque enlargement and result in acute ischemic stroke. In order to examine this hypothesis, we investigated the relationship between gene polymorphisms of PPARγ, MMP-9 and the risk of developing ischemic stroke.

537 ischemic stroke patients and 537 controls were recruited in this study. A total of 537 acute ischemic stroke patients aged between 30 to 95 years old from the department of neurology of Chi-Mei, Lotung Poh-Ai, Wan-Fang and TMU hospitals in Taiwan were recruited as cases. 537 Stroke-free subjects from the regular health examination in Shin-Kong WHS Memories and Wan-Fang hospitals in Taipei in 2004 were controls. Cases and controls were frequency matched by age and sex. We genotyped four SNPs of PPARγ (C-681G, C-2821T, Pro12Ala, and C161T) and three SNPs of MMP-9 (C-1562T, R279Q, and P574R) using polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) method. All subjects were interviewed by trained research assistants to collect questionnaire data including conventional vascular risk factors and disease history. Fasting glucose, lipid profile, CRP, and uric acid were from routine biochemistry tests. Logistic

regression model was used to estimate the odds ratio (OR) and 95% confidence interval (CI).

Study subjects with the G allele of C-681G or C allele of C-2821T significantly increased the risk of ischemic stroke (OR=1.44, 95% CI : 1.13-1.84 and OR=1.35, 95% CI : 1.06-1.71, respectively). A significantly increased risk of ischemic stroke was also found in subjects with the GCC haplotype (C-681G, C-2821T, Pro12Ala ; OR=1.32, 95% CI : 1.09-1.59), even after the Bonferroni correction. Moreover, we found the significant joint effect between PPARγ C-681G, C-2821T and hypertension, diabetes mellitus on the risk of ischemic stroke. Our results also showed that there was interaction between the PPARγ and MMP-9 genes in predicting the risk of ischemic stroke as the C reaction protein was higher than 0.2 mg/dL.

In conclusion, our results provide the evidence of association between PPARγ gene polymorphisms and ischemic stroke in a Taiwanese population. Both C-681G G allele and C-2821T C allele of PPARγ gene significantly increased the risk of ischemic stroke. We also found a strong synergistic effect between the PPARγ gene and hypertension, diabetes mellitus on the risk of ischemic stroke. Furthermore, our results indicated that there was interaction between the PPARγ and MMP-9 genes in predicting the risk of ischemic stroke.