# EXOSOMES DERIVATION FROM PERIVASCULAR STEM CELLS

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**Introduction:** Exosomes have recently been highlighted as a new strategy for using in cell therapy because they are fundamental in the processes of cell growth, differentiation, invasion and angiogenesis by regulating cell-to-cell and cell-to-extracellular matrix interactions. Recently literature data shows that perivascular stem cells-derived exosomes secrete proteins include growth factors, cytokines and hormones or other soluble mediators which have therapeutic functions in a paracrine manner. In this regard, the purpose of this study was to isolate exosomes from adipose-derived perivascular stem cells.

**Methods:** Perivascular stem cells (PSCs) were initially cultured in exosome-production media containing a-MEM supplemented with exosome-free fetal bovine serum. When cells reached a confluence of 80%, extensive washes in PBS were performed to remove any possible residue of FBS. The cells were transferred in exosome-production medium and the culture split into two subcultures maintained for 48 hours. Exosomes were isolated by differential centrifugation at 300g for 10 minutes, 2,000g for 20 minutes, 10,000g for 30 minutes to eliminate cells and debris. Obtained supernatants were depleted of residual floating cells and cell debris by filtration with 0.22 urn, followed by two consecutive steps of ultracentrifugation at 100,000g for 90 minutes, including a washing step in PBS, to precipitate exosomes. The protein concentration was measured by Nanodrop. Morphological and ultrastructural characteristics of exosomes were studied using transmission electron microscopy (TEM). **Results:** Our results show that yield of proteins in exosomes after 48 hours cultivation of 1x10' PSCs was reached 1.5 mg/ml. Furthermore, TEM revealed that exosomes have a round-shaped form and their size ranges from 80 to 120 nm which characterize exosomes of adult stem cells.

**Conclusion:** Thus, our study demonstrate that exosomes for the first time were successfully isolated from PSCs. Derived exosomes from PSCs can be used as a novel immunomodulatory therapeutic agent for regenerative processes.