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Developments in Clinical Cell Therapy

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Abstract

Immunotherapy has become an important part of hematopoietic stem cell (HSC) transplantation and cancer therapy. Regenerative and reparative properties of somatic cell-based therapies hold tremendous promise for repairing injured tissue, preventing and reversing damage to organs, and restoring balance to compromised immune systems. The principles and practices of the diverse aspects of immune therapy for cancer, HSC transplantation and regenerative medicine have many commonalities. This meeting report summarizes a workshop sponsored by the National Heart, Lung and Blood Institute (NHLBI) and Production Assistance for Cellular Therapies (PACT), held on 23 – 24 April 2009 at the National Institutes of Health (NIH, USA). A series of scientific sessions and speakers highlighted key aspects of the latest scientific, clinical and technologic developments in cell therapy, involving a unique set of cell products with a special emphasis on converging concepts in these fields.

Keywords

cancer; *cell therapy*; *hematopoietic stem cell transplantation*; *regenerative medicine*

Workshop Mission

A workshop sponsored by the National Heart Lung and Blood Institute (NHLBI) and Production Assistance for Cellular Therapies (PACT) "*Converging Concepts in Cell Therapy*" was held on April 23–24, 2009, at the National Institutes of Health (NIH, USA). A series of scientific sessions and speakers highlighted key aspects of the latest scientific, clinical and technological developments in cell therapy involving a unique set of cell products with a

Final Remarks Cellular therapies involving are making important contributions to HSC transplantation, cancer therapy and regenerative medicine. We are privileged to work with the investigator community to enhance the biology, advance the clinical applications, and realize the promise of cell therapies.

special emphasis on converging concepts in these fields. This 2-day workshop stimulated discussions regarding the advancements made in developing cellular therapies, discussed the risks and benefits associated with the clinical use of these therapies, and provided a venue for information exchange.

Clinical Trials using Umbilical Cord Blood (UCB)-derived T-Regulatory (T-reg) Cells

Dr. David McKenna from the University of Minnesota discussed methods for establishing the best culture conditions for expansion of human peripheral blood and UCB-derived T-regs. Evidence indicates that cell-based artificial antigen presenting cells (aAPCs) are superior to beads in expanding functional T-regs, and studies are underway to confirm potency *in vivo*.

A dose-escalation study of peripheral blood-derived $CD4^+/CD25^+$ Treg cells in patients undergoing hematopoietic stem cell transplantation was recently initiated by Claudio G. Brunstein at the University of Minnesota. Also, a Phase I clinical trial investigating the safety of UCB-derived Treg cell infusion using a non-myeloablative double UCB transplant platform is ongoing at the University of Minnesota. The primary endpoints are maximum tolerated dose and dose-limiting toxicity of UCB-derived Treg cells. Thus far, the study has shown that infusing *ex vivo* expanded and activated UCB-derived T-regs is safe in patients with hematologic malignancies at tested dose levels, and UCB-derived T-regs are detectable up to 14 days post infusion. A new trial of UCB-derived Treg cells involving restimulation with anti-CD3/28 beads to provide larger doses of functional T-reg cells is underway. Efforts led by Dr. Bruce L. Levine from the University of Pennsylvania are to expand UCB-derived T-regs with cell-based aAPCs [1].

Mesenchymal Stem Cells (MSCs) in Phase I Cardiac Repair and Phase II Acute Graft Versus Host Disease (GVHD) Trials

Dr. Adrian Gee from the Baylor of College of Medicine briefly discussed the expanded use of MSCs in cardiac and cancer applications as evidenced by the tremendous increase in publications supporting these applications. MSCs are a promising cell source for the repair of ischemic cardiac damage because they can form a stem cell niche and secrete growth factors and cytokines. Dr. Ian McNiece from the University of Miami presented data on studies in pig models that found reduced scar size in both acute and chronic treatment settings, GATA-6 and Ki-67 positive myocytes, MSC washout, reduced myocyte apoptosis, and increased tissue perfusion and mature arterioles. Phase I clinical trial dosing studies are now examining the use of MSC transplantation in patients with ischemic cardiomyopathy undergoing coronary artery bypass surgery. Dr. Katarina LeBlanc from the Karolinska Institutet is using MSCs in Phase II HSC transplantation trials to treat steroid-resistant severe acute GHVD [2]. The Karolinska Institutet group has also co-transplanted MSCs and HSCs in patients undergoing high-risk HSC transplant, without rejection or graft failure. Several of these patients had rejected a previous graft, but all had good engraftment after co-transplantation [3].

Adoptive T-Cell Therapy Clinical Trials involving Antigen-Specific T Cells

Dr. Cliona Rooney of Baylor College of Medicine described how T cells are effective for the treatment of viral infections in immunosuppressed patients. The treatment of malignancy is more challenging, but tumor-specific T cells have produced clinical responses and complete remissions of Hodgkin's disease and melanoma [4].

Dr. Philip D. Greenberg from the University of Washington presented data on a Phase I clinical trial conducted at University of Washington Medical Center that assessed tumor associated

antigen targets in metastatic melanoma using a mixed population of leukemic cells with a T cell clone that recognizes the antigen. The study demonstrated that the adoptively transferred T cell clones persist *in-vivo* and mediate antigen-specific immunity [5]. Dr. Shelly Heimfeld from the Fred Hutchinson Cancer Research Center in Seattle, WA spoke on genetically engineered leukemia-specific T cells that are being used in Phase I/II clinical trials using peripheral blood mononuclear cells and a large-scale rapid T-cell expansion protocol to treat lymphoma [6].

Engineered T Cells used in the Treatment of Melanoma and HIV

Dr. Bernard Fox of the Earle A. Chiles Institute in Oregon opened the engineered T cell session with a discussion of how T cells can be engineered by inserting genes than serves as a marker so that these cells can be tracked, adding a function that alters the T cells' function or growth (using growth factors, effector cytokines, or effector molecules), or modifying the cells so that they traffic differently to tumor sites (using chemokine receptors). It is also possible to make T cells more resistant to other negative signals or use T cell receptor gene transfer to redirect the T cell to the tumor site.

Dr. Steven A. Rosenberg from the National Cancer Institute discussed how T cell-based immunotherapy can mediate the regression of large vascularized, invasive metastatic melanoma in humans [7]. The ability to genetically modify human T cells opens the possibility of extending cell transfer immunotherapy to patients with common epithelial cancers. Dr. Carl H. June and his group from the University of Pennsylvania are testing the expression of antisense to the HIV envelope gene in CD4 cells using a second-generation lentiviral vector. Within 7–10 days, the process produces nearly a whole body's worth of T cells that are 100% transduced. A Phase I clinical trial conducted using the lentiviral vector was well tolerated and exhibited sustained gene transfer [8].

Adoptive Cancer Immunotherapy Clinical Trials involving Natural Killer (NK) Cells

Dr. David Stroncek highlighted issues regarding the production, expansion and quality of NK cells. Hematopoietic stem cell transplantation is permanent and can lead to GVHD or transplant-related mortality. In contrast, adoptive transfer is safer and more transient and cells can expand *in vivo*. Dr. Jeffrey S. Miller from the University of Minnesota and colleagues published their platform using haploidentical NK cell infusions. The investigators chose the lymphodepleting chemotherapy of high-dose cytoxan and fludarabine to promote the *in vivo* expansion of melanoma-specific cytotoxic T lymphocytes. A clinical update in 2008 ten of the 32 patients in this trial experienced remission. Of these patients, three received an allogeneic transplant and experienced disease-free survival for at least 2.5 years. The investigators found no correlation with KIR ligand mismatch. The data suggest that *in vivo* expansion is important for efficacy [9]. Researchers have recently explored methods to alter tumors as targets to make them more susceptible to killing by autologous NK cells. VELCADE® (bortezomib) is an FDA-approved proteasome inhibitor used to treat multiple myeloma. Dr. Richard W. Childs presented preliminary data of a Phase I trial being conducted in the NHLBI is evaluating the safety and the anti- tumor effects of escalating doses of adoptively infused *ex vivo* expanded autologous NK cells against metastatic cancers or hematological malignancies sensitized to NK cytotoxicity with Bortezomib. Irradiated EBV-LCL cells are used as feeder cells to expand these NK cell populations under GMP conditions [10]. The study is currently accruing patients into NK cell dose level 3, with 9 patients having received a total of 19 NK cell infusions, with 19/20 *ex vivo* NK cultures expanding successfully to achieve the target NK cell dose. Five patients with stable disease following NK cell therapy and have gone on to receive subsequent cycles of NK cells (range 2–5 cycles).

Dendritic Cell (DC)-based Cancer Vaccine Trials

Dr. Theresa Whiteside from the University of Pittsburgh led a discussion on dendritic cell-based vaccines. Alpha DC1-based vaccines effectively induce effector T cells (CTLs and Th1) in cancer patients who frequently are immunosuppressed and require potent antigenic stimulation for mounting memory responses. Polarized alpha DC1 make effective vaccines because they display a unique set of chemokines and preferentially interact with effector T cells rather than T-regs. Dr. Pawel Kalinski of the University if Pittsburgh discussed current Phase I trials which are comparing the effectiveness of intralymphatic DC delivery with standard delivery routes and the effectiveness of alpha DC1s to that of standard DCs [11]. A Phase I/II trial of therapeutic immunization with autologous DCs pulsed with autologous, inactivated HIV-1-infected, apoptotic cells is being conducted by Dr. Sharon Riddler at the University of Pittsburgh. The hypothesis is that this therapeutic vaccination approach is safe for HIV-infected adults and will result in a decrease in the HIV-1 RNA set point [12]. Each participant will receive three vaccines of 10 million cells each at 2-week intervals followed by an interruption of the ART and one booster vaccine dose. The post-interruption viral load will be compared to the pre-treatment viral load and immunologic correlates of vaccine response will be investigated.

Understanding clinical efficacy of cell-based therapies *in vivo* - Cell Imaging Techniques

Dr. Marianna Sabatino from NIH provided an overview of cell imaging and trafficking techniques. Cellular imaging is an important strategy to understand the effects of cell therapy *in vivo*. Different techniques have been developed and among these radionuclide technology and (MRI) have being investigated the most due to their high resolution and clinical applicability. There are two major methods to label cells for imaging studies: directly with super-paramagnetic iron oxide agents or radionuclides or indirectly using reporter genes transferred into the cells before administration. The genes can later be traced using a specific radio- or optical labeled probes allowing repeatable tracking over a long period of time. Dr. Zhenghong Lee of the University Hospitals Case Medical Center described a similar indirect labeling approach which uses porous ceramic cubes loaded with transduced stem cells for quantitative PET imaging in addition to the popular luminescent imaging. Validation assays have shown that MSCs retain their properties after *in vivo* induction [13]. Dr. Eric T. Ahrens from Carnegie Mellon University has been focusing on ex vivo labeling to sort and select cell populations using perfluorocarbons. Cationic perfluorocarbons have been introduced as contrast agents for cell labeling. The main advantage of one such agent, 19F MRI, is that it allows imaging of the cells without any background, since there are no endogenous fluorine atoms present in the body [14].

Erin Simonds of Stanford University discussed the use of phosphospecific flow cytometry as an imaging strategy that uses intracellular signaling as it occurs and its particularly value when materials are scarce, samples are heterogeneous, or networks are complex [15,16].

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