NICOTINAMIDE ATTENUATES IN VITRO DIFFERENTIATION AND ELEVATES BONE MARROW HOMING POTENTIAL OF CULTURED CD34+ CELLS

Topic: Hematopoietic cell expansion in vitro
T. Peled[1], S. Adi[1], I. Peleg[1], N. G. Rosenheimer[1], A. Peled[2];
[1]Gamida Cell, Jerusalem, ISRAEL, [2]Hadassah University Hospital, Jerusalem, ISRAEL.
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Experimental evidence suggests that progenitor cells generated by cytokine-supplemented-cultures lose some of their homing capacity. This critical change may compromise their biological function and potential clinical utility, despite their efficient numerical expansion.
We have previously shown that nicotinamide (NA), a specific inhibitor of NAD(+)-dependent ADP-ribosyltransferases, inhibits in vitro differentiation of hematopoietic progenitors. Three-week treatment of cord blood (CB) derived cultures with NA shifted the balance toward the early progenitor cell compartment, resulting in a CD34+ cell fraction highly enriched with CD34+CD38-Lin- cells (18.6±3%) compared with control cultures (0.7±0.06%), and further augmented long-term culture potential.

In the present study we evaluated homing related parameters of NA-treated, ex-vivo expanded cells. To this end, CB-derived CD34+ cells were cultured for 3-weeks with SCF, TPO, IL-6 and FLT3, ±NA (5mM). FACS-analysis of integrin receptors demonstrated comparable percentages of VLA-4 and LFA-1 in NA-treated and control cultures. Percentages of CD34+CXCR4+ cells were higher in NA-treated cultures, while percentages of dipeptidylpeptidaseIV(CD26) were 50% lower ($n=4, p<0.05$). Re-isolated CD34+ cells from 3-week NA-cultures displayed increased migratory activity toward an SDF1 gradient over control culture-derived CD34+ cells (36±19% and 11±4%, $n=3$, $p<0.05$, respectively).
To evaluate homing potential, NOD/SCID mice were transplanted with $20x10^6$ cells derived from 3-week cultures treated with or without NA or with non-expanded cells. Marrow homing of human cells was evaluated 24hr post-transplantation by FACS. Data pooled from two consecutive experiments demonstrate that homing of CD45+CD34+ cells was 6 and 2-fold higher in the NA group and the control group, respectively, relative to the non-expanded cell group ($n=14, p<0.05$). Overall homing of CD45+ cells was comparable in all groups.
Given that, together with levels of progenitor content, homing efficiency is likely a critical preliminary requirement for efficacious cell-based therapeutics, the results of this study suggest the clinical utility of CD34+ cells cultured in the presence of NA