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Deciphering the role of substrate stiffness on stem cell transfection using lipid-based nanocarriers to deliver angiogenic genes

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Substrate stiffness is an important parameter regulating stem cell morphology and cytoskeletal organization. It may also play a critical role in the internalization of lipid-based nanocarriers for gene delivery. Based on this hypothesis, the present study investigates the role of substrate stiffness on non-viral transfection of human adipose-derived stem cells (hASCs) with the aim to maximize hASCs expression of vascular endothelial growth factor (VEGF) and enhance their wound healing potential. hASCs display more actin stress fibers when cultured on stiffer silicone substrates that mimicked the elasticity of hard tissues such as bone. This change in cytoskeletal composition facilitates a greater internalization of plasmid/lipofectamine nanocomplexes. Additionally, the investigation the main pathways involved in endocytosis is reported by studying the gene expression post-transfection. Caveolin-mediated genes are upregulated in the hASCs cultured on the stiffer silicone substrates, thus confirming the role of actin as a key mediator in the internalization of plasmid nanocomplexes. Transfected hASCs seeded on more rigid substrates exhibit higher levels of VEGF expression when compared to hASCs cultured on softer hydrogels. Overall, these findings suggest that a higher plasmid internalization and corresponding VEGF expression can be obtained when hASCs are cultured on hydrogels resembling the elasticity of hard tissues.

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