出國報告（出國類別：開會＋其他）

第3屆日本心臟學會基礎心臟血管學研究會議
(3rd Council Forum on Basic CardioVascular Research)

服務機關：臺中榮民總醫院心臟血管中心
姓名職稱：梁凱偉／一般心臟醫學科主任
派赴國家/地區：日本東京
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一、 摘要

Heme oxygenase-1 gene promoter $(GT)_n$ repeats polymorphism in cardiovascular diseases

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Background: Heme oxygenase (HO)-1 is a rate-limiting enzyme for degrading heme into carbon monoxide and promotes vasodilatation and anti-oxidation. Shorter $(GT)_n$ repeats of the HO-1 gene (HOMX1) promoter are associated with higher transcriptional activity in vitro. The longer $(GT)_n$ repeats of HOMX1 promoter have been correlated with incidence of coronary artery disease (CAD). We have done researches for investigating the $(GT)_n$ repeats of the HOMX1 promoter in different cardiovascular diseases, such as CAD with different Jeopardy severity scores, microvascular angina (MVA), and non-ischemic dilated cardiomyopathy (DCM).

Methods: Leukocyte genomic DNA was extracted and polymerase chain reaction was done for amplifying the HOMX1 promoter $(GT)_n$ repeating segment for determining the repeat number. The study cohort was from the patients who received cardiac catheterization and agreed donating blood samples for research use.

Results: The severe CAD group (Jeopardy Score ≥ 8) had a significantly lower frequency of S allele (3.7% vs. 8.9%, $p = 0.042$) than the moderate CAD group (Jeopardy Score < 8). In a multivariate binary logistic analysis, carrier of shorter GT repeats was a significant negative predictor (odds ratio 0.393, $p = 0.024$) for a higher Jeopardy Score-graded CAD. In another study, MVA subjects had higher ratio of carriers of L allele (83%) vs. healthy controls (66%) ($p<0.001$). In multivariate analysis, carrier of L allele (odds ratio 2.772, $p<0.001$) was a significant predictor for the diagnosis of MVA. In a study for DCM subjects, moderately-reduced left ventricular ejection fraction (EF) group (EF 36-40%) had higher ratio

of carriers of S allele (86%) than the severely reduced EF group (EF≤35%) (68%) (p=0.012). In a multivariate analysis, carrier of S allele (odds ratio 0.407, p=0.047) was a significant negative predictor for the diagnosis of severely reduced LVEF (≤ 35%) in non-ischemic DCM. **Conclusions:** Shorter $(GT)_n$ repeat in HMOX1 promoter was associated with a lower Jeopardy severity score in patients with significant CAD. Subjects with MVA had longer HMOX1 promoter $(GT)_n$ repeats than the healthy controls. Non-ischemic DCM with severely reduced LVEF (≤ 35%) had longer HMOX1 promoter $(GT)_n$ repeats than those with moderately reduced LVEF (36-40%).

Keywords: coronary artery disease (CAD); non-ischemic dilated cardiomyopathy (DCM); heme oxygenase-1 (HO-1); jeopardy severity score; microvascular angina (MVA); promoter polymorphism.

二、 目的

心臟血管中心一般心臟醫學科主任：梁凱偉奉准於 108 年 9 月 6 日公假參加於日本東京舉行的日本心臟學會 (The Japanese Circulation Society) 第 3 屆基礎心臟血管學研究會議 (3rd Council Forum on Basic CardioVascular Research)，擔任邀請演講者(invited speaker)於 Symposium 6: Genomics in Cardiology 發表口頭演講 (invited speaker)。

三、 過程

Presidential address---日本慶應大學心臟內科教授 Dr. Fukuda, 介紹 iPSC 誘導性多能幹細胞（induced pluripotent stem cell），又稱人工誘導多能幹細胞，常簡稱爲 iPS 細胞（iPSC），是一種由哺乳動物成體細胞 (ex. Skin fibroblast)經轉入轉錄因子等手段脫分化形成的多能幹細胞，在心衰竭 或 心肌梗塞後 心肌再生的相關研究。最早由日本學者山中伸彌的研究團隊於 2006 年發現。山中伸彌團隊在發表 iPS 誘導

技術時使用實驗材料爲小鼠細胞。2007 年，研究人員又證明 iPS 誘導技術可以應用於人體細胞[2]。最初由山中伸彌團隊發現的誘導方法是透過慢病毒載體將 *Oct4*、*Sox2*、*c-Myc*、*Klf4* 四種轉錄因子基因轉入成體細胞將其轉化爲類似於胚胎幹細胞的多能幹細胞。其後，研究人員又先後發現了更優化的誘導方法，如使用質體載體轉染、腺病毒感染、脂質體導入等非基因組整合的方法進行誘導、透過細胞融合誘導、使用小分子藥物進行誘導、轉入 miRNA（微干擾 RNA）進行誘導等。iPS 細胞與胚胎幹細胞擁有相似的再生能力，理論上可以分化爲成體的所有器官、組織。而相比胚胎幹細胞，iPS 細胞面臨的倫理道德爭議較小，且應用該技術可以產生基因型與移植受體完全相同的幹細胞，規避了排異反應的風險，因而 iPS 細胞在一定程度上衝擊了胚胎幹細胞在再生醫學中的地位，被認爲在再生醫學及組織工程方面擁有較爲廣闊的應用前景，有望爲治癒糖尿病、關節炎等疾病提供新的思路。同時，iPS 細胞在新藥開發、疾病模型構建領域也有望得到應用。但 iPS 誘導技術同樣面臨著誘導效率低、用於治療可能存在長期風險等挑戰。iPS 技術的發明人山中伸彌於 2012 年與核移植及複製技術研究的先驅者約翰·格登爵士一同獲諾貝爾生理醫學獎。

Keynote lecture, Dr. Marlene Rabinovitch, Professor of Pediatric Cardiology, Stanford University Bone morphogenetic protein (BMP) receptor type 2 (BMPR2) mutation 與原發性肺高壓的發生機制 *BMPR2* was identified as the first PAH-predisposing gene. This gene is located on the long arm of chromosome 2 (2q31-32) and encodes a type II receptor (BMPRII) belonging to the TGF-β receptor superfamily. The BMPRII receptor is involved in the regulation of growth, differentiation, and apoptosis of pulmonary artery endothelial and smooth muscle cells. Loss of BMPR2 causes proliferation of PA SMCs in response to TGF-β1 and BMP2, in contrast to the inhibition of SMC proliferation and increased susceptibility to apoptosis normally observed with these morphogens. Normal BMPR2 signaling negatively regulates PDGF and likely other growth-promoting factors implicated in the pathobiology of PAH, such as EGF. Genetic studies have demonstrated that 70% or more of patients with hereditary PAH, and 10%－20% of patients with sporadic IPAH, are heterozygous for a mutation BMPR2. BMPR2 is a member of the TGF-β superfamily of growth factor receptors. Mutations can affect different functions of BMPR2, namely the

ligand-binding domain, the signaling mechanism, and the interaction of the receptor with the cytoskeleton. BMPR2 is expressed ubiquitously and, in association with a coreceptor (usually BMPR1A), can signal through many different pathways, including pSmad1/5,p-p38, pERK, JNK, and Akt/PI3K.

Dr. Rabinovitch 's group has shown that BMPs, via BMPR2, activate the canonical Wnt signaling pathway to induce PA EC survival and proliferation and activate the noncanonical pathway to induce migration, critical features in angiogenesis and regeneration of damaged blood vessels. Further work by Rabinovitch' s group showed that a complex between β-catenin and PPARγ is essential in directing these functions and that a key downstream gene transcribed by this complex is apelin. Apelin has autocrine effects in promoting PA EC survival and migration and paracrine effects in suppressing aberrant PA SMC growth. Mice with PPARγ deleted in ECs develop spontaneous PAH that is reversed by treatment with apelin.

四、 心得

三天會期內有 3 場 Keynote lecture 皆為大師級演講，及一場 presidential address(Professor Keiichi Fukuda---Keio University)內容涵蓋血管分子生物學，regenerative medicine (stem cell),肺高壓基礎研究。此外還有 6 場基礎或臨床的 symposium，皆為 updated basic research。與會研究報告，以日本在基礎心臟再生醫學，血管分子生物醫學及動脈硬化相關機制研究，領先我們許多，尤其是大量使用 knock-out mice, tissue-specific knock-out mice, or organ-specific knock-out mice, or transgenic mice 來驗證其臨床或動物模式或細胞模式研究結果，值得我們學習趕上。

五、 建議

無。

六、 附錄

請詳見附件。