remained difficult to assess given the requirement of human thymus tissue, and the limited number of progenitor T-cells that can be readily analyzed.

[0010] Previous work from the inventors' laboratory established that human T-lineage differentiation can be induced from umbilical cord-blood (UCB)-derived HSCs cocultured with OP9-DL1 cells (La Motte-Mohs et al., 2005). The inventors showed normal stage-specific expression of various cell surface molecules, including the generation of immature DP T-lineage cells. However, these studies were not performed using quantitative clonal analyses, and it was unresolved whether different UCB $CD34^+$ subsets could give rise to T-lineage cells and whether Delta-like/Notch signals influence the T-progenitor frequency of $CD34^+$ UCB cells. Additionally, it was unclear whether functional T-cells could be generated. Finally, the inventors' initial studies (La Motte-Mohs et al., 2005) showed that during the early stages of HSC/OP9-DL1 differentiation a population of cells resembling T-progenitors became apparent, however the potential of these cells to serve as effective T-cell progenitors was not addressed.

Summary

[0011] ~~The inventors examined the e~~Early stages of human T-cell development in vitro were examined, and ~~performed~~ limiting dilution and single-cell assays to address the T-cell progenitor frequency of various UCB-derived $CD34^+$ stem/progenitor subsets were performed. The ~~inventors assessed the~~ effect of Delta-like/Notch interactions in enhancing T-cell progenitor potential among Notch-signaled $CD34^+$ subsets was assessed. Furthermore, using limiting dilution thymus-reconstitution approaches, ~~the inventors~~ findings revealed that different tissue culture-derived T-progenitor subsets vary in their thymus-engrafting effectiveness, although these cells display a similar potential to give rise to T-lineage cells when assayed on OP9-DL1 cells. In particular, two distinct subsets, $CD34^+$ $CD7^{++}$ $CD5^-$ $CD1a^-$ (proT1) and $CD34^+$ $CD7^{++}$ $CD5^+$ $CD1a^-$ (proT2) were analyzed. It was shown~~The inventors also showed~~ that mature functional T-cells are generated in vitro, and that these cells upon TCR-stimulation display T-cell effector-function. The pro-T cells can also give rise to natural killer (NK) cells when cultured with IL-15.

[0012] Together, these findings support the use of the progenitor T cells (pro-T cells) for the generation and study of human T-cells and NK cells, and provide support for the use of in vitro-generated pro-T cells, mature T-cells and NK cells in cell-based immune-reconstitution approaches.

[0012a] In one aspect, there is disclosed herein a use of an effective amount of a progenitor T cell having a phenotype $CD34^+CD7^+CD5^+CD1a^-$ in the manufacture of a medicament for treating a condition requiring an increase in the number of T cells in an animal in need thereof.

[0012b] In another aspect, there is disclosed herein an isolated progenitor T cell having a phenotype $CD34^+CD7^+CD5^+CD1a^-CD2^-$.

[0012c] In yet another aspect, there is disclosed herein an isolated progenitor T cell having a phenotype $CD34^+CD7^+CD5^+CD1a^-CD2^-$ for treating a condition requiring an increase in the number of T cells in an animal in need thereof.

[0012d] In a further aspect, there is disclosed herein a pharmaceutical composition comprising an isolated progenitor T cell as defined herein and a pharmaceutically acceptable diluent or carrier.

[0012e] In yet another aspect, there is disclosed herein a method of generating a pro-T cell having a phenotype $CD34^+CD7^+CD5^+CD1a^-$ comprising (a) culturing a sample comprising stem cells or progenitor cells with cells that express a Notch ligand and (b) isolating pro-T cells and (c) fractionating said progenitor T cells to obtain said progenitor T cell having the phenotype $CD34^+$ $CD7^+$ $CD5^+$ $CD1a^-$.

[0013] Accordingly, one example~~aspect~~ of the present application provides an isolated progenitor T cell having the phenotype $CD34^+CD7^+CD1a^-$. In one embodiment, the isolated progenitor T cell has the phenotype $CD34^+CD7^+CD5^-$ $CD1a^-$. In another embodiment, the isolated progenitor T cell has the phenotype $CD34^+CD7^+CD5^+CD1a^-$.

[0014] In another example~~aspect~~, the present application provides a pharmaceutical composition comprising an isolated progenitor T cell in admixture with a suitable diluent or carrier.

[0015] In another example~~aspect~~, the present application provides the use of the human progenitor T cells in all applications, including preparing mature T cells, preparing NK cells, engrafting a thymus, immune reconstitution, the treatment of conditions requiring an increase in T cells as carriers for genes used in gene therapy.

[0016] Other features and advantages of the present application will become apparent from the following detailed description. It should be understood, however, that the detailed description and the specific examples while indicating preferred embodiments of the application are given by way of illustration only, since various changes and modifications within the spirit and scope of the application will become apparent to those skilled in the art from this detailed description.

Brief description of the drawings

[0017] Advantages of the present application can be readily appreciated as the same becomes better understood by reference to the following detailed description when considered in connection with the accompanying drawings. The following is a brief description of the drawings, which are presented only for the purposes of further illustrating the applications and not for the purposes of limiting the same.