UNIVERSITI TEKNOLOGI MARA

THE PREVALENCE AND FUNCTIONAL IMPACT OF RARE GENOMIC COPY NUMBER VARIATION IN HYPERTENSION-RELATED LEFT VENTRICULAR HYPERTROPHY

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AUTHOR'S DECLARATION

I declare that the work in this thesis was carried out in accordance with the regulations of Universiti Teknologi MARA. It is original and is the result of my own work, unless otherwise indicated or acknowledged as referenced work. This thesis has not been submitted to any other academic institution or non-academic institution for any degree or qualification.

I, hereby, acknowledge that I have been supplied with the Academic Rules and Regulations for Post Graduates, Universiti Teknologi MARA, regulating the conduct of my study and research.

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ABSTRACT

Left ventricular hypertrophy (LVH) is an independent risk factor for cardiovascular morbidity and mortality, and a powerful predictor of adverse cardiovascular outcomes in hypertensive patients. The current community based study shows that of the 992 male hypertensive participants, 26.6% had LVH at echocardiography. A majority of the hypertensive LVH (HT LVH+) participants were from rural areas suggesting lower socio-economy status, lower awareness, treatment and control rate of hypertension have a role to play. Copy number variation (CNV) may contribute to the development of HT LVH+. Three hundred hypertensive participants among whom 100 had LVH, underwent genome-wide genotyping of copy number variation using microarray. CNV analysis was performed using three independent algorithms: CNV partition v2.3.4, PennCNV and iPattern. CNVs that passed the QCs were then subjected to Gene Ontology and Pathway analysis using DAVID (Database for Annotation, Visualization and Integrated Discovery, version 6.7), Ingenuity and GeneGO Metacore. A total of 208 rare CNVs were identified in HT LVH+. Analyses revealed the involvement of candidate genes such as *IQGAP2, CDH15, F2R*, and *VAV3* that are known to be functionally involved in cardiac development and phenotypes. Network enrichment analyses suggested that the gene-set was, directly or indirectly, involved in the transcription factors regulating the "foetal cardiac gene programme" which triggered the hypertrophic cascade ie. Sp1, p53 and CREB1, and androgen receptor signalling cascades. This suggests that regulation of mechanisms related to foetal cardiac gene could be a key point for the treatment of LVH. To improve the understanding on the mechanism of foetal cardiac gene program in the pathogenesis of HT LVH+, a functional validation was carried out by targeting the Sp1 signalling pathway using *in vitro* model. Mithramycin, a Sp1 inhibitor was used to imitate regulation of foetal cardiac gene. Losartan (Angiotensin II receptor blocker) was used to see whether regression of LVH was directly related to BP control or indirectly by triggering foetal cardiac gene program. Cultured human cardiomyocytes (HCM) were exposed to Angiotensin II (Ang II) at a final concentration of 1 μM for 24 hours to induce cell hypertrophy. Mithramycin 150 nM and losartan at a final concentration 10000 nM was then added to the hypertrophied-HCM cultures for 24 hours to reverse the hypertrophy. Global gene expression of the cells was then characterized using Illumina platform (HumanHT-12 v4). Both losartan and mithramycin treatments showed significant regression of the hypertrophied-HCM. When differential expressed genes were compared between these groups, 207 genes ($p<0.001$) shared similar patterns. Enrichment analysis revealed the involvement of these genes in the cardiac development via Sp1 signalling. Thus the development of HT LVH+ was through activation of transcription factor which was highly expressed during cardiac development. Regression of cardiac hypertrophy by losartan treatment not only works directly via blood pressure reduction and but also at least in part, attenuating foetal gene program activation. In conclusion, more than a quarter of hypertensive male Malaysians had LVH and genetic factor (rare CNV) plays a contributing factor to the development of HT LVH+ through alteration of the functional regulation of foetal cardiac gene programme. This study complements the understanding of underlying molecular mechanism of HT LVH+, providing new insights on the fundamental mechanism of action of losartan in regression of HT LVH+.

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