UNIVERSITI TEKNOLOGI MARA

INVESTIGATION ON THE IMPACTS OF BISPHENOL A EXPOSURE ON THE *IN VITRO* MODEL OF HEART AND *IN VIVO* MODEL OF PREGNANT RATS AND FOETUSES

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PhD

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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 results 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 Graduate, Universiti Teknologi MARA, regulating the conduct of my study and research.

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ABSTRACT

In utero exposure to environmental contaminants such as bisphenol A (BPA) has been reported to alter the profile of microRNA (miRNA) and subsequently increase the risk of cardiovascular disease (CVD) development. Therefore, this study aimed to investigate the impact of BPA exposure on cardiomyocytes using primary neonatal rat cardiomyocytes cultured (CMs) and in utero BPA exposure on heart of mothers and foetuses using pregnant rats. In CMs, cells were exposed to 0.1 to 100 μM of BPA and beating frequencies were significantly reduced in BPA- exposed cells (48%-64%, p=0.000), reduction of cell viability (54%, p=0.047) altered CMs morphology, in comparison to non-exposed CMs. For the pregnant rats, animals were divided into tween-80 (vehicle control), 0.05 mg/ml and 0.2 mg/ml BPA via drinking water for 19 days; from pregnancy day 2 till 21. In BPA-exposed animals, non-significant weight gained in mother and foetuses' weight, number and size of foetuses were observed in comparison to control ($p < 0.05$). BPA-exposed pregnant rats showed significant increase in blood pressure (BP) ($p < 0.05$) and altered in cardiac miR-499-5p expression in comparison to control group ($p < 0.01$). Foetal heart of BPA-exposed mother showed significant reduction in glycogen content and significant upregulation of miR-17-5p, -208-3p, and -210-3p ($p < 0.05$) expression. H&E staining of BPA-exposed foetal hearts showed signs of fibrosis while BPA-exposed mothers showed muscle remnant. Number of cells and sizes of heart were analysed and found not significant between the groups of exposure. The presence of fibrosis was further confirmed by Masson trichrome staining in BPA-exposed foetal heart. Remarkably, reduced expression of cardiac troponin I (cTnI) was also observed in foetus of BPA-exposed mother in comparison with foetus of control mother. In contrast, alpha fetoprotein (AFP) expression was well distributed in cytoplasm of control foetal heart while less expression of AFP was observed in heart of foetus from BPA-exposed mother. Hypoxia induced factor-1 alpha (HIF-1α) was elevated in expression in BPA-exposed foetal heart compared to the control. For CMs, BPA affected both morphology and function of cells. In accordance, BPA exposure in pregnant rats led to increase of BP, altered cardiac miR-499-5p and formation of muscle remnant in heart tissues with diffused cellularity and less defined striation. For foetus, BPA exposure altered heart tissue morphology, miRNA profile and muscle protein expression. In conclusion, the findings in here suggest the risk of in utero BPA exposure on both foetus and mother, which may increase the risk of CVD in later life by altering the expression of cardiac miRNA profile and protein crucial for heart development and function.

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