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Viability and Glycosaminoglycan Content in Chondrocytes Transfected with SRY (Sex-Determining Region Y)-box 9 and Telomerase Reverse Transcriptase Genes

Noorhidayah Md Nazir[1], Ahmad Hafiz Zulkifly[2], Kamarul Ariffin Khalid[2], Ismail Zainol[3], Zaitunnatakhin Zamli[1], Munirah Sha'ban[1]

1 Department of Biomedical Science, Kulliyyah of Allied Health Sciences, International Islamic University Malaysia (IIUM), Jalan Sultan Ahmad Shah, Bandar Indera Mahkota, 25200 Kuantan, Pahang Darul Makmur, Malaysia.
2 Department of Orthopaedics, Traumatology and Rehabilitation, Kulliyyah of Medicine, International Islamic University Malaysia (IIUM), Jalan Hospital Campus, 25100 Kuantan, Pahang Darul Makmur, Malaysia.
3 Department of Chemistry, Faculty of Science and Mathematics, Universiti Pendidikan Sultan Idris (UPSI), 35900 Tanjong Malim, Perak Darul Ridzuan, Malaysia.

Email: noorhidayahmdnazir@gmail.com

Glycosaminoglycan (GAG) is a major component of cartilage matrix. This essential polysaccharide has the capacity to attract water molecules. Such capacity is important in cartilage primarily because it absorbs shock and provides lubrication in joint. Cartilage degeneration is often a normal ageing change. Along the process, the cartilage cell; chondrocyte may become less viable and thus, downregulate GAG production. This study evaluates cell viability and GAG content in chondrocytes using monolayer culture model. Chondrocyte usually loses its phenotype after several replications in culture. Hypothetically, replicative senescence contributes to ageing indirectly. Hence, this study compares SOX9 and/or TERT transfected and non-transfected chondrocytes to see whether or not the cells could remain viable and produce GAG after serial passages. While SOX9 is essential for chondrogenesis, TERT is responsible for cells longevity. Upon research approval (IIUM/IACUC/Approval/2015/[5]/[24]), rabbits' articular cartilages (n=2) were harvested. Isolated chondrocytes were seeded in 6 well-plate with an initial seeding of 5,000 cells/cm^2. At passage-1, the chondrocytes were transfected with SOX9 and/or TERT genes via lipofection. The four groups namely non-transfected (control), SOX9-, TERT- and SOX9/TERT-transfected chondrocytes were evaluated at passage-1, -2 and -3. It can be appreciated that

the post-transfected chondrocytes have cells viability and GAG content comparable to that of the non-transfected group. They exhibited similar downregulation pattern for viability and GAG content as postulated in theory of ageing. The viability ranged from the highest 93.43% to the lowest 78.49% throughout the culture. The total GAG contents are 25.23µg/ml, 23.87µg/ml, 21.59µg/ml and 23.86µg/ml for SOX9-, TERT-, SOX9/TERT-post-transfected chondrocytes and control, respectively. While cells viability gives an indication that transfection may have non-toxic effect on cells, this study raises some significant questions whether or not SOX9 and TERT could maintain or enhance chondrocyte properties at structural and ultrastructural levels. Detailed investigations into molecular and protein levels are also needed.

Keywords: Cartilage; Chondrocyte; Glycosaminoglycan; Gene Transfer; SOX9; TERT