3rd International Conference on

WOUND CARE, TISSUE REPAIR & REGENERATIVE MEDICINE

September 11-12, 2017 | Dallas, USA

A COMPARISON BETWEEN TWO NEW METHODS TO ISOLATE ADIPOSE-DERIVED STEM CELL

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Introduction: In literature, while there is unanimous agreement on the harvesting procedure for adipose tissue, there is not a standardized protocol to isolate ASCs for clinical application. We experienced two different isolation procedure and we compared the results to prove which is the most effectiveness. The former used an enzymatic digestion procedure to maximize the number of adipose derived stem cells the latter is based only on mechanical isolation.

Material and Methods: Adipose tissue was obtained by conventional liposuction procedure performed on healthy people. With the first procedure, the fat was centrifuged at 1600 rpm for 3 minutes and then mixed with collagenase digestion solution Subsequently, the solution obtained was incubated for 30 minutes at 37°C in a shaker-incubator and, then, it was centrifuged at 200 relative centrifuge force (RCF) for 4 minutes. Finally, the SVF gained was washed two times and then centrifuged again at 200 RCF for 4 minutes. With the second procedure, the fat was subjected to a vibration cycle in a cell shaker at 600 vibration/ minute for 3 minutes and centrifuged immediately after at 1600 rpm for 3 minutes, without using enzymatic digestion. The procedure was performed under a laminar flow cabinet.

Results: Biological cell features were evaluated by cytometric flow analysis of specific surface antigens. In each method, the adipose-derived stem cells marker CD45-/CD34+/CD31- and the mesenchymal marker CD90+ were found positive. The main difference observed between the two procedures is the adipose-derived stem cells final number obtained. Starting from 100 ml of adipose tissue, we found a mean of 9.06 x 105 stem cells using enzymatic isolation, and 6.5 x 105 stem cells using only mechanical procedure.

Conclusion: We obtained a viable cellular pellet of adipose-derived stem cells with each procedure, but the enzymatic isolation allowed the collection of more cells than the mechanical one.