Keynote Speech *"Genetic reprogramming of T cells for human applications"* by Dr. Laurence Cooper
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***Abstract***

The immune system can be successfully manipulated *ex vivo* for *in vivo* applications. Proof-of-principle has been established for T cells genetically modified to redirect specificity through constitutive expression of chimeric antigen receptors (CARs) and T-cell receptors (TCRs). The associated laboratory processes encompass, activation, propagation, and genetic manipulation of the T-cell product all in compliance with current good manufacturing practice (cGMP) for Phase I/II trials. These “first generation” bio-engineering technologies have demonstrated success targeting some hematologic malignancies and solid tumors. For example, CARs targeting cell surface CD19 (a B-lineage antigen) and GD2 (a disialoganglioside expressed on tumors of neuroectodermal origin) demonstrate responses in recipients with B-cell malignancies and neuroblastoma, respectively. For example, TCR targeting intracellular NY-ESO-1 (a cancer‐testis antigen) in the context of human leukocyte antigen (HLA) show responses in recipients with multiple myeloma and synovial sarcoma. Follow-on studies within the first-generation technologies include (i) identification of safe CAR and TCR targets (especially to target solid tumors) and the use of suicide genes for conditional *in vivo* ablation, (ii) experimenting with alternative cellular templates other than abTCR+ T cells to express CAR and TCR (*e.g.*,  gdTCR+ T cells, NKT cells, NK cells), (iii) reducing burden on manufacturing to produce T cells in real time (such as delivering off-the-shelf T cells with potential for “on demand” disease control), and (iv) improvements in bio-processing to bias the infusion product towards “young” T cells that maintain their replicative potential (preserve telomere length) and have capacity for self-renewal or memory (*e.g.*, infusing T cells with central memory phenotype). This suite of first-generation technologies in aggregate will provide therapeutic benefits for patients with cancer. However, additional “generations” of technologies will be needed for T-cell therapy to achieve its full potential and thus to address one of the hall marks of cancer, namely its genetic instability. This leads to heterogeneity of expression of many “targeted” antigens within a given patient’s tumor and heterogeneity of expression between potential patients. The former gives rise to antigen-escape variants (*e.g.*, CD19neg malignant B cells) and relapse after monotherapy upon infusion of T cells and the latter results in the need to customize the T-cell products for different recipients. Thus, “second-generation” technologies will be based on combinations. These will likely encompass (i) co-infusing T cells with multiple specificities (*e.g.*, more than one CAR species), (ii) combining the introduced immunoreceptor with cytokine (*e.g.*, CAR and interleukin (IL)-15) to improve potency, (iii) combining genetic elements for the conditional and induced (controlled) expression of introduced transgenes, (iv) combining genetic insertion (*e.g.*, to express CAR and TCR) with genetic editing (*e.g.*, to eliminate checkpoints) and (v) combining T-cell therapy with other immunotherapies (*e.g.*, checkpoint inhibitors) to recycle effector functions within the tumor microenvironment. This second-wave of T-cell therapies will help to bridge the gap between the therapeutic responses seen between B-cell leukemias/lymphomas and other types of malignancies. A third generation of technologies will likely be needed before T cells can reliably and efficiently execute all manner of tumor cells. This will be based on the targeting of neo-antigens which arise from driver mutations expressed exclusively by the patient’s malignant cells. T-cell therapy targeting neo-antigens will require embracing technologies which build on the prior two generations, but also harness the ability to identify individualized targets and to re-program the effector cells with TCRs specific for neo-antigens. This approach will need to account for the dual pressures of (i) the time to generate the T-cell products and (ii) the cost. The former will be addressed through efficient processing and the latter through the use of non-viral approaches to gene transfer, such as using the DNA plasmids from the *Sleeping Beauty* system. Programs that undertake first, second, and third generation approaches to immunotherapy are destined to develop powerful therapeutics genetically programmed to carefully address the recipient with cancer.