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ORIGINAL ARTICLE

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Functional differences of Toll-like receptor 4 in osteogenesis, adipogenesis and chondrogenesis in human bone marrow-derived mesenchymal stem cells

Fatemeh Khodabandehloo[1] | Reza Aflatoonian[2] | Zahra Zandieh[3] | Farzad Rajaei[4] | Forugh-Azam Sayahpour[5] | Marjan Nassiri-Asl[4,6] | Mohamadreza Baghaban Eslaminejad[5]

[1]Department of Molecular Medicine, Qazvin University of Medical Sciences, Qazvin, Iran

[2]Department of Endocrinology and Female Infertility, Reproductive Biomedicine Research Center, Royan Institute for Reproductive Biomedicine, ACECR, Tehran, Iran

[3]Department of Anatomy, School of Medicine, Iran University of Medical Sciences, Tehran, Iran

[4]Cellular and Molecular Research Center, Research Institute for Prevention of Non-Communicable Disease, Qazvin University of Medical Sciences, Qazvin, Iran

[5]Department of Stem Cells and Developmental Biology, Cell Science Research Center, Royan Institute for Stem Cell Biology and Technology, ACECR, Tehran, Iran

[6]Department of Pharmacology and Neurobiology Research Center, Shahid Beheshti University of Medical Sciences, Tehran, Iran

Correspondence
Mohamadreza Baghaban Eslaminejad, Department of Stem Cells and Developmental Biology, Cell Science Research Center, Royan Institute for Stem Cell Biology and Technology, ACECR, Tehran, Iran.
Email: eslami@royaninstitute.org

Marjan Nassiri-Asl, Cellular and Molecular Research Center, Research Institute for Prevention of Non-Communicable Disease, Qazvin University of Medical Sciences, Qazvin, Iran.
Email: mnassiriasl@qums.ac.ir

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Abstract

Multipotent human bone marrow-derived mesenchymal stem cells (hMSCs) are promising candidates for bone and cartilage regeneration. Toll-like receptor 4 (TLR4) is expressed by hMSCs and is a receptor for both exogenous and endogenous danger signals. TLRs have been shown to possess functional differences based on the species (human or mouse) they are isolated from therefore, the effects of knockdown of TLR4 were evaluated in humans during the differentiation of MSCs into bone, fat and chondrocyte cells in vitro. We investigated the expression profile of TLR4 during the differentiation of hMSCs into three different lineages on days 7, 14 and 21 and assessed the differentiation potential of the cells in the presence of lipopolysaccharide (LPS, as an exogenous agonist) and fibronectin fragment III-1c (FnIII-1c, as an endogenous agonist). TLR4 expression increased following the induction of hMSC differentiation into all three lineages. Alkaline phosphatase activity revealed that FnIII-1c accelerated calcium deposition on day 7, whereas LPS increased calcium deposition on day 14. Chondrogenesis increased in the presence of LPS; however, FnIII-1c acted as a reducer in the late stage. TLR4 silencing led to decreased osteogenesis and increased adipogenesis. Furthermore, Wnt5a expression was inversely related to chondrogenesis during the late stage of differentiation. We suggest that understanding the functionality of TLR4 (in the presence of pathogen or stress signal) during the differentiation of hMSCs into three lineages would be useful for MSC-based treatments.