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Ex vivo expansion of Primate CD34+ Cells isolated from Bone Marrow and Human Bone Marrow Mononuclear Cells using a Novel Scaffold

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Background:
Bone marrow derived CD34+ cells have been in clinical application in patients with haematological malignancies. One of the major problems with this treatment is the non-availability of matched donors or the necessity of multiple transfusions depending upon the pathology. Recently evidences have been accumulating to prove the safety and efficacy of autologous CD34+ cells in diseases such as myocardial dysfunction, peripheral vascular diseases and neurological certain conditions. However there are only a few reports in the literature on ex vivo expansion of the bone marrow derived CD34+ cells. We have in two different studies proven that isolated CD34+ cells from baboon bone marrow and non-isolated BMMNCs from human bone marrow could be expanded with increase in percentage of CD34+ cells using a novel scaffold.

Methods:
Study I: Bone marrow was derived from healthy baboons posterior iliac crest and mononuclear cells were isolated using density gradient method. Then the BMMNCs were subjected to magnetic bead (Miltenyi Biotech) separation of CD34+ cells, which were expanded ex vivo for one week.

Study 2: A portion of the bone marrow drawn for diagnosis or bone marrow aspirated for clinical application from human patients was used after informed consent. The BMMNCs were subjected to ex vivo expansion without any further separation. Initial and post-expansion CD34+ percentage and quantity were evaluated at different intervals starting from 7 to 21 days. Both the studies used the same Thermo-Gelation Reversible Polymer Scaffold impregnated with same culture cocktails prepared in NCRM.

Results:
Study I revealed increase in the quantity of CD34+ cells which formed colonies in the culture from the 5th day onwards. The study 2 revealed a significant increase in the total quantity of CD34+ cells percentage increased from 0.91% to 2.26% on day 7 (n=4), 1.53% to
2.93% on day 14 (n=1) and 1.32% to 11.71% (n=3) on day 21 on an average quantified using flow cytometry.

**Conclusion:**

This study has shown that ex vivo expansion of CD34+ cells with and without isolation is feasible. This technology may become a good tool to hemato-oncologists who may in need of a precious matched unit to be expanded multifold or may cryopreserve the donors' bone marrow for subsequent application to same or similar patients. Patients and health adults who haven’t had an opportunity to store their cord blood also now have an option to store their own bone marrow cells in several small aliquots which could be thus expanded for future application when needed.