Mechanical Processing of Emulsified Lipoaspirate Results in a Dose-Dependent Upregulation of Stem Cell Markers and Populations

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INTRODUCTION: Mechanical processing of lipoaspirate (LA) is a commonly employed technique prior to reinjection for the purposes of lipofilling and skin rejuvenation. Our group has previously demonstrated that one form of mechanical processing, 'nanofat grafting,' results in a significant upregulation of multipotent mesenchymal stem cell (MSC) markers, adipose-derived stem (ADSCs) and endothelial progenitor cell populations (EPCs).[1] Recently, a pluripotent population termed multilineage stress-enduring (Muse) cells was described after subjecting lipoaspirate to various extreme stress conditions.[2] Based on these findings, we hypothesized that modulation of shear-stress alone would result in a correlative induction of markers associated with multipotency and/or pluripotency.

METHODS: Two microfluidic devices were created from acrylics and methacrylic ester using laser etching and 3D printing. Each multichannel construct consists of expansion and constriction regions with minimal widths of 500 µm (v4) or 250 µm (v2) where the narrower the channel, the greater shear force generated. Standard LA (n = 7) was set aside as a control or processed as nanofat.[3] Subsequently, two nanofat samples were processed via microfluidic devices regulated by a syringe pump (12.5 ml/min for 10 passes). Finally, each sample was subjected to collagenase digestion and the resulting stromal vascular fraction (SVF) pellets were subjected to automated cell count and multicolor flow cytometry panels.

RESULTS: On average, nanofat processing with or without microfluidic device yielded a four-fold decrease in nucleated cells when compared to control SVF. A dose-dependent pattern of stress-to-phenotype induction was observed for markers CD34 and CD13, as well as the subpopulations of MSCs, Muse cells, EPCs and ADSCs. The induction of MSCs ($p < 0.003$), Muse cells ($p < 0.002$), EPCs ($p < 0.04$) and ADSCs ($p < 0.05$) was much greater in all mechanically emulsified groups when compared to control, with v2 stress resulting in the largest populations.

CONCLUSION: Mechanical shear stress results in a dose-dependent induction of mesenchymal stem cell markers as well as multipotent/pluripotent populations. More detailed in vitro and in vivo studies are currently being explored to elucidate the mechanisms at play and examine the clinical significance of these findings. Additionally, we are working to determine the optimal stress needed to produce a potent progenitor mix for various clinical applications.

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