

# CHARACTERIZATION OF UMBILICAL CORD BLOOD CD34<sup>+</sup> CELLS AFTER SERUM-FREE *EX-VIVO* EXPANSION WITH ANGIOGENIC FACTORS

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**Introduction:** Endothelial Progenitor cells (EPCs) are isolated from the mononuclear cell (MNC) fraction of umbilical cord blood (UCB), bone marrow, or peripheral blood. This small fraction of cells has been proven to be an attractive cell source not only for cardiovascular application but also for various medical conditions. However, to obtain an adequate number of cells from limited blood volume is challenging in some clinical situations because of the low frequency of EPC. Mobilization of EPCs with cytokine from bone marrow is now the common method for obtaining large number of cells, but this is considered invasive and inconvenient for patients. In this report, we describe a novel serum-free *ex-vivo* expansion (ExV) culture system for UCB derived CD34<sup>+</sup> cells to obtain large cell numbers and to enhance their therapeutic potential.

**Methods:** CD34<sup>+</sup> cells were isolated from UCB using immunomagnetic beads. ExV culture with serum-free medium containing SCF, TPO, VEGF, IL-6 and Flt-3 ligand was applied to UCB-CD34<sup>+</sup> cells ( $n = 6$ ) for 7 days. Post-ExV cell numbers were counted and compared with pre-ExV number. Gene expressions were analyzed by real-time PCR. Freshly isolated UCB-CD34<sup>+</sup> cells ( $n = 4$ ) were also used as control.

**Results:** The ExV culture resulted in significant increase in the cell number by 20-30 folds after 7 days. Data of real-time PCR showed up and down-regulation of several angiogenic and anti-inflammatory factors post ExV.

**Conclusion:** The findings of this study showed that our novel, serum-free ExV system could provide a significant increase of cell number with enhancement of their therapeutic potential. Utilizing ExV culture to obtain larger number of cells may eliminate or decrease the dose of cytokine for mobilization of cells. Moreover, modifications of angiogenic and anti-inflammatory genes may provide new insights into the therapeutic potential of using ExV expanded CD34<sup>+</sup> cells not only in cardiovascular regeneration but also various applications of EPC induced organogenesis.