Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Elsevier hereby grants permission to make all its COVID-19-related research that is available on the COVID-19 resource centre - including this research content - immediately available in PubMed Central and other publicly funded repositories, such as the WHO COVID database with rights for unrestricted research re-use and analyses in any form or by any means with acknowledgement of the original source. These permissions are granted for free by Elsevier for as long as the COVID-19 resource centre remains active.
302 Somatic Stem Cells: Hematopoietic Stem/Progenitor Cells ENDOTHELIAL PROGENITOR CELLS OVEREXPRESSING ENDOTHELIAL NO-SYNTHASE MAY IMPROVE INFARCT HEALING: RESULTS FROM THE ENHANCED ANGIOGENIC CELL THERAPY – ACUTE MYOCARDIAL INFARCTION (ENACT-AMI) TRIAL
D. J. Stewart1, M J Kutryk1, H Q Ly2, C A Glover1, A Dick3, K A Connelly1, S G Goodman1, H Leong-Poy1, L Carlin4, R Gaudet4, M Taljaard5, D W Courtman6
1Division of Cardiology, St Michael’s Hospital Keenan Research Centre for Biomedical Science, Toronto, ON, Canada; 2Research Centre, Institut De Cardiologie de Montreal, Montreal, QC, Canada; 3Cardiology, University of Ottawa Heart Institute, Ottawa, ON, Canada; 4Sinclair Centre for Regenerative Medicine, Ottawa Hospital Research Institute, Ottawa, ON, Canada; 5Clinical Epidemiology, Ottawa Hospital Research Institute, Ottawa, ON, Canada; 6Mount Sinai Hospital, New York, NY, United States.

Keywords: Heart Failure, Cell-based gene therapy, Angiogenesis.

Background & Aim: Introduction: The goal of the Enhanced Angiogenic Cell Therapy-AMI (ENACT-AMI) trial was to determine whether intracoronary delivery of early outgrowth endothelial progenitor cells (EPCs) engineered to overexpress endothelial NO-synthase (eNOS) would improve global left ventricular ejection fraction (LVEF) (primary outcome) and infarct size as assessed by cardiac magnetic resonance imaging (CMR) in patients with anterior wall acute myocardial infarction (AMI) treated with evidence-based therapies.

Methods, Results & Conclusion: ENACT-AMI (NCT00936819) was a double-blind placebo-controlled trial in which participants were randomized to one of 3 arms: 1) saline placebo; 2) EPCs; or 3) eNOS-transfected EPCs using a plasmid DNA vector (pVax) containing the human eNOS sequence combined with JetPEI (Polyplus). Target sample size was 100 participants. Groups were compared using analysis of covariance (ANCOVA).

The trial was terminated due to slow recruitment after 47 patients were enrolled at three Canadian sites over six years (n=18 placebo, n=15 EPCs; n=14 eNOS-transfected EPCs). The groups were comparable with respect to demographic variables including age (56.1±9.7 years), cardiac risk factors, pre-existing cardiac disease, peak CK and troponin values and baseline LVEF by cardiac magnetic resonance imaging (40.7±9.3). Intracoronary cell delivery (20M cells; 20±5 days post-AMI) was well tolerated. At 6 months, there were no significant differences in the primary endpoint of LVEF between groups (p=0.30, mean difference between the average of the two EPC groups vs. placebo: 0.5% [95% Confidence Interval (CI) -2.9% to 3.9%]). The secondary outcome of left ventricular infarct mass indexed to LV mass at 6 months demonstrated no significant difference between the average of the two EPC groups versus placebo (p=0.72); however, a significant difference was seen in those receiving eNOS-transfected EPCs compared to EPCs (-6.6; CI -12.0 to -1.1, p=0.02). Only four patients with large anterior wall AMI consistent with improved scar healing. These findings have important implications for remodelling and require confirmation in larger clinical trials.

303 Somatic Stem Cells: Hematopoietic Stem/Progenitor Cells CRYOPRESERVATION OF ALLOGENIC STEM CELL COLLECTIONS DURING COVID-19 PANDEMIC: PRODUCTS CHARACTERIZATION AND ENGRAFTMENT OUTCOME
A. Keyzner1,2, R Jakubowski1, Y Sinitsyn1, S Tindle1, S Shpontak1, U Ozbek1, L Isola1,2, C Iancu Rubin1,2
1Mount Sinai Hospital, New York, NY, United States; 2Icahn School of Medicine at Mount Sinai, New York, NY, United States.

Keywords: Allogeneic Stem and Progenitor Cells, Cryopreservation, Engraftment.

Background & Aim: The COVID19 pandemic has affected the practice of allogeneic hematopoietic stem cell transplantation at multiple levels including donor selection, evaluation and stem cells collection and processing. Due to the risk of SARS-CoV2 infection and the impact of pandemic-related restrictions on transportation and handling, on March 13 2020, the National Marrow Donor Program/Be the Match recommended cryopreservation of allogeneic stem grafts which have been traditionally infused fresh. Here we report a single center experience on cryopreservation of allogeneic products and engraftment outcome after their infusion.

Methods, Results & Conclusion: A total of 35 products [graft sources, 13 bone marrow (HPC-BM) and 22 apheresis (HPC-A)] were cryopreserved and infused at Mount Sinai Hospital between March and December 2020. The median dose of CD34+ cells was 1.57×10^6 cells/Kg and 4.46×10^6 cells/Kg for thawed BM and HPC-A (Table 1) as calculated based on the post-thaw total nucleated cells recovery of 88% and 96%, respectively. The median time between collection and freezing was 24 hours and storage time ranged from 7 to 35 for HPC-BM and 7-262 days for HPC-A products. The time to absolute neutrophil count (ANC) and platelet (PLT) engraftment in patients receiving HPC-BM was 16 and 26 days, respectively. When compared to the engraftment times observed after infusion of fresh products during 2019, there was no significant difference for either ANC or PLT (Table 1). Importantly, none of the patients receiving cryopreserved grafts experienced neutrophil engraftment.

Table 1 (abstract 303)
Comparison between fresh and cryopreserved grafts

|          | HPC, Bone Marrow | HPC, Apheresis |
|----------|------------------|----------------|
| # of patients | 10 | 13 | 28 | 22 |

CD34+ cells/kg (fresh)
- Average: 2.54E+06
- Median: 1.94E+06
- Range: 8.9E+05-6.87E+06

CD34+ cells/kg (cryopreserved)
- Average: N/A
- Median: 1.57E+06
- Range: 3.51E+06

Time to ANC engraftment
- Average: 19 days
- Median: 18 days
- Range: 11-32 days

Time to PLT engraftment
- Average: 27 days
- Median: 26 days
- Range: 14-107 days

*p value

*Neutrophils ≥0.5×10^9/L the first of 3 consecutive days
**Platelets ≥20×10^9/L the first of 3 consecutive days 7 days after the last platelet transfusion.
HPC-BM experienced primary graft failure. The time to engraftment in patients receiving HPC-A was 14 days for ANC and 23 days for PLT. While there was no significant difference between the times to ANC or PLT engraftment in 2020 vs. 2019 (Table 1), comprehensive analyses including long-term clinical outcomes are required to determine the impact of cryopreserved allografts on transplantation. Of note, 3 of the patients receiving cryopreserved HPC, A died prior to engraftment due to severe infections at days 12, 22 and 32. In summary, we report that cryopreservation resulted in acceptable TNC recovery and CD34+ cell doses and infusion of thawed products did not significantly affect the time to ANC or PLT engraftment when compared to that observed after transplant of fresh products. Our results, corroborated with those of others, support the implementation of cryopreservation of allogeneic products as an adequate approach in the management of HSCT during challenging times and beyond.

304
Somatic Stem Cells: Hematopoietic Stem/Progenitor Cells
CONVERTING A LEUKEMIC TRANSCRIPTION FACTOR INTO A POWERFUL TOOL FOR LARGE-SCALE EX VIVO PRODUCTION OF HUMAN PHAGOCYTES
R. Windisch, S. Soliman, A. Hoffmann, L. Chen-Wichmann, S. Lutz, E. Kellner, E. Redondo-Monte, S. Vosberg, O. Weigert, L. Hartmann, S. Schneider, F. Beier, C. Strobl, O. Windisch, M. Schuendeln, J. Bernhagen, A. Humpe, C. Brendel, H. Klump, P. Greif, C. Wichmann
1Department of Transfusion Medicine, Cell Therapeutics and Hemostaseology, University Hospital LMU Munich, Munich, Germany; 2Chair of Vascular Biology, Institute for Stroke and Dementia Research, LMU Munich, Munich, Germany; 3Department of Anaesthesiology, University Hospital, LMU Munich, Munich, Germany; 4Department of Medicine II, University Hospital, LMU Munich, Munich, Germany; 5German Cancer Consortium (DKTK), Munich, Germany; 6German Cancer Research Center (DKFZ), Heidelberg, Germany; 7Laboratory for Leukemia Diagnostics, Department of Medicine III, University Hospital, LMU Munich, Munich, Germany; 8Institute of Human Genetics, University Hospital, LMU Munich, Munich, Germany; 9Department of Hematology, Oncology, Hemostaseology and Stem Cell Transplantation, Medical Faculty University Hospital Aachen, RWTH Aachen University, Munich, Germany; 10Pediatric Hematology and Oncology, Department of Pediatrics III, University Hospital Essen and the University of Duisburg-Essen, Essen, Germany; 11Munich Cluster for Systems Neurology (SyNergy), Munich, Germany; 12Division of Pediatric Hematology/Oncology, Boston Children’s Hospital, Dana-Farber Cancer Institute, Harvard Medical School, Boston, MA, United States; 13Institute for Transfusion Medicine, University Hospital Essen, Essen, Germany.

Keywords: Expansion, Differentiation, Macrophages.

Background & Aim: The key hurdle for ex vivo expansion of human CD34+ hematopoietic stem and progenitor cells (HSPCs) represents the rapid cellular differentiation after detachment from the supporting bone marrow stem cell niche. However, a few leukemia-related chimeric transcription factors, including MLL fusion proteins, are able to circumvent this issue.

Methods, Results & Conclusion: Here, we fused the coding sequence of an FKBP12-derived destabilization domain (DD) to the fusion gene MLL-ENL and subsequently expressed the protein switch (DD-ME) in human CD34+ progenitors derived from healthy donors. The DD-specific ligand Shield1 was added to regulate DD-ME protein turnover yielding massive and long-term expansion of HSPC-derived late monocytic precursors without additional driver mutations and with normal karyotype preserved. Upon removal of Shield1, the cells completely lost self-renewal and colony-forming properties and spontaneously differentiated, even after 12 months of ex vivo expansion. In the absence of Shield1, a six-day stimulation with IFN-γ, LPS, and GM-CSF triggered robust monocytic differentiation.

Immunophenotypic characterization revealed upregulation of the surface markers CD14, CD68, CD80, CD163 and MHC class I and II, concordant with monocytic morphology as judged by cytospin preparations. Upregulation of inflammatory markers such as IL-6, IL-10 and CCL2 on mRNA level was detected by qRT-PCR. Furthermore, no counter gene expression analysis covering a total of 770 myeloid specific genes revealed the cells’ identity as differentiated phagocytes. In functional assays, we demonstrated the ability of the obtained cells to migrate towards the chemokine CCL2, attach to VCAM-1 under flow and shear stress, to produce reactive oxygen species and engulf both bacterial particles and apoptotic cells. Finally, we demonstrated IgG Fc domain binding and phagocytosis of lymphoblastic tumor cells, including Daubi, Raji and patient-derived MCL cells in an antibody-dependent manner.

Taken together, we demonstrate the conversion of a leukemia-associated transcription factor to a useful tool for ex vivo blood cell production. Using this engineered protein switch, we were able to obtain HSPC-derived monocytic progenitors in large-scale and differentiate those towards functional monocytes under well-defined ex vivo conditions, both efficiently and quantitatively. This controllable system could set the stage for directed generation of patient-derived monocytes for cell-based immunotherapeutic approaches.

305
Somatic Stem Cells: Hematopoietic Stem/Progenitor Cells
IMPACT OF FANCONI ANEMIA GENOTYPE ON OUTCOME AFTER HEMATOPOIETIC STEM CELL TRANSPLANT: A SINGLE CENTRE EXPERIENCE OF 20 YEARS
S. Jain, A. Mauguen, A. R. Djavid, J. A. Kennedy, A. Smogorzewska, M. Walsh, J. J. Boelens, M. Cancio
1Pediatric BMT and cellular therapy, Memorial Sloan Kettering Cancer Center, New York, NY, United States; 2Department of Epidemiology and Statistics, Memorial Sloan Kettering Cancer Center, New York, NY, United States; 3Department of Pediatric BMT and cellular therapy, Memorial Sloan Kettering Cancer Center, New York, NY, United States; 4Department of Medicine, Memorial Sloan Kettering Cancer Center, New York, NY, United States; 5Laboratory of Genome Maintenance, The Rockefeller University, New York, NY, United States; 6Columbia University Vagelos College of Physicians and Surgeons, New York, NY, United States; 7Stem Cell Transplantation and Cellular Therapies, Memorial Sloan Kettering Cancer Center, New York, NY, United States.

Keywords: Fanconi anemia, Genotype, Transplant.

Background & Aim: The effect of Fanconi anemia (FA) genotype on hematopoietic stem cell transplant (HSCT) outcome is not well elucidated in literature. Our aim was to compare HSCT outcome of patients with FANCA vs non- FANCA mutations.

Methods, Results & Conclusion: Retrospective data from hospital electronic records of FA patients who underwent HSCT from April 1998 to April 2019 were retrieved. FANCA mutation group was compared with non- FANCA for demographics, physical abnormalities, HSCT characteristics and outcome. Fisher’s exact, Wilcoxon and log rank tests, and Kaplan-Meier were used for analysis. Event was defined as primary graft failure, relapse or death.

Data on complementation group was available for 50 out of 53 patients. FANCA mutations were found in 38/50 (76%) patients; FANCG in 5/50 (10%); FANCC in 4/50 (8%); FANCD1, FANCD2 and FANCL each in 1/50 (2%) patients. Most common physical abnormality was pigmentary abnormalities in 37/74%. Microcephaly was higher in the non-FANCA group (5/12 vs 5/38, p = 0.05). In both FANCA and non-FANCA patients, half underwent HSCT for severe aplastic anemia (SAA) and half for MDS/AML. The HSCT characteristics were similar between the two groups except donor population(Table 1).