Insights in Stem Cells

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Stem Cell Research 2017- Metadichol and CD34 Expression in Umbilical Cord Cells - Raghavan PR - Nanorx Inc

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Introduction:

Molecules that modulate the fate of adult or embryonic stem cells can facilitate the use of stem cell therapies for a multitude of diseases. The well-known characterized adult stem cells are hematopoietic stem cells (HSC). The expected clinical potential did not materialize due to the lack of defined culture conditions for their expansion . Human umbilical cord (CB) blood is rich in hematopoietic stem cells (HSCs) similar to those found in bone marrow. CB transplants are increasing every year worldwide. The growth of blood banks that store CB and the clinical data that support this point to HLA transplants incompatible with a low risk of b host disease (GVHD) [5]. The widest use of CB is limited by the low number of CSH per unit, and most CB units do not have enough stem cells for adults. Much work has been devoted to the development of technologies for the ex vivo expansion of HSCs to allow the transplantation of CB. Low numbers of hematopoietic stem and progenitor cells in cord blood units limit their widespread use in human transplant protocols. A characteristic of hematopoietic stem and progenitor cells is the presence of CD34 antigen cells which are multi-potent stem cells with automatic renewal which lead to all the blood cells of the immune system and to erythrocytes) and lymphoid cells (T cells, B and NK cells cells) lines . The highly specialized cells that result from HSC are essential for defending the body from infection and disease. Today, bone marrow transplants use stem cells and also harness their potential to regenerate damaged tissue. Cord blood cells have relatively long telomeric DNA compared to their peripheral blood or bone marrow analogs . Cord blood cells are capable of hematopoiesis longer. They generate more divisions and produce a greater number of descendants (daughter cells). The increase in expression of the CD34 antigen mainly precedes cell differentiation. Hematopoietic cell transplants have played an essential role in the treatment of disease. They are very promising for clinical application involving gene therapy, tolerance induction to facilitate transplants of allogenic or xenogenic organs. Recent work shows that the potential of HSC is limitless for generating whole organ systems . Boitano and colleagues recently reported a purine derivative called SR1 which greatly expands human HSC CBs in culture . The authors examined a chemical library of 100,000 heterocyclic compounds for their ability to increase the number of CD34 CB cells in culture. They showed that SR1 supports a 50-fold multiplication of CD34 cells. SR1 is an antagonist of the aryl hydrocarbon receptor protein (AhR), which generally intervenes in xenobiotic responses but has also been implicated in the regulation of hematopoietic stem / progenitor cells. The suppression of AhR resulted in sustained proliferation of CD34 cells in culture. A recent clinical trial of ex vivo CD34 cells from expanded umbilical cord blood used SR-1 and demonstrated better transplantation and better early recovery of leukocytes, suggesting that ex vivo expansion of HSC might be potentially feasible.

Likewise, co-cultures of CD34 cord blood cells with mesenchymal stromal cells led to expression of CD34 cells by a factor of 30.1, improving the time required for the neutrophil transplant at 15 days, against 24 days in recipients who received an unhandled cord. blood CD34 cells . Growing evidence indicates that targeting metabolism and cell stress for HSC expansion could potentially lead to successful HSC expansion approaches for transplant therapies in the future . Metadichol® [17] is a lipid emulsion of long chain alcohols, and we have recently shown that it is an inverse agonist of AHR [18], the only one in the medical literature. Given Boitano's work that AHRs develop CD34 cells in UBC (umbilical cord cells), our results show that metadichol, which is a safe food ingredient, can increase the expression of CD34. in the blood of the umbilical cord at the picogram level.

Reagents

Antibody: CD34-FITC conjugates: BD Pharmingen

Cell Line: Umbilical Cord (UC) cells were sourced from Hi-Media labs. Germany, they were human Wharton's

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jelly cord blood cells. They were not frozen and expanded after five passages.

Procedure:

Culture 1 × 106 cells in a 6-well plate containing 2 ml of complete media.After 24 h of incubation, cells are treated with 1 pg, 100 pg, 1 ng, 100 ng and 1µg in serum-free DMEM media and incubated for 72 h. After 72 Hr. of treatment, cells were collected and, pelleted cells at 4000 rpm for 5 minutes at room temperature and discard the supernatant. Washed the cell pellet twice with 1X PBS. The cell pellet suspended in 100 µL of Sheath fluid and incubated with CD34-FITC antibody for 20mins in the dark. Post incubation, the cells were once washed with 1X PBS and resuspended in Sheath fluid. The treated and untreated cell populations were determined using FACS Caliber (BD Biosciences, San Jose, CA).

Results:

UC cells treated with Metadichol at 1 pg, 100 pg, 1 ng, 100 ng, and 1 µg of for 72 hrs. Cells treated at 1 ng has shown the highest increase in expression of CD34 compared to untreated Control. The cells treated at 1pg, 100pg has demonstrated the multiplicity of CD34 expression as indicated by peak shift compared to treatment with 1 ng, 100 ng, and 1 µg. The fold increase is at various concentrations is difficult to calculate as the control cells, the signals for CD 34 look more like background noise. Nonetheless, the CD34 cell expression at 100 pg is seen and is very significant.

References

1. Mimeault (2007) Stem cells: a revolution in therapeutics-recent advances in stem cell biology and their therapeutic applications in regenerative medicine and cancer therapies. Clin Pharmacol Ther 82: 252. [PubMed]

2. Kondo M (2003) Biology of hematopoietic stem cells and progenitors: implications for clinical application. Annu Rev Immunol 21:759. [PubMed]

3. Hofmeister CC (2007) Ex vivo expansion of umbilical cord blood stem cells for transplantation: growing knowledge from the hematopoietic niche. Bone Marrow Transplant 39:11. [PubMed]

4. Song Chou (2010) Expansion of Human Cord Blood Hematopoietic Stem Cells for Transplantation, Cell Stem Cell 7: 427-428. [PubMed]

5. Hwang WY (2007) A meta-analysis of unrelated donor umbilical cord blood transplantation versus unrelated donor bone marrow transplantation in adult and pediatric patients. Biol Blood Marrow Transplant. 13: 444-453. [PubMed]

6. Anna H (2015) Characteristics of hematopoietic stem cells of umbilical cord blood Cytotechnology. 67: 387-396. [PubMed]

7. Jackson KA, Mi T, Goodell MA (1999) Hematopoietic potential of stem cells isolated from murine skeletal muscle. Proc Natl Acad Sci USA 96: 14482- 14486. [PubMed]

8. Jackson KA (2001) Regeneration of ischemic cardiac muscle and vascular endothelium by adult stem cells. J Clin Invest 107: 1395-1402. [PubMed]

9. Goodell MA (2001) Stem cells: is there a future in plastics? Curr Opin Cell Biol 13: 662-665.

10. Horwitz EM (1999) Transplantability and therapeutic effects of bone marrow derived mesenchymal cells in children with osteogenesis imperfecta. Nat Med 5: 309-313. [PubMed]

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