April 19, 2017

Equity Research

WELLS FARGO SECURITIES

Deep Dive on Emerging Cell Therapies for Cancer

Sector Rating: Biotechnology, Overweight

Company Name	Rating	Price 04/19/17	FY EPS 2017E	FY EPS 2018E	FY P/E 2017	FY P/E 2018
Biotechnology						
Adaptimmune Therapeutics PLC (ADAP)	2 V	$5.41	$-0.21	$-0.22	NM	NM
Amgen, Inc. (AMGN)	2	161.26	12.21	12.65	13.2x	12.8x
Bellicum Pharmaceuticals, Inc. (BLCM)	1 V	13.46	-2.66	-2.53	NM	NM
bluebird bio, Inc. (BLUE)	1 V	83.95	-3.84	-4.19	NM	NM
Celgene Corp. (CELG)	1	122.92	7.25	9.20	17.0x	13.4x
Cellectis SA (CLLS)	1 V	22.13	-2.34	-2.95	NM	NM
Fate Therapeutics, Inc. (FATE)	1 V	4.49	-0.97	-0.81	NM	NM
Juno Therapeutics, Inc. (JUNO)	1 V	24.61	-2.74	-3.17	NM	NM
Kite Pharma, Inc. (KITE)	2 V	79.55	-9.08	-4.37	NM	NM
Lion Biotechnologies, Inc. (LBIO)	1 V	6.30	-1.15	-1.24	NM	NM
ZIOPHARM Oncology, Inc. (ZIOP)	2 V	7.08	-0.55	-0.59	NM	NM

Source: Company data and Wells Fargo Securities, LLC estimates *1= Outperform, 2 = Market Perform, 3 = Underperform, V = Volatile,*
= Company is on the Priority Stock List *NA = Not Available, NC = No Change, NE = No Estimate, NM = Not Meaningful*

- The Wells Fargo Securities biotechnology has undertaken a detailed review of the adoptive cellular therapy (ACT) space, encompassing CAR-T, engineered T-cell receptor (eTCR), and tumor infiltrating lymphocyte (TIL) therapeutics for cancer. With more than 250 ongoing trials described in our report and more than 100 investigational new drug (IND) applications pending, we view the ACT space as one of the most complex and exciting areas of biotechnology drug development and one of the most significant next product waves for our coverage universe.
- Following recent research initiations of CAR-T therapeutic leaders Kite Pharma (KITE) and Juno Therapeutics (JUNO), and ahead of expected approval of KITE's leading CD19 CAR-T therapeutic Axi-Cel for diffuse large B-cell lymphoma (DLBCL), filing by competitor Novartis for CTL019, and pivotal trial advancement for JUNO's JCAR017 in DLBCL, we see significant opportunity for product profile improvement even within the CD19 CAR-T space. In particular, with a 90% rate for CD19 CAR-T products in ALL as a guide, we believe that a 30% CR rate in DLBCL supporting Axi-Cel filing can certainly be improved upon. In this report we focus on important aspects of CAR-T cell composition, CAR-T construct, and the importance of different co-stimulatory domains, and the manufacturing process and avoidance of "exhausted cells," as well as other factors in optimizing CAR-T therapeutics. We favor JUNO among leaders in this regard.
- With expertise in gene therapy, gene editing, immunology, cell engineering, and transplant medicine converging on the development of next-generation adoptive cellular therapies (ACT), we believe that extending early success beyond CD19 CAR-T to more difficult targets will be complex, and expect progress beyond CD19 leaders. In particular, we highlight recent success for gene therapy leader Bluebird Bio (BLUE) in engineering a BCMA CAR-T for myeloma with a 100% CR rate and absent cytokine release syndrome, with a superior profile to large Pharma CAR-T leader Novartis (NVS), where response rates were lower and CRS was prominent. We expect myeloma to represent a significant market opportunity for adoptive cellular therapies (ACT), and with success of antibody therapeutic DARZALEX in targeting CD38, we highlight a CD38 CAR-T for myeloma emerging from Cellectis (CLLS) and note the important role of its gene editing platform to remove CD38 from the CAR-T product so it doesn't turn on itself. (*Continued on next page*).

Jim Birchenough, MD, Senior Analyst
(415) 947-5470
jim.birchenough@wellsfargo.com
Chuck Whitesell, Associate Analyst
(212) 214-5067
chuck.whitesell@wellsfargo.com
Nick Abbott, Associate Analyst
(206) 542-2492
nick.abbott@wellsfargo.com
Yanan Zhu, Associate Analyst
(415) 396-3194
yanan.zhu@wellsfargo.com

BIOTECH041617-194148Please see page 92 for rating definitions, important disclosures and required analyst certifications. All estimates/forecasts are as of 04/19/17 unless otherwise stated. 04/19/17 13:08:34 ET

Wells Fargo Securities, LLC does and seeks to do business with companies covered in its research reports. As a result, investors should be aware that the firm may have a conflict of interest that could affect the objectivity of the report and investors should consider this report as only a single factor in making their investment decision.

Together we'll go far

- Ultimately, the greatest opportunity for adoptive cellular therapy (ACT), in our view, lies within the larger category of solid tumors, where exclusive targets have been more elusive and where early patient deaths have highlighted the huge challenge. With a limited number of large protein targets for CAR-T therapeutics, we see greater opportunity for novel intra-cellular targets for engineered T-cell receptor (eTCR) therapeutics. Early success with eTCR leader Adaptimmune (ADAP) with an eTCR targeting NY-ESO1 is promising, with the potential for broader success, in our view; however, we believe that finer control of eTCR will be required for targets that likely exist to some extent on normal tissues. To that end, we highlight Bellicum Pharmaceuticals (BLCM) with its controllable CAR-T and eTCR therapeutics and switches to turn on and off T-cells and suggest that a focus on other controllable ACT products could potentially be important investment opportunities. Overall, our detailed review highlights that there is a significant challenge to eTCR therapeutics in solid tumors, and that in the near term, we prefer the less elegant approach of Lion Biotechnologies (LBIO) in expanding existing tumor infiltrates (TIL, which have been selected out for tumor specificity.

TABLE OF CONTENTS

Adoptive Cellular Therapy - Executive Summary 5
Adoptive Cellular Therapy (ACT) Overview 12
History of Adoptive/Activated Cell Therapy 15
Adoptive Cellular Therapy and 3-leter acronyms - SCT, TIL, TCR and CARs 16
TILs – Considering the Process and Is there utility outside of melanoma? 19
Engineering the T Cell Receptor – Hampered by Technological Limitations? 20
Human Leucocyte Antigen (HLA) Matching – limits the utility of eTCR approaches 20
Loss of HLA is a key tumor escape pathway 21
Chimeric Antigen Receptors – broader applicability than eTCRs 23
With Increased Potency comes Increased Risk for Toxicity 24
Cytokine release syndrome, neurotoxicity and left ventricular dysfunction 25
CAR-T immunogenicity 26
Engineered TCRs – Natural vs Affinity Matured TCR Therapeutics 29
In summary - Current Applications of Adoptive Cellular Therapy (ACT) : 32
Looking to the Future of ACT 36
The Potential Role of Suicide switches to Turn off CAR-T and eTCR Therapeutics 38
Deployment switches for Boolean activation 38
On-board cytokines and chemokines 38
Activation switches and rheostats 39
Off the shelf cells for ACT 43
Graft versus Host Disease and Host versus Grafted reactions in the allogeneic setting 44
Induced Pluripotent Stem Cells, iPSC 46
Non-viral transfection and patient-specific neoantigens 48
ACT Beyond αβT cells 49
Convergence of eTCR and CARs 50
Adoptive Cellular Therapy (ACT) of the future –Summary 53
CD19 – Proof of Concept for Adoptive Cellular Therapy (ACT) 54
What is the Bar for Success of ACT in Hematologic Malignancy? Considering endpoints 54
KITE/NCI –- CD28 murine CD19 CAR (Axi-Cel) – retrovirus 57
The University of Pennsylvania (Novartis) – 41BB murine CD19 CAR- lentivirus 61
CD19/41BB/ lentivirus or CD19/CD28 /Retrovirus 65
MD Anderson (ZIOP) –CD19/CD28 with Sleeping Beauty 67

Adoptive Cellular Therapy (ACT) – A Selection of Who's Invested 68
A 67
B 69
C 70
E 73
F 73
G 74
I 74
J 74
K 75
L 76
M 77
N 77
O 78
P 78
S 78
T 79
U 79
Z 79
ACT Clinical Trial Review 80
Corporate ACT trials 80
Solid Tumor ACT Trials 84
Paying for Adoptive Cellular Therapies (ACT) 90
Medicare reimbursement 91

Adoptive Cellular Therapy -- Executive Summary

Adoptive Cellular Therapy - CAR-T, eTCR, and TILs

Adoptive Cellular Therapy (ACT) is a rapidly evolving category of targeted cell therapies for the treatment of cancer and includes chimeric antigen receptor T-cell (CAR-T) therapeutics, engineered T-cell receptor (eTCR) therapeutics, and tumor-infiltrating lymphocytes (TILs).

At its most basic level, these adoptive cellular therapies (ACT) involve genetic engineering of immune cells, most commonly, immune T-cells, outside of the body to target proteins, or protein fragments (antigens) on cancer cells.

Examples of these cancer targets include the following:

- CD19 protein on cancerous B-cells in leukemia and lymphoma, targeted by CD19 CAR-T products;
- BCMA protein on myeloma cells, targeted by BCMA CAR-T products;
- NY-ESO-1 antigen on a variety of solid tumors, and targeted by NY-ESO-1 specific eTCR products; and
- Tumor-infiltrating lymphocytes extracted from melanoma tumors, and expanded and optimized before re-introduction to the patient.

Central to the success of these adoptive cellular therapies (ACT) is identification of targets unique to cancer cells, the genetic engineering of immune cell products that hone specifically to these targets, the optimization of immune cell products to generate active tumor killing and persistent effect, the design of protocols for dosing of these immune cell products to optimize the therapeutic index, and consideration of all these factors, and more in optimizing efficacy and limiting toxicity.

Early Promise of CAR-T Therapeutics in Blood Cancers

Excitement around adoptive cellular therapeutics (ACT) has arisen from early success, primarily with CAR-T therapeutics in blood cancers, and more limited success of eTCR products and TILs products in select solid tumors, such as melanoma.

Initial success in adoptive cellular therapy (ACT) came from the CD19 CAR-T products from Juno, Kite, and Novartis, where complete response (CR) rates of about 90% have been observed in patients with acute lymphocytic leukemia (ALL) failing all available therapies. More limited success of CD19 CAR-T therapeutics has been observed in aggressive diffuse large B-cell lymphoma (DLBCL) where a 30% complete response (CR) rate with most advanced product AxiCel has been demonstrated.

Early success in targeting the CD19 protein on the surface of cancerous B-cells has recently been followed by limited success in targeting the BCMA protein on cancerous plasma cells in patients with multiple myeloma, another blood cancer.

Initial results from the National Cancer Institute (NCI) for a BCMA CAR-T demonstrated 3 responses, including 1 complete response (CR) in patients with relapsed/refractory myeloma, with more recent results from Bluebird (BLUE) and partner Celgene (CELG) suggesting a 100% overall response rate (ORR) in patients treated at the highest dose.

Despite the early success of CD19 CAR-T and BCMA CAR-T therapeutics, some limitations have been seen in terms of persistence of T-cell effect, duration of response, toxicity, and overall therapeutic index. In addition, differences in therapeutic index, and the level of toxicity between the different CD19 CAR-T and BCMA CAR-T products have highlighted the importance of all aspects of CAR-T design, patient selection, patient dosing, and overall patient care.

Target Selection More Difficult in Solid Tumors

The challenge in extending CD19 CAR-T and BCMA CAR-T success to other cancer targets, in particular, cancer targets beyond blood cancers, has been highlighted by disappointments in early efforts with CAR-T therapeutics in patients with solid tumors.

In particular, we note patient deaths in early experience with CAR-T therapeutics targeting Her2 in non small cell lung cancer (NSCLC), a target thought to be unique to lung cancer, tumor cells.

In considering the difficulty in identifying protein targets on the surface of solid tumor cancer cells, it has been estimated that there have been roughly 300 separate protein targets on solid tumors interrogated, but none has yet to be specific to the cancer cell.

Given the significant cell killing ability of targeted T-cells and the potential for significant toxicity, patient deaths and severe adverse events (SAEs) should not be unexpected when protein targets on cancer cells are also expressed on normal cells.

In approaching this tougher hurdle of targeting cellular immune therapies at more difficult to treat solid tumors, both an exploration of novel targets, as well as optimization of the therapeutic index (balance of efficacy and toxicity) of these cellular immune therapies, are being pursued.

Considering First Generation CAR-T Therapeutics and Importance of the Construct, and Concepts of Conditioning and Dosing

With the focus on optimizing the therapeutic index of existing CAR-T therapeutics, and increasing efficacy extending duration of effect and limiting toxicity, we believe the first generation CAR-T therapeutics are a good starting point in considering approaches that might be used to increase the likelihood of success against protein targets in solid tumors that aren't exclusively expressed on tumor cells.

The importance of optimizing all aspects of CAR-T product design, dosing, and delivery has been highlighted, with significant differences between initial CD19 CAR-T and BCMA CAR-T therapeutics. In particular, within the CD19 CAR-T category, differences in cytokine release syndrome (CRS) and cerebral edema have been seen between leaders Juno and KITE, with Juno having been put on temporary clinical hold for cerebral edema with JCAR015, and having additional cases reported at the American Society of Hematology (ASH) annual despite protocol changes, and with KITE having no issues with cerebral edema for its CD19 CAR-T product xx.

In the BCMA CAR-T category, similar differences in efficacy and toxicity have been observed between products from Bluebird Bio/Celgene, Novartis, and original data from the National Cancer Institute (NCI). In particular, for BLUE/CELG, a 100% complete response (CR) rate is seen at the highest dose tested to date, with 2 patients with absence of minimal residual disease (MRD-), and with an absence of any significant level of cytokine release syndrome (CRS) seen with both NCI experience and recent data from Novartis at the American Society of Hematology (ASH) meeting.

In considering what might drive differences between CAR-T therapeutics targeting the same protein target, various aspects of CAR-T therapy have been highlighted, including the following.

- **The actual CAR-T construct** and what is called the **co-stimulatory domain**, which is a second signaling switch within the signaling machinery of the CAR-T construct inside the cell. Two common co-stimulatory domains have been contrasted with the 41BB domain, thought to cause less rapid expansion of the CAR-T cells after dosing, with lower risk of cytokine release sydrome (CRS) and cerebral edema vs. the CD28 domain thought to cause more rapid expansion of CAR-T cells, and associated with the cerebral edema seen with JUNO's CD19 CAR-T, JCAR015.

- **The conditioning regimen --** The process of dosing patients with CAR-T cells involves a process similar to that with stem cell transplant, where to provide an environment for the CAR-T cells to expand, patients are treated with a conditioning chemotherapy to eliminate cells that would compete for growth factors in the body. Conditioning chemotherapy with a combination of Fludarabine + cyclophosphamide, has been speculated to potentially lead to more rapid expansion of the CAR-T cells than with cyclophosphamide, alone. The theory of fludarabine's contribution to cerebral edema with JUNO's CD19 CAR-T was somewhat questioned at the recent ASH meeting, where cerebral edema still occurred despite removal of Fludarabine from the conditioning regimen.

- **Avoiding "tonic signaling** of CAR-T cells in absence of the protein target" -- At the recent ASH meeting, Bluebird Bio and partner Celgene highlighted significant work on choosing the right CAR-T construct to avoid "tonic signaling," or the activation of the CAR-T cells without interaction with the protein target.

How "Next-Generation" CAR-T Therapeutics Might be Designed to Optimize Effect and Therapeutic Index

- **Designing in signals to CAR-T products for sustained and more potent effect** - In looking at the key properties of CAR-T cells, and T-cells in general, we believe one should consider persistence, expansion, and activity of the cells, that is, how long do they stay around, how large the population of cells can expand to be, and how active are the cells at killing targeted cancer cells. In seeking to optimize CAR-T therapeutics across these domains, companies are looking to design CAR-T cells that release key factors, cytokines, that stimulate the cells. Examples of these factors include interleukin-2 (IL-2), IL-7, IL12, IL-15, and IL-21. In general, IL-12 is a factor that enhances the cell killing ability of T-cells, whereas the other factors, in particular, IL-2 and IL-15, are important for the persistence and expansion of T-cells. JUNO's so-called "ARMORED CAR" is a next-generation CAR-T product candidate that is designed to secrete IL-12 to enhance the cell-killing capability of the CAR-T, whereas Ziopharm Oncology (ZIOP) is looking to evaluated a next-generation CAR-T therapeutic that also produces IL-15 for more sustained effect and greater expansion of the CAR-T cell numbers.

- **Designing in "shut-off" switches to turn off CAR-T cell activation on toxicity** -- Another approach to optimizing the therapeutic index, or balance of efficacy and toxicity, is the design of a so-called "kill-switch," or "shut-off switch" into the CAR-T cell, which can be activated by a separate small molecule drug. Bellicum (BLCM) has designed into its immune cell products a "kill-switch" that is activated by a small-molecule drug called rimiducid, which brings together two proteins inside the CAR-T cell that create a suicide signal. The discovery of the suicide switch, which BLCM is designing into its immune cell products, was discovered by scientists at Ariad Pharmaceuticals (ARIA), and the activators of the suicide switch are called rapalogues, of which rimiducid is one. BLUE and Cellectis (CLLS) are also looking at designing into their CAR-T cell products suicide switches, also triggered by rapalogues.

- **Designing in checkpoint inhibitors to counteract the immuno-suppressive tumor micro-environment** -- One of the central discoveries in the area of immuno-oncology, and the basis for the approval of drugs like Merck's KEYTRUDA and Bristol-Myers Squibb's OPDIVO, is the discovery of checkpoint proteins on the surface of cancer cells, which attach to checkpoint receptors on T-cells, to turn them off. In essence, checkpoint proteins make cancer cells invisible to T-cells, or at a minimum, create a state of exhaustion in the T-cell where it is no longer active against the tumor cell. Checkpoint inhibitors (CPI), like KEYTRUDA and OPDIVO, block this interaction of the checkpoint protein (ligand), called PDL-1, on tumor cells, and the PD1 receptor on T-cells, to essentially unmask the tumor cell to T-cell attack. While CAR-T therapeutics are designed to overwhelm the immuno-suppressive PDL-1 signal from tumor cells, one might surmise that a T-cell that lacked the PD-1 receptor would be immune to this suppressive signal.

The Parker Foundation, through funding of the University of Pennsylvania, is developing a next-generation CAR-T therapeutic where the PD-1 receptor, naturally found on the surface of the T-cell, is engineered off. In principal, any company with gene editing capabilities that can cut out a gene for PD-1 within the T-cell, could also design a next-generation CAR-T therapeutic impervious to PDL-1 suppression from the tumor. Indeed, leading gene editing companies are doing just that, with Bluebird Bio's Mega-Tal gene editing being evaluated in "knocking out" PD-1 preclinically, with Cellectis (CLLS) having demonstrated this pre-clinically, and with published data on a collaborative effort between gene editing leader Sangamo (SGMO) and the National Cancer Institute (NCI) using SGMO's gene editing platform to "knock out" PD-1, as well. Presumably the collaboration between Intellia (INTC) and Novartis (NVS) could, in part, focus INTC's CRISPR CAS9 editing "scissors" on designing out PD-1 on NVS's CAR-T therapeutics. Other gene editing companies operating in this area include Editas, CRISPR, and Caribou.

- **Engineering in capabilities for CAR-T cells to hone more effectively toward tumor cell areas** -- Ultimately, for CAR-T therapeutics and other adoptive cellular therapy (ACT) products to work, the cells themselves need to migrate to the tumor site(s) to attack the cancer cells directly. This honing is assumed to occur naturally and by sheer number of cells infused, but efforts are being undertaken to optimize the "honing" of CAR-T cells, and other ACT products, to tumor cells. FATE therapeutics (FATE) has developed technology to optimize honing of immune cells to specific locations and has an extensive collaboration with JUNO, although details of next-generation CAR-T products with optimized tumor honing have been limited and could potentially gain visibility in 2017.

- **Designing in more than one target for the CAR-T cell to avoid tumor resistance --** the best tumor target, which is highly over-expressed on a tumor, may not be present on every tumor cell. In addition, as a resistance mechanism, tumors expressing a certain protein target may avoid immune attack by CAR-T cells by downregulating that protein target. To address this limitation of first-generation CAR-T therapeutics, efforts are under way to target multiple proteins on cancer cell surfaces. One such effort, as a

next-generation of JUNO's CD19 CAR-T, is a CAR-T therapeutic targeting both CD19 and CD22 proteins on cancerous leukemia and lymphoma cells.

Engineered T-cell Receptor (eTCR) Therapeutics as Alternative to CAR-T Therapeutics

One significant disadvantage of CAR-T therapeutics is that larger protein targets are typically found on normal cells, as well, and while these proteins may be over-expressed and in higher abundance on tumor cells, they are not unique or exclusive to the tumor, itself. In essence, targets of CAR-T therapeutics are tumor associated antigens (TAA), as opposed to what would be classified as tumor rejection antigens (TRAs) that are unique to the tumor. CD19 CAR-T and BCMA CAR-T therapeutics are effective because while the CD19 and BCMA targets are also found on normal blood cells, these blood cells are temporarily dispensable and should grow back quickly when the cancer is eliminated. Normal cells in the rest of the body, and involved in solid tumors are not so dispensable and, as such, a more selective approach is required. One such selective approach is the engineering of T-Cell Receptor or, eTCR therapeutics.

While CAR-T cells are engineered to target larger proteins on the surface of cancer cells, the actual T-cell receptor (TCR) on our normal T-cells is designed to recognize smaller protein fragments, or smaller peptides, which are present on cell surfaces in association with larger proteins called MHC. To advance the quest for engineered T-cells that might target these smaller MHC associated peptides on the surface of cancer cells, engineered T-cell receptor (eTCR) are being developed as an alternative to CAR-T cell therapeutics.

While the CAR construct on T-cells involves a genetically engineered antibody that is fused with the internal signaling machinery of the T-cell, these eTCR therapeutics represent actual engineered T-cell receptors that are designed to target actual smaller peptides on the cancer cell. The advent of eTCR therapeutics has opened up a whole new area of cancer cell targets, as these peptide fragments on the surface of cancer cells actually emanate from the inside of the cancer cell and offer a litany of new potential cancer targets.

While the understanding of first generation CAR-T therapeutics involves some understanding of how a simple antibody interacts with a protein on cancer cells, the actual understanding of eTCR design requires some appreciation of how T-cells in the body typically work and a deeper understanding of how T-cells interact with foreign peptides in the body.

Explanation of a few terms before delving into first generation eTCR Therapeutics:

- **Proteins vs peptides** -- Peptides are short linear sequences of roughly 9-10 amino acids. Amino acids are the building blocks of all peptides and proteins in our body and themselves are chemical structures in a library of 20 blocks. While peptides are short linear sequences, proteins have a much longer sequence of about 400 amino acids, on average, and take on a 3-dimensional shape as this much longer sequence folds on itself. However, if a protein were unraveled, it, too, would have a linear sequence with about 400 building blocks.

- **MHC associated peptides** -- How proteins are expressed as peptide fragments on the surface of tumor cells -- proteins that are either produced in a tumor cell, or taken up by a tumor cell, are broken down within the cell into discreet 8-10 amino acid length peptides and then transported to the surface of the tumor cell for presentation to the immune system in combination or association with the MHC protein. A protein can be cut into many different peptide sequences for presentation on the tumor cell for the immune system to see, and these sequences may be overlapping (e.g., amino acids 10-20, 13-23, 18-28, etc.). As such, if we take one such tumor rejection antigen (TRA), the 180-amino acid NY-ESO-1 protein, it may be presented on the tumor cell as dozens of different NY-ESO-1 peptides in association with the MHC protein.

- **The MHC protein, itself** -- the MHC protein, or major histocompatability complex (MHC) protein, is expressed on every cell in our body and on all tumor cells. MHC protein is either MHC class 1, or MHC class II. Most relevant to eTCR therapeutics is MHC class 1, which presents peptides on the surface of all cells in the body (except red blood cells) and all tumor cells. MHC class II is expressed on specialized immune cells such as dendritic cells.

- **HLA Restriction –** When T-cells interrogate a peptide on the surface of a target cell (as either foreign, or "self"), their T-cell receptor (TCR) interacts with the peptide in association with an HLA protein.

Considering First Generation eTCR Therapeutics

In designing an eTCR therapeutic to target a specific MHC associated peptide, the engineered T-cell receptor has to be designed to recognize the specific peptide sequence, as well as a specific portion of the MHC protein. To understand how eTCR therapeutics are designed, it might be worthwhile to consider the process of their creation.

- Step 1 - Remove lymphocytes (T-cells, B-cells, Natural Killer cells) from a patient's tumor to identify the lymphocytes that are infiltrating the tumor or so-called tumor infiltrating lymphocytes (TILs).
- Step 2 - Run an experiment in which all potential peptide sequences of the cancer protein are represented separately and identify which peptide sequence that the tumor infiltrating lymphocytes (TILs) from the patient's tumor interact with. In a 400-peptide cancer protein, TILs can recognize linear sequences of 9-11 peptides, and these are scanned in sequence, i.e., peptides 1 to 10, 2 to 11, 3 to 12, 4 to 13, etc.
- Step 3 - Genetically engineer eTCR product with the T-cell receptor (TCR) on the surface of the engineered T-cell corresponding to the specific peptide sequence identified in step 2.
- Step 4 - Give the eTCR product to patients over-expressing the tumor protein.

With solid tumors as the "holy grail" of adoptive cellular therapy (ACT) success, early success has been seen in several areas with eTCR products, targeted at novel MHC associated peptides. In particular, some degree of validation has been seen with eTCR therapeutics targeted at NY-ESO-1, in cancers like sarcoma and myeloma, as well as individual patient success targeting mutated KRAS protein.

- AdaptImmune (ADAP) – ADAP is in clinical development with an eTCR therapeutic targeting NY-ESO-1 and has demonstrated a 50% overall response rate (ORR) in patients with NY-ESO-1 over-expressing melanoma.
- KITE (KITE) - KITE had an eTCR program targeting NY-ESO-1, but has since abandoned the program, at least the first generation program.
- National Cancer Institute (NCI) - NCI also has an eTCR program targeting NY-ESO-1.
- Beyond the lead eTCR programs targeting NY-ESO-1, other programs under active development include eTCR programs against MAGE-A3, MAGE-A10, and mutated KRAS protein.

Importance of Target Selection for eTCR Therapeutics With a More Complex Spectrum Of Options

In considering the different categories of peptide targets for eTCR therapeutics, we highlight several categories of MHC associated peptides to target. These include (1) mutated neo-antigens that are either common between patients, or unique to each patient; (2) cancer testis peptides, which are present on normal cells during embryonic development, but which are supposed to disappear in adult life; (3) viral associated peptides; and (4) peptides that represent unique modifications of sugar chains. An example of a mutated neo-antigen is KRAS, whereas an example of a cancer testis antigen is WT-1. A virally associated antigen could come from HPV or EBV infection, and alternate sugar patterns define MUC-1 and MUC-16 targets.

A Simpler Alternative to CAR-T and eTCR

While the design of CAR-T products and eTCR therapeutics is quite elegant, a potentially simpler approach is to remove T-cells from the actual patient's tumor, so-called tumor infiltrating lymphocytes (TILs), optimize the selection of these TILs outside of the body, and then, like CAR-T and eTCR products, give these cells back to the patient in relatively large numbers.

Whereas the elegant design of the NY-ESO-1 eTCR has generated a 50% overall response rate (ORR) in melanoma patients that have tumors over-expressing NY-ESO-1, a simpler TILs approach had the same 50% response rate in a broader melanoma population.

Considering First Generation TILs Therapeutics

Early success with TILs therapy has been seen with Lion Bio (LBIO) in melanoma, as well as with Dr. Rosenberg's group at the National Cancer Institute (NCI).

Putting It All Together - What Might We Learn in 2017

CAR- T Therapeutics

- KITE plans to file for approval of KTE-C19 for diffuse large B-cell lymphoma (DLBCL) following an update based on a 6-month data update. Will the Food and Drug Administration (FDA) accept and approve this as one of the first CAR-T products to be considered for approval?
- JUNO should have data for its alternate CD19 CAR-T product, JCAR017, which has the different 41BB co-stimulatory domain, in DLBCL.

TCR Therapeutics

- ADAP should gain agreement with the FDA on a phase 3 design of its NY-ESO-1 SPEAR in synovial sarcoma.
- ADAP should get additional data from its SPEAR programs targeting MAGE A3 and MAGE A10.
- ADAP should identify targets for next development, which could include an eTCR (targeting AFP in liver cancer.) from the recent BLCM collaboration.
- KITE should enter clinical development with an HPV antigen targeted eTCR.
- JUNO should progress with a WT-1 targeted eTCR in non small-cell lung cancer (NSCLC.)
- ZIOP should provide an update on its eTCR program targeting tumor neo-antigens.
- BLCM should enter clinical development with its eTCR product targeting PRAME in melanoma.

TILs Therapeutics

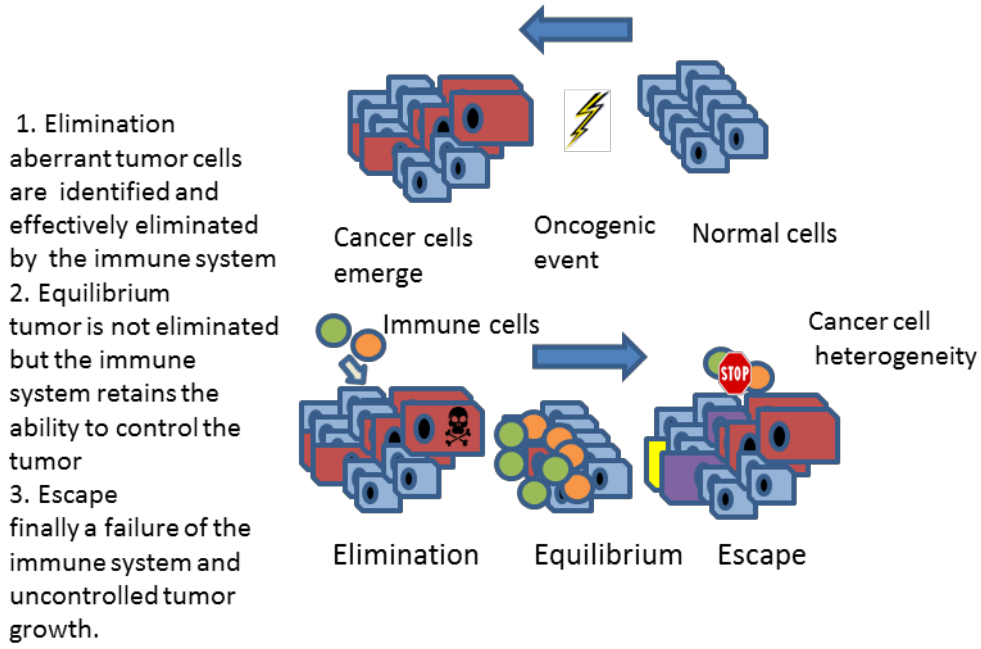
- LBIO should gain visibility on whether NCI data in melanoma can be replicated with a more commercially relevant manufacturing process across a multi-center phase 2 study reporting at ASCO 2017.
- There could potentially be visibility on a next-generation TILs product that engineers out PD-1.
- There could be advancement of an HPV targeted TILs product in 2017.

Adoptive Cellular Therapy Overview

Despite advances in chemotherapy and the introduction of targeted kinase inhibitors and monoclonal antibodies, cancer remains largely refractory to cure and a diagnosis of metastatic cancer still delivers a dismal prognosis. Cancer arises, in part, due to a failure of the body's immune system to identify and eliminate malignant cells**. A key objective of Adoptive cellular Therapy (ACT) is to overwhelm the inhibitory tumor microenvironment, by sheer weight of numbers, with immune cells modified outside the body able to potently target cancer cells.**

It should be noted that cancer emerges after a series of oncogenic (cancer promoting) events that integrate genetic and environmental factors. Following initial establishment of the tumor, the role of the immune system evolves during a process referred to as immuno editing involving elimination of the tumor in stage 1, adaptation to reach an equilibrium in stage 2, and finally, tumor escape in stage 3, as depicted in Exhibit 1.

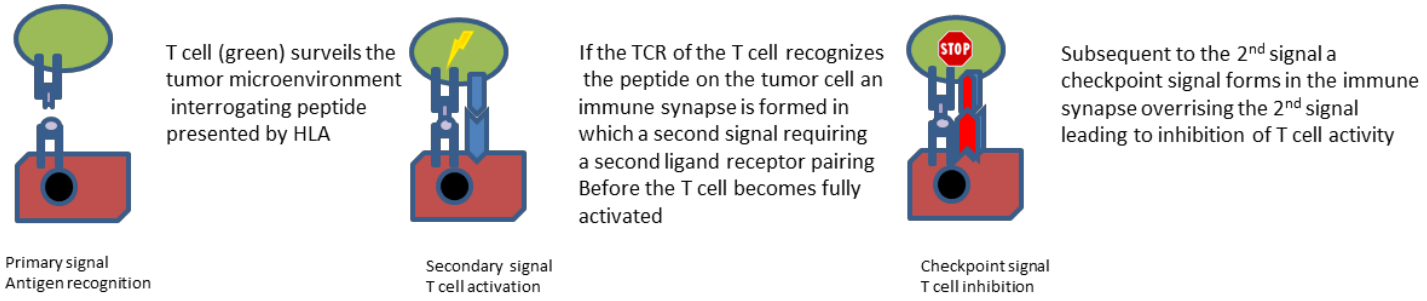
Exhibit 1. Evolution of the Immune Response to Cancer



Source: Wells Fargo Securities, LLC

Recent progress in understanding the interplay between immune cells and cancer cells has begun to uncover some of the ways in which cancer cells hide from and neuter immune cell function. As noted in Exhibit 2, binding of a tumor antigen by the T cell is required, but is insufficient, for T cell activation, which requires a second signal, as depicted in the middle panel in the following exhibit. In a functioning immune system, professional antigen presenting cells (APCs) provide both the primary and secondary signals leading to T cell activation. These activated T cells can then find and kill cells expressing the target antigen, and clonal expansion allows for a logarithmic increase in tumor targeting T cell numbers. In the case of T-cells engineered outside of the body and then re-introduced as engineered T-cell receptor (eTCR) therapeutics, both the primary and secondary signals need to be designed into the actual T-cell therapeutic.

Exhibit 2. T Cell Activation and Inhibition



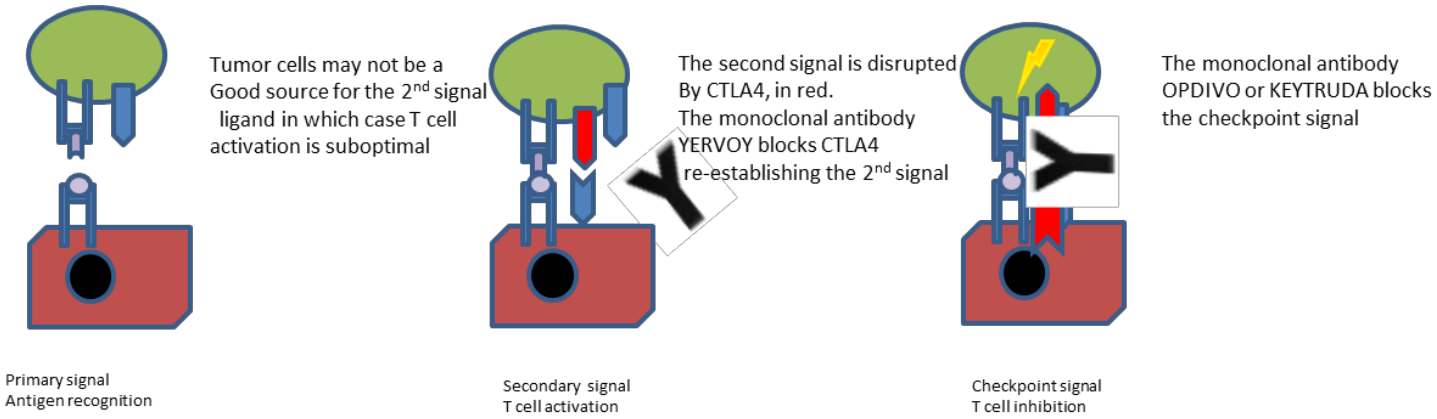
Source: Wells Fargo Securities, LLC, HLA Human leukocyte antigen

To prevent the immune system from running amok and leading to unwanted collateral damage, a series of temporally regulated checkpoint inhibitor pathways act to control T cell function, as depicted in the right-hand panel of Exhibit 2..

As noted in Exhibit 3, tumor cells often lack key molecular components required for T cell and immune cell activation, and, in addition, premature upregulation of CTLA4 on the T cell displaces the CD28 2nd signal, leading to a short circuiting of T cell priming and activation. The first approved checkpoint target antibody, YERVOY, acts to re-establish the 2nd signal by clocking CTLA4. A distinct class of antibodies targeting a checkpoint of activated T cells, the programmed death 1 pathway (PD-1) inhibitors, OPDIVO, KEYTRUDA, TECENTRIQ, and most recently, BAVENCIO, are able to reverse PD-1 mediated T cell dysfunction.

Striking data for monoclonal antibodies that bind to these naturally occurring checkpoint inhibitors of T cell/immune cell function have reignited interest in the role of the immune system as a way to target cancer. While drugs such as YERVOY and KEYTRUDA/OPDIVO have produced unprecedented data in solid tumors such as melanoma and non-small cell lung cancer (NSCLC), the reality is that most patients remain resistant to these agents, and while combination therapy offers an opportunity for improved efficacy, tolerability is often reduced. **One of the potential benefits for ACT is the ability to overcome a variety of resistance mechanisms through infusion of millions to billions of T cells able to target the cancer.**

Exhibit 3. Approved Monoclonal Antibodies can Help Initiate and Maintain an Anti-Tumor Response



Source: Wells Fargo Securities, LLC

Many tumors contain T cells that are able to recognize and kill tumor cells, but their function is neutralized by the tumor microenvironment which subverts immune modulating pathways such as the programmed death 1 (PD-1) pathway. The clinically observed effects of agents such as PD-1 pathway inhibitor KEYTRUDA invoke a thesis that T cells capable of targeting and eliminating the tumor are actually present, and in some patients at least, their moribund state can be reversed by neutralization of a single immune checkpoint, in this case, the PD-1 pathway. Early data suggest that the number of distinct T cell clones able to effectively target the tumor may be as few as 1. Van Rooji and colleagues noted that of the 448 immunologically relevant epitopes that could be identified by a tumor infiltrating lymphocyte (TIL) in a melanoma patient responding to YERVOY, only 2 were able to trigger a patient-specific tumor response, Clin Oncol. 2013;31:439–42., and similarly, Rizvi and colleagues showed that **in a non small-cell lung cancer**

(NSCLC) patient responding to KEYTRUDA,T-cell mediated clinical activity and anti-tumor response was mediated by a single mutation, Science, 2015;348:124–8.

In the absence of tumor-targeting T cells in the tumor microenvironment, antagonists of checkpoint inhibitors are assumed to be ineffective. We find it interesting, therefore, that the tumors where the checkpoint inhibitory agents have shown most promise are those with a high number of DNA mutations in the tumor, referred to as somatic mutations, which provide an opportunity for recognition as foreign by a T cell. In the case of NSCLC, this DNA damage is most often secondary to cigarette smoking and, assuming that at least some of the damaged DNA is translated into protein, it can be presented by APCs to T cells in lymph nodes. If a T cell recognizes the mutated protein as non-self, it can become activated, leading to a cascade of events including rapid expansion of the T cell clone, which can lead to killing of tumor cells displaying that particular target protein.

From a cancer biology perspective, the function of these mutated proteins can be critical. If they are oncogenic, i.e., the mutation is driving the uncontrolled growth of the cancer, such as is the case with the BCR-ABL abnormality in chronic myeloid leukemia, targeting the biochemical pathway associated with the abnormality with therapeutic proteins or small molecules can have a profound benefit. Yet from an ACT perspective, the function of the mutated/abnormal protein is likely less relevant since the ACT T cell is agnostic to a functional or non-functional protein as long as it is recognized as foreign and leads to T cell activation. It is reasonable to assume, however, that if the target of the ACT approach is not critical to the oncogenic phenotype of the cancer, resistance is more likely to occur as clones lacking the target emerge or are selected for.

In addition to mutated proteins, cancer cells often display proteins that are out of context for that tissue, or that are in overabundance compared to the level observed on healthy cells in the tissue. DNA that should be silenced is often not so in cancer, and proteins that played a role in neonatal development, for example, are often re-expressed in cancer cells and can become targets for ACT. These neonatal proteins are referred to as cancer-testis antigens, while antigens associated with development of a specific tissue are often referred to as differentiation antigens.

Cancer biologists have captured a dichotomy of cancer targets with tumor-specific antigens (TSA) comprising immunologic targets that are unique to the cancer and that are mostly mutated or viral proteins seen as non-self by the immune system and tumor-associated antigen (TAA) that are self-proteins, which are either over expressed, or expressed out of context. Targeting a unique TSA would be expected to have a better safety profile than targeting a TAA since the TSA is unique to the cancer, but a TAA may be found on both healthy and cancerous tissue, although in some cases the tissue targeted via a TAA may not be critical such as the prostate, B cell, or ovary.

Another important consideration is whether the cancer is addicted to the pathway that uses the antigen. Emerging data for CD19 targeting ACT, as an example, suggests that failure of ACT is often linked to loss of the target on the cancer cell**, thus targeting a TSA that is also part of a critical pathway to the cancer cell could lead to a lower rate of escape by antigen loss than targeting a non-critical TSA or a TAA. Examples of TSA and TAA that are being targeted in the clinic are shown in Exhibit 4.**

Exhibit 4. Tumor-Specific and Tumor-Associated Antigens

Tumor Specifc Antigen	
Non-viral	EGFR-VIII
	Neo-epitopes
Viral	HPV
Tumor Associated Antigen	
Cancer testis	NY-ESO-1
Lineage	PSA
	CD19
Over expressed	Her2
Mutated	KRAS
Post translational	Mucin

Source: Wells Fargo Securities, LLC

History of Adoptive/Activated Cell Therapy

It has long been observed that immune cell stimulation in cancer patients with cytokines like interleukin 2 or interferon alpha can lead to dramatic anti-tumor responses. It has also been observed that patients with blood cancers receiving immune cells from a donor can be cured. In following up on these observations, investigators have explored the utility of harvesting tumor infiltrating lymphocytes (TIL) directly from tumors in patients, expanding them and reinfusing them by the billion. While this strategy showed some success, it lacked precision and led to strategies to identify relevant TCRs that could be engineered into the T cell prior to expansion and re-infusion. Both TIL and engineered TCR strategies require presentation of antigen by HLA at the surface of the tumor; however, a common mechanism of escape is loss of antigen presentation. An alternate approach was to use synthetic biology to combine aspects of antibody technology with T cell biology to create a chimeric antigen receptor (CAR), which would target antigens that are found on the cell surface without the need for HLA presentation, but use the TCR signaling machinery inside the cell to mimic the signaling from a native TCR.

In the setting of B cell hematologic malignancy for example, T cells have been redirected to target B cells and their pre-cursors on the assumption that removing the B- cell precursors can stem the tide of immature cancerous B cells that cause leukemia and lymphoma. Collectively, these immune cell-based approaches comprise a new field often referred to activated cell therapy (ACT). From a procedural perspective, ACT is arguably an extension of stem cell transplant (SCT), and given complexities of the procedures and attendant safety concerns, ACT remains under the purview of the bone marrow transplant unit, and for the foreseeable future we see ACT remaining beyond the reach of community oncology.

No genetically engineered T cell products have been approved by regulatory authorities in the United States and, despite impressive clinical data for some products, regulatory approval will likely require adequate demonstration of safety and efficacy, and the ability to reliably manufacture the product. Acknowledging that these products are more challenging to manufacture than traditional proteins on the one hand creates barriers to entry, but on the other hand, presents additional challenges for the pioneering companies that are developing commercial manufacturing processes from academic processes that were designed without a view to regulatory approval. NVS recently announced acceptance by the U.S. FDA of its CTL109 CAR-T product biologics license application (BLA) for accelerated review and KITE recently completed the Axi-Cel (CD19 CAR-T) BLA filing for diffuse large B cell lymphoma. Thus, 2017 becomes a landmark year for ACT as the FDA vets the safety and efficacy of the first ACT products.

Adoptive Cellular Therapy and 3-leter acronyms -- SCT, TIL, TCR, and CARs

As noted, we consider adoptive cellular therapy (ACT) an extension of hematopoietic stem cell transplant (HSCT), but involving more sophisticated genetic engineering approaches to maximize safety and efficacy. Stem cell transplant (SCT) requires the use of high-dose chemotherapy with or without radiation therapy as a preparative regimen to kill as much of the cancer as possible; but as a corollary, the preparative regimen also leads to a loss of healthy immature and mature hematopoietic cells. Such a loss would be fatal if not for the add-back of hematopoietic stem cells (HSC) as part of the transplant, which are either collected from the patient ahead of time, or from a donor.

HSC are harvested by apheresis from the patient prior to the preparative regimen in the case of an autologous HSCT (auto-HSCT) or from a donor in the case of an allogeneic transplant (allo-HSCT). In both settings it is hoped that the healthy transplanted cells are able to target any remaining cancer cells. However, in the allo-HSCT setting, the donor cells can also attack the recipient's healthy cells in a graft versus host (GvHD) reaction. Like HSCT, ACT therapy involves obtaining cells from the patient or a donor; however, these cells need to be expanded because either the number of cells harvested is low, or the apheresis product is separated to select for T cells, which are then engineered and expanded. In both HSCT and ACT the patients receive preparative chemotherapy prior to receipt of the HSC/ACT product, and while in the HSCT procedure this step is myeloablative, for ACT it is lymphodepleting. Chemotherapy used in both procedures is associated with morbidity. In both HSCT and ACT the post-transplant period is critical and may require considerable use of healthcare resources such as admission to the intensive care unit. Finally, while there is no cost in purchasing an HSCT graft in the setting of an autologous transplant, there is for an allo-transplant, which, in the case of a cord blood transplant, can cost more than $100,000 in the United States based on the use of two cord blood units; clearly there will likely be a cost associated with a third party such as KITE engineering an autologous graft.

We believe that HSCT provides a framework for adoptive cellular therapy (ACT) as a way to build on the transplant experience, but with more sophisticated approaches to maximize safety and efficacy.

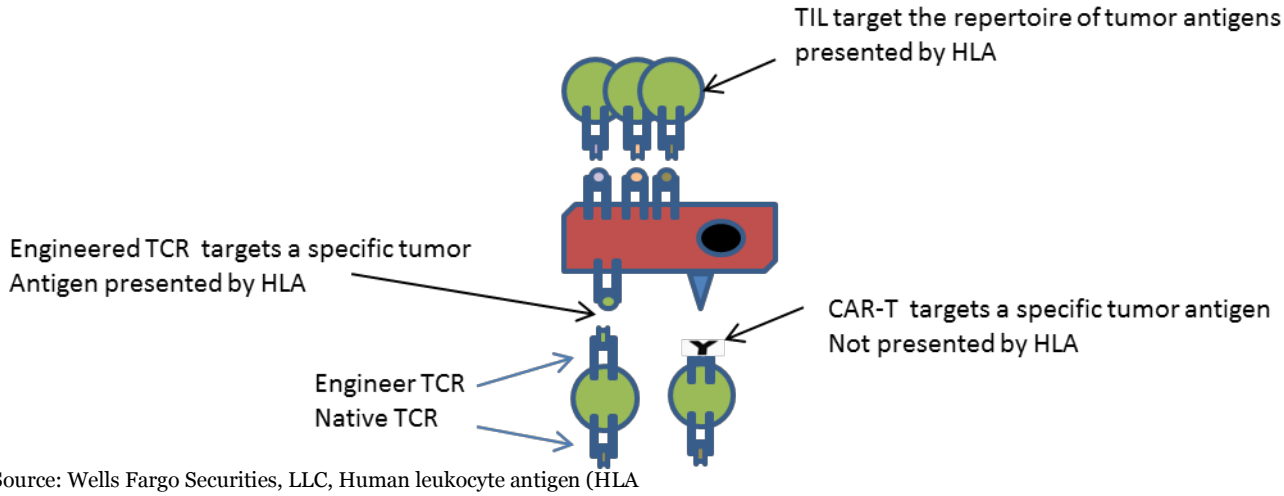
There are three basic types of adoptive cellular therapies (ACT), all of which involve expansion of T cells in the laboratory before re-infusing them into a patient in a fashion similar to HSCT:

(1) Tumor-infiltrating T cells or TILs are removed directly from the patient's tumor and, as noted in the top panel of Exhibit 5, these TIL are polyclonal recognizing several antigens presented by the tumor cell, which may include tumor-associated antigens (TAA) or tumor-specific antigens (TSA).

(2) Engineered T-cell receptor (e TCR) T cells add a new T-cell receptor (TCR) to a T cell to target the desired tumor antigen, and as noted in the bottom left of Exhibit 4, these T cells still carry their naturally occurring TCR.

(3) A chimeric antigen receptor (CAR) is added to a T cell and uses an antibody fragment or protein to target a tumor antigen, as depicted in the bottom right of Exhibit 5. In this case the CAR is a synthetic construct and, as was the case with the eTCR T cells, the CAR-T carries both its TCR and the CAR.

Exhibit 5. Targeting a Cancer Cell with a TIL, Engineered TCR or a CAR-T



Source: Wells Fargo Securities, LLC, Human leukocyte antigen (HLA

Scientists reasoned that if one could identify a TIL that targeted the tumor, and not healthy tissue, one could isolate those T cells from a patient, expand them in the laboratory, and return them to the patient in an attempt to overwhelm the tumor. Further, since the expanded T cells came from the patient where they were not causing damage to healthy tissue, the risk for graft versus host disease (GvHD) would be expected to be very small with such an approach.

In solid tumors, the most obvious place to look for such T cells was the tumor itself and the best disease to try was one with readily accessible tumor that was responsive, albeit rarely, to immune-based therapy – melanoma. Several laboratories have isolated TIL, and while several different tumors have been evaluated, the collective experience is greatest in melanoma where at the National Cancer Institute (NCI) alone in the United States has treated more than 200 patients with TIL therapy.

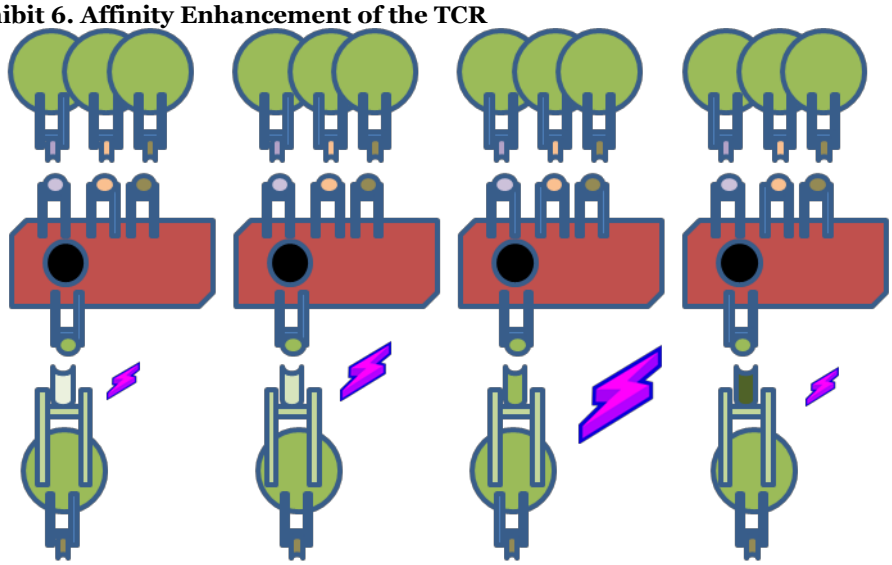
By promoting the non-specific expansion of tumor-derived T cells as part of the TIL manufacturing process, a strength of the TIL approach is that different T cell populations are expanded potentially identifying numerous targets on the tumor. Important to note is that such a polyclonal approach may be relevant because solid tumors are often heterogeneous and by targeting multiple different tumor proteins, the chance that part of the tumor is left untouched because it expresses too little of a particular target to trigger T cell activation is reduced.

While the unbiased approach of TIL is a strength, immunologically it represents a random-chance event since non-tumor targeting T cells, or those able to recognize but not kill a tumor, may potentially dilute the effect of those T cells able to efficiently kill the tumor. A TIL approach assumes that tumor-reactive T cells can be identified and expanded efficiently from the tumor; however, this is not always the case. Recently the NCI published a case study in the setting of triple negative breast cancer where the Assadipour *et al.,* Clinical Cancer Research on line 4/4/17. Assadipour and colleagues were able to identify a TIL that targeted a neoantigen specific for the tumor; however, they noted unexpected failure in producing a highly potent TIL product, likely due to the prolonged manufacturing time required, which led to a decrease in the frequency of the desired TCR.

As noted by Assadipour, sequencing and cloning of the TCR into a T cell may address the challenge to generating a TIL. The ability to identify a neoantigen is a recent advance in the TIL field and TCR-based therapies started in the setting of tumor associated antigens. One example is the Wilm'stumor-1 (WT-1) antigen, which is found to be over-expressed on several tumors including acute myeloid leukemia (AML). By identifying a T cell in a healthy individual that surprisingly was able to potently kill WT-1 expressing AML cells, scientists have previously been able to follow a parallel path to the TIL pioneers – grow up large numbers of these WT-1 positive T cells and give them to patients who were at high risk for relapse following a transplant for a WT-1 positive tumor. Unfortunately, results in the clinic were underwhelming, illustrating a truism – the human immune system does not allow the development of highly potent T cells that target self-proteins.

In contrast to self-proteins, the specific activity of T cells targeting viruses, for example, can be very high and so scientists reasoned that the part of a T cell that recognizes a protein as self or non-self, the **T cell receptor (TCR) could be engineered to increase its potency,** a process referred to as **affinity enhancement,** as depicted in Exhibit 6.

Exhibit 6. Affinity Enhancement of the TCR



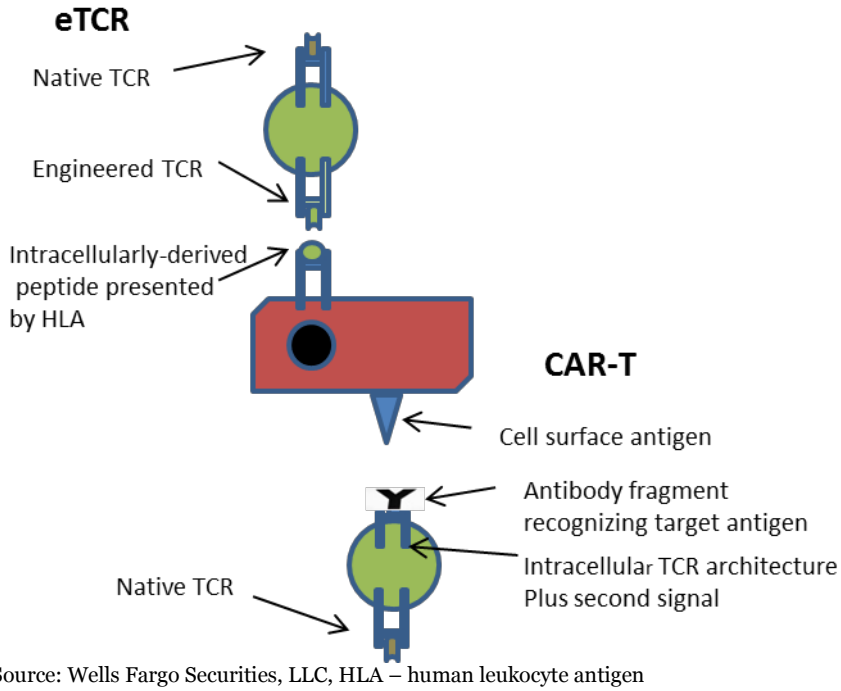
Source: Wells Fargo Securities, LLC

As a result, several affinity-enhanced engineered TCRs (eTCR) targeting a handful of different tumor-associated antigens (TAAs) have been evaluated in the clinic. Central to both eTCR and TIL approaches is the basic concept that the target cell must be expressing the target antigen. In the case of TIL therapy, this target antigen must be recognized by the TCR that is already present on the T cell, while in the case of eTCR, by the TCR genetically engineered onto the T cell.

One common escape mechanism for cancer to render TILs or eTCR as a therapeutic option ineffective is downregulation of the machinery required to express peptides at the cancer cell surface. In doing so, cancer cells prevent interrogation of target antigen by the TCR or the surveilling TIL, or eTCR T cell. On this point, Ribas and colleagues published mechanisms of resistance to KEYTRUDA monotherapy observed in their clinical experience, NEJM 2016 Jul 13. Of 78 patients treated, 42 achieved an objective response, of whom 15 went on to relapse. Of these 15 patients that relapsed, 3 met criteria for interrogation of the resistance mechanisms in which two patients were mediated by mutations in beta-2-microglobulin (B2M) in one and the JAK pathway in another. These translate to an absence of antigen presentation associated with loss of B2M and interruption of IFNγ signaling in two. A CAR strategy does not rely on B2M, but IFNγ signaling is important for T cell mediated killing.

An alternative ACT approach obviates the need for the TCR recognition of an antigen and replaces the extracellular TCR domain with an antibody fragment to create a chimeric antigen receptor (CAR), which can be introduced into a T cell to create a CAR-T. With the ability to leverage 30 years of experience developing antibodies, combined with the exceptional quality of data from CAR-T therapies targeting B cells in B cell malignancies, CAR-T-based therapy has arguably become the most active area in ACT. The basic schematic of the eTCR and CAR-T approaches is shown in Exhibit 7. While not shown explicitly, activation of an eTCR requires both the primary signal through the TCR and a secondary signal, which is provided between the T cell and either the rumor cell, or an APC. In the setting of a CAR, this second signal is appended to the CD3 component of the TCR that is used as part of the CAR architecture.

Exhibit 7. Basic Schematic of Engineered TCR and CAR-T Products



Source: Wells Fargo Securities, LLC, HLA – human leukocyte antigen

TILs – Considering the Process and Is there utility outside of melanoma?

Tumor-infiltrating lymphocyte, or TIL, therapy represents the simplest approach to ACT and has been most widely explored in melanoma.

TIL Process - In short, **for a TIL regimen, specimens of tumor are disrupted and cultured using reagents that support T cell growth. Over a period of weeks, billions of T cells are generated, which can then be re-infused into the patient.** Initial TIL studies were disappointing as the re-infused cells quickly disappeared; but borrowing a leaf from the hematologic transplant community, later TIL trials incorporated the use, collectively lymphoid cells, creating a niche for the incoming cells and removing competition for homeostatic cytokines including interleukin 15 (IL-15**). Incorporating lymphodepletion prior to administration of TILs has demonstrated encouraging clinical activity in melanoma, and preliminary data in human papilloma virus (HPV) associated cancer is also encouraging.** In theory, tumors that are sensitive to checkpoint inhibition could be sensitive to TIL therapy and combinations of TIL with checkpoint inhibitors could extend the utility of TIL. Protocols incorporating newer manufacturing methods for TIL, and the addition of checkpoint inhibitors may expand the utility of TIL beyond melanoma and virally associated cancer.

Hematologic malignancies can also be highly antigenic, and while some tumors are associated with nodal involvement and provide a source for TILs, the most obvious place to look for T cells that recognize tumor is involved bone marrow. Marrow-infiltrating T cells, MILs have been identified, isolated, expanded in a similar manner to TIL, and evaluated in myeloma with encouraging results.

Engineering the T Cell Receptor – Hampered by Technological Limitations?

Each of the approximately 1 billion unique T cells in a human can, in theory, recognize a single antigen because it only displays multiple copies of the same T cell receptor. Rather than trying to find the one T cell in 1 billion that binds the desired cancer cell target, scientists have developed the ability to add a second engineered TCR to a patient's T cells, re-directing them to the desired target. Many researchers have actually selected host T cells that recognize viral antigens for retargeting with an eTCR on the basis that these engineered T cells can both target the tumor with the eTCR and protect against re-emergence of viral pathogens via their native TCR. The new eTCR can be engineered and affinity matured to create a more potent TCR than would be possible using the originally isolated unmodified TCR.

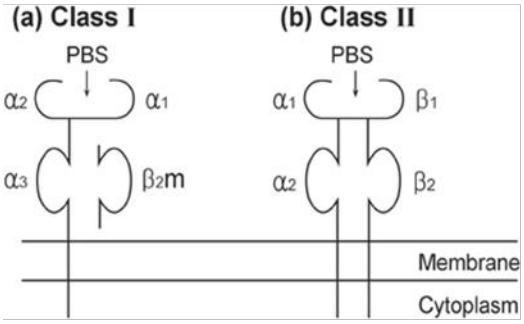
Once the TCR has been designed, nucleic acid coding for the alpha and beta chains of the eTCR can be packaged into a virus that is used to infect donor T cells. Integration of the nucleic acid into donor T cell leads to expression of the eTCR along with the native TCR. One of the theoretical toxicities associated with such an eTCR approach is for alpha beta chain mispairing between the native and engineered TCRs leading to a new TCR that by chance recognizes an antigen on a healthy tissue. However, use of murine (mouse) constant regions for eTCRs has been employed to prevent mispairing since the murine elements will not pair with endogenous human TCR chains. In addition, the use of powerful promoters for engineered human TCR genes ensures that the majority of TCR's expressed by the T cell are the engineered ones and any mispaired TCRs would be in the minority. One reason that naturally occurring TCR's have low affinity for their target is that in theory, each TCR sequence can recognize several distinct human leukocyte antigen (HLA) peptide complexes in the human, and while most of these may never occur in nature, altering the sequence of naturally occurring TCRs to increase avidity presents challenges to identifying potentially unwanted cross-reactivity. This was tragically demonstrated in two early eTCRs tested in the clinic where unwanted cross-reactivity led to fatalities; avidity incorporates both the affinity of the TCR to bind to its target and the durability of that binding – the "on" time.

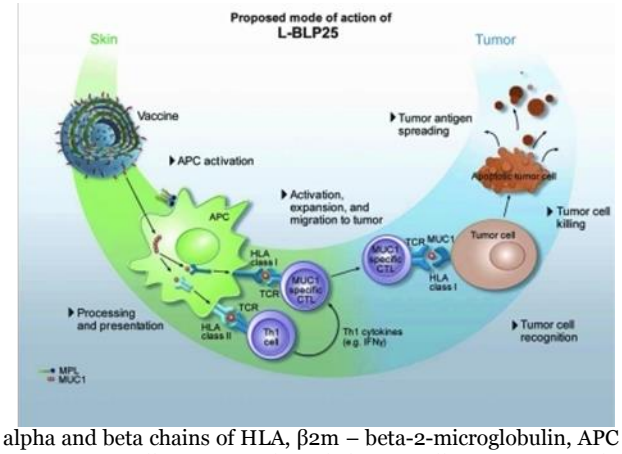
More recently, isolation of neoantigens that are both tumor and patient specific has led to the ability to engineer a unique patient-specific eTCR. In the laboratory, mouse tumors can be interrogated for neoantigens, **those neoantigen peptides synthesized and tested for reactivity against mouse tumors within 24 hours to form the basis of a vaccine or alternatively, inform the development of eTCRs targeting the same neoantigens. Using viral methods for T cell transduction, it is estimated that it would take at least 8 weeks to manufacture a patient-specific neo-antigen T cell product; ZIOP aims to reduce this to 1-2 days using its non-viral Sleeping Beauty T cell transfection technology.**

Human Leucocyte Antigen (HLA) Matching – limits the utility of eTCR approaches

The TCR on a T cell interrogates a peptide that represents a small part of the full-length parent protein. The protein is processed into peptides by the immunoproteasome and each resultant peptide can be presented in a special grove of the human leukocyte antigen (HLA) protein. The TCR recognizes elements of both the peptide and the HLA. HLA proteins are expressed by all nucleated cells, with the exception of germline tissue, where HLA expression is silenced. The HLA gene family codes for a group of related proteins that are key to distinguishing self from non-self. In humans there are more than 200 genes that code for Class I, II, and III HLA proteins. Within Class I there are three major proteins: HLA-A, HLA-B, and HLA-C, whose role is to present peptides at the cell surface, derived from intracellular proteins, to CD8 T cells. In terms of HLA class II, there are 6 main genes potentially coding 10,000 genes and of these, the most important, with respect to clinical utility, is HLA-DRB1. Unlike Class I molecules, Class II molecules are found on dendritic cells, macrophages, and B cells, which represent collectively antigen presenting cells (APC) that present peptides derived from extracellular proteins to CD4 T cells. As noted in the left panel of the figure below, HLA Class I expression is dependent on co-expression of a separate protein, beta-2-microglobulin, which does not associate with HLA Class II. In the right panel the concept of a cancer vaccine is represented, with the vaccine particles bearing tumor antigens being taken up by an antigen-presenting cell (APC) where peptides are processed and presented at the cell surface via the HLA molecules to CD4+ve helper and CD8+ve cytotoxic T cells (CTL).

Exhibit 8. Structures of HLA Class I and II, and Role in Tumor Cell Targeting





Source: Gov, PBS – peptide binding site, α1,2,3 & β1,2 – alpha and beta chains of HLA, β2m – beta-2-microglobulin, APC – antigen presenting cell, HLA – human leukocyte antigen, TCR – T cell receptor, Th1 – helper T cell, CTL – cytotoxic T lymphocyte, MHC o major histocompatibility complex – synonym for HLA

Each HLA gene, such as HLA-A, has several closely related alleles, denoted by additional characters following an asterisk – HLA-A*02 represents a grouping with additional subtypes, HLA-A*020 and HLA-A*0202, for example. Because the TCR interrogates a peptide and an HLA, TCR-based therapy is HLA restricted – thus, while the target protein may be shared with all patients, the TCR will only bind the peptide in the subset of patients sharing the same HLA. Thus, if the original TCR was isolated from an HLA-A*02 individual, engineered T cells using the native or affinity matured version of this TCR can only be used in patients with HLA-A*02. **To date, most eTCRs evaluated in the clinic have been engineered from individuals with HLA-A*2 as approximately 60% of Caucasians share HLA-A*2.**

Loss of HLA is a key tumor escape pathway

T cell mediated tumor cell killing requires T cell priming by HLA presentation of a recognized peptide to the TCR on the T cell and a secondary activation signal, which together form an immune synapse. Absence of the second signal leads to T cell anergy, wherein the T cell remains alive for an extended period of time, but in a hyporesponsive state. The machinery required to present peptides in the context of HLA is complex and the multistep processing required presents tumor cells with **multiple opportunities to lose antigen presentation.**

- Genetic loss of HLA component expression may prevent functional HLA protein from assembling in the endoplasmic reticulum of the cell.
- Immunoproteasomal dysregulation may lead to a lack of peptide to display.
- Transporter dysfunction may prevent peptide transport into the endoplasmic reticulum from the immunoproteasome.
- B2 microglobulin loss prevents formation of a functional class I HLA.
- Oncogene expression, e.g. *c-myc* may lead to inhibition of HLA-DR and loss of HLA class II expression.

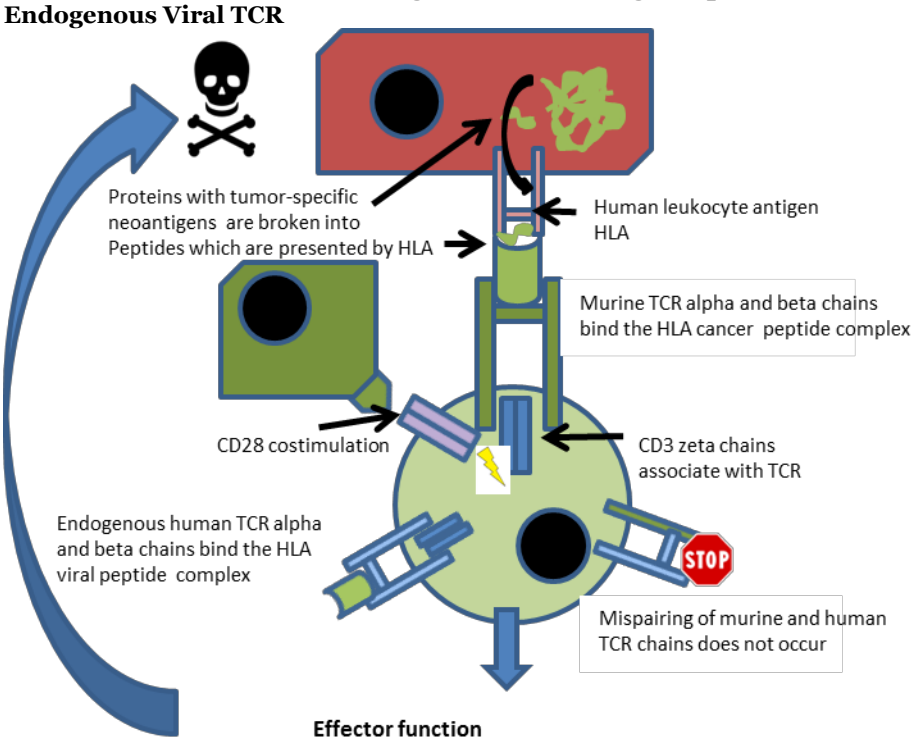
Immunologists refer to each of these as resulting from soft or hard lesions, reflecting different mechanisms. Soft lesions refer to epigenetic dysregulation and present the opportunity for re-induction with the use of hypomethylating or histone deactylating (HDAC)agents, for example. Hard lesions are structural genetic changes that can only be rectified by genetic engineering and thus, present a more intractable problem. Optimal T cell activation requires both Class I HLA function targeting CD8 T cells and Class II HLA targeting CD4 T helper cells and thus, loss of HLA class II alone is sufficient to blunt an immune response. As previously noted, there are several checkpoint inhibitors that can downregulate T cell activation; but in the context of HLA class II, expression of lymphocyte activation gene 3 (LAG-3) on the T cell can outcompete CD4 for binding to Class II HLA given its higher affinity. LAG-3 expression by T cells is a marker of T cell exhaustion.

In a recent review by Garrido *et al.,* Current Opinion in Immunology V39, 44-51 2016, the authors note that 65-90% of tumors have lost Class I HLA expression, of which hard loss is estimated to occur in 30-40%. In addition, Garrido notes that Class I HLA loss increases as the cancer progresses by virtue of T cell mediated

immune pressure selecting tumor sub-clones that are Class I HLA negative. Given the role of B cells as antigen presenting cells, Class II HLA and costimulatory molecule expression could make B cell malignancies an obvious target for T cell-based therapy. However, loss of HLA-DR has been noted in 12-36% of diffuse large B-cell lymphoma (DLBCL), while soft lesions have been noted in 68% of follicular lymphoma (FL) and 15% in Hodgkin lymphoma, a tumor that has been shown to be sensitive to PD-1 inhibition. Expression of *c-myc* occurs in the most difficult to treat lymphomas and results a priori in loss of Class II HLA expression.

Conceptually, eTCR-based ACT can take two basic approaches – targeting a unique patient-specific antigen, in which case each eTCR can only be used in one individual, and secondly targeting a shared tumor antigen, which allows for use of the same eTCR across a broader group of patients who share the same HLA type and subtype. In the latter case, each patient receives an autologous product made from their own T cells. The need to match HLA dictates that for each target, several separate eTCR products will likely be required to treat the broadest population. Most tumors express Class I HLA and not Class II, and therefore, the majority of eTCR products for solid tumors target Class I HLA, although there are several Class II eTCR products in the clinic for solid tumors. B cells express Class II HLA and thus, B cell tumors may be amenable to an eTCR approach targeting Class II HLA.

Exhibit 9. Schematic of an Engineered Neoantigen Specific TCR Using a T cell with an Endogenous Viral TCR



Source: Wells Fargo Securities, LLC, HLA – human leukocyte antigen

In this simplified schema, the green T cell interrogates and recognizes a tumor cell expressing a unique tumor antigen. Since this antigen is unique to the tumor, it is not going to be expressed by a dendritic cell, and tumor cells do not often provide co-stimulation. Successful activation requires the presence of an accessory cell or co-stimulation from the endogenously activated TCR.

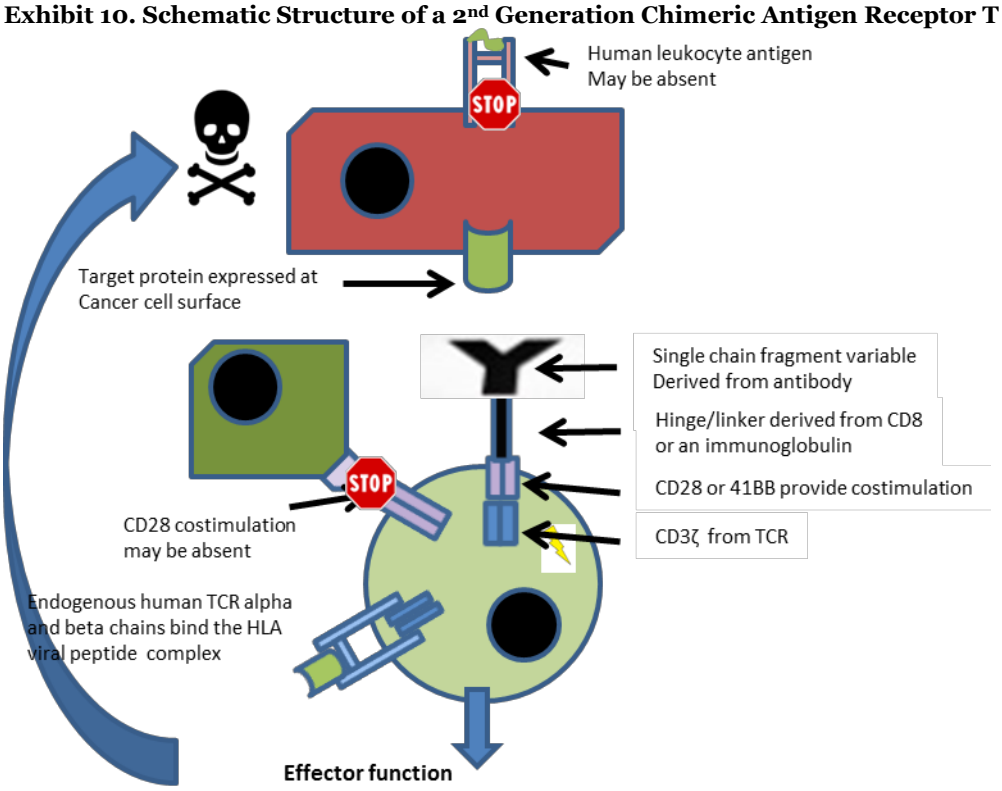
Chimeric Antigen Receptors – broader applicability than eTCRs

Chimeric antigen receptors (CAR) comprise elements of an antibody or receptor to bind the target antigen, grafted on to the modified internal structure of the TCR complex. Specifically in a classic CAR the single chain fragment variable (scFv) region of an antibody, which binds the antigen, replaces the extracellular domain of the TCR and is linked to the transmembrane domain of the TCR complex via a hinge to provide spatial separation.

TCR signaling requires a co-receptor, CD3 which comprises gamma, delta, epsilon, and zeta chains, but necessary and sufficient for signaling is the CD3 zeta (CD3ζ) chain. Data for first generation CARs comprising an scFv with a CD3ζ co-receptor were unimpressive as full activation of the T cell requires both a primary – antigen binding signal and a secondary signal classically comprising CD80/86 on an antigen presenting cell binding to CD28 on the T cell. However, the tumor cell may be a poor source of CD80/86 and the critical second signal is suboptimal.

Integrating this second signal into the CAR architecture obviated the need for ligand activation and, as shown elegantly by scientists at Memorial Sloan Kettering (MSKCC), these 2nd generation CARs were more effective than 1st generation CARs. Scientists from MSKCC compared a CAR with an integrated CD28 co-signaling domain to a 1st generation CAR with separate CD28 co-stimulation domain, which relies on the provision of external CD28 activating ligands, CD80/86. MSKCC showed that the 1st generation CAR plus CD28 was inferior to the 2nd generation CAR due to interference from cytotoxic T-lymphocyte-associated protein 4 (CTLA4), which binds preferentially over CD80/86 to CD28, leading to a dominant inhibitory signal to the T cell, Condomines *et al.,* PLoS One, 2015 Jun 25;10(6). Along with CD28, CD137 or 41BB is also used as a common co-stimulatory signal in 2nd generation CAR-T cells. Cartoons of the TCR along with a CAR are shown in Exhibit 10, which also notes examples of where a CAR would be effective where a TCR would not.

Exhibit 10. Schematic Structure of a 2nd Generation Chimeric Antigen Receptor T cell



Source: Wells Fargo Securities, LLC

Using a CD19 CAR-T in acute lymphocytic leukemia (ALL) as a model, CD28 CARs appear to be more potent, as evidenced by a more rapid increase in CAR-T number, but with shorter persistence than a 41BB CAR. Third generation CAR constructs combining CD28 with 41BB do not appear to show additive benefit, although academics researchers in Germany have suggested that combining CD28 and OX40 is superior to CD28 alone, and **Baylor and the NCI are collaborating on a 3rd generation CD28/OX40 CAR targeting the ganglioside antigen GD2 in neuroblastoma.** Providing a third co-stimulatory pathway separate from the CAR has also been investigated with MSKCC researchers, suggesting

that providing 41BB ligand separately from the CAR may allow for additivity between CD28 and 41BB signaling by ligating 41BB on the same cell, but perhaps more important, on neighboring T cells.

Beyond co-simulation, 3rd-5th generation CAR-T products are evaluating the inclusion of suicide switches for rapid removal of CAR-T in the event of toxicity, provision of cytokines, activators, and chemokines that improve T cell performance, incorporation of transcription factors to optimize T cell function and reduce sensitivity to apoptotic signals, expression of metabolic factors to improve function in oxygen and nutrient-poor environments, and deletion of receptors/pathway coding for checkpoint pathways. Orchestrating the ability to turn these various pathways on and off will likely require the incorporation of switches and rheostats, allowing for precise and individual control of these engineered elements, as well as use of Boolean logic to increase the therapeutic window – that is to say that CAR activation may require ligand A AND B or A NOT B, or even A OR B, BUT NOT C to be present.

While CAR-T most commonly uses antibodies to target tumor antigens, non-antibody-based structures can be used to target relevant cancer antigens including soluble receptors such as the IL-13Rα CAR-T product under development at Mustang Therapeutics or, as recently reported by the Wistar Institute, the follicular stimulating hormone receptor, Perales-Puchalt *et al.,* Clinical Cancer Research published online July 2016.

One challenge facing those developing the next generation of ACT products is the ability to rationally evaluate the many different options/devices for improving T cell function, particularly in the setting of solid tumors. At Seattle Children's, JUNO founder Mike Jensen described an in-vivo competitive screen at the recent 2017 AACR. The Jensen lab uses a truncated EGFR as a tag used to purify CD19 containing CAR-T products and has extended this concept to use a truncated HER2 tag for each test device. Thus, EGFRt/HER2t, double +ve CD4 and CD8 T cells can be purified, with each containing a CAR and a device; recall Jensen uses a 1:1 CD4/CD8 product. Experimentally then each of 4 flanks can be seeded with a different solid tumor engineered to express CD19 and mice treated with a mixture of CD4/8 products containing a range of devices and a CD19 CAR-T. Following days 7 and 14, for example, the tumors can be removed and their T cell content analyzed. In such an experiment not only can the change in differentiation status of the CAR-T be evaluated, but changes can be correlated to the relative abundance of each device, with Jensen noting that different devices may be important not only for different tumors, but at different times following CAR-T administration. This implies the ability to control devices by the incorporation of small molecule controlled switches, which Jensen stated are being incorporated.

With Increased Potency comes Increased Risk for Toxicity

Based on current clinical data, the three modes of ACT reflect a spectrum of toxicity ranging from relatively benign for TIL to potentially fatal for eTCR and CARs. **With most ACT clinical experience on CD19 CAR-T, our view of ACT toxicity is biased by that clinical experience.** Following administration ACT, it is hoped that the cells find their target antigens leading to activation and clonal expansion of the ACT, resulting in rapid destruction of tumor cells. However, despite the common theme of T cell-mediated cell-killing, TIL, CAR-T and eTCR have shown unique and different safety profiles.

TIL trials have been shown to be relatively safe, although some graft versus host disease (GvHD) has been reported, likely reflecting on-target toxicity resulting from T cells derived from the tumor-recognizing tumor-associated antigens such as MAGE, which can also be found on healthy tissue. Such toxicity likely occurs in the context of widespread immune activation following TIL administration.

For the eTCR and CAR products, rapid destruction of tumor can lead to acute inflammation, resulting in a clinical syndrome referred to as cytokine release syndrome (CRS) or, at its worst, macrophage activation syndrome (MAS), which is potentially fatal. Neurotoxicity has been observed with the CD-19 CAR-T trials and has been particularly problematic in NHL and for one CAR-T product in adult ALL, where several fatalities have been reported. Recently, JUNO's JCAR-015 was put on clinical hold following cases of fatal cerebral edema; however, other CD19 CAR-T programs, including JUNO's other CD19 CAR-T programs, have not reported fatal cerebral edema. The FDA has identified left ventricular dysfunction as a CAR-T associated toxicity, but little has been published on this event to date. Immunogenicity has been reported to occur in some patients, leading to immune-mediated clearance of the CAR-T and loss of activity; however, the introduction of humanized and fully human scFv's for CAR-T should reduce this phenomenon. On-target off-tumor toxicity occurs when the transfected T cells target healthy tissue that expresses the same antigen. In the CD19, field loss of B cells is an acceptable trade-off for targeting B cell malignancies. Similarly targeting the prostate, for example, may be an acceptable consequence of targeting a prostate cancer antigen shared with the healthy prostate tissue. In other instances on-target off-tumor toxicity is unacceptable and has been observed with several solid tumor CAR-T products: HER2 targeting lung tissue, CAX9, renal tissue, and CEA the GI tract. Similarly eTCR products have been associated with fatalities due to on-target off-tissue

toxicity involving the heart and brain. Finally U Penn has described a patient relapsing with a CD19 CAR-T +ve, CD19+ve ALL, which arose during product manufacture. This case is highly instructive and debunks the theory that CD19 CAR-T manufacturing would be self-sterilizing since the T cells would be expected to kill CD19+ve B cells. Of course, this risk only applies to the autologous situation as in the allogeneic setting, T cells are derived from healthy volunteers.

Exhibit 11. Summary of ACT-Related Toxicity

Event	Commentary
Cytokine release syndrome	Well known phenomenon associated with massive tumor lysis Correlates with peak CAR-T levels at days 7-10 post infusion Can be fatal Can be blunted with IL-6 inhibition Most problematic with ALL
Neurotoxicity	Most commonly seen with CD19 CAR-T Unknown etiology Correlates with peak CAR-T levels at days 7-10 Can be fatal Most problematic with NHL No effective prophylaxis or treatment
Cerebral edema	Fatal cases limited to one of JUNO's CD19 CAR-T products, JCAR-015 Occurs 1-2 days after CAR-T infusion Was initially though to be associated with fludarabine
Left Ventricular Dysfunction	Identified by FDA as a CAR-T toxicity Little known about incidence, etiology severity or treatment
Immunogenicity	Associated with use of murine scFv Can lead to immune response by patient Can result in loss of CAR-T from blood Can prevent successful retreatment
On-Target Off-Tissue	If target of CAR/TCR is not unique to the cancer Can be fatal
Off-target Off-Tissue	Cross reactivity of TCR with non-target antigen can be fatal
Transduced Cancer Cell Contaminant	Observed in CD19 CAR-T ALL Caused at least 1 relapse Frequency unknown

Source: Wells Fargo Securities, LLC

Cytokine release syndrome, neurotoxicity and left ventricular dysfunction

Cytokine release syndrome (CRS) has long been recognized as a common and potentially fatal side effect associated with release of acute phase reactants such as IL-6. CRS is often accompanied with very high temperature and low blood pressure, and in severe cases requires admission to the intensive care unit. CAR-T and in particular, CD19 CAR-T products developed by different sponsors, have shown a similar CRS toxicity profile. Specifically, severe cytokine release syndrome (sCRS) and severe neurotoxicity (sNT) have been problematic for the CD19 CAR-T products, and fatalities have been reported secondary to sCRS and sNT. Rates of sCRS in the CD19 CAR-T trials are commonly reported at rates of 20% or higher and have led to institution of management algorithms using the anti-IL-6 monoclonal ACTEMRA with or without steroids to blunt CRS. Recently the U Penn group published their analysis of CRS, concluding that a 3 cytokine signature can accurately predict the development of sCRS, Teachey *et al,*. Cancer Discov. 2016 Jun;6(6):664-79. While CRS appears to be related to antigen load, in the case of CD19 on both healthy and malignant B cells, the etiology of severe neurotoxicity is unclear. U Penn recently listed a clinical trial evaluating early intervention with ACTEMRA in the setting of high ALL tumor burden, defined as 40% or more blasts in the bone marrow prior to CART19 infusion, NCT02906371. Definitions used to define sCRS vary and at a recently AACR presentation, SCRI's Jensen noted that using the U Penn definition of sCRS would result in a 0% rate noted for the PLAT-02 JCAR017 trial. Similarly, JUNO has commented that levels of sCRS and sNT are lower with JCAR017 in the setting of DLBCL than reported by others. Using the SCRI criteria, Jensen and colleagues have reported 5 cases of sCRS, 3 of sNT and 4 of sCRS with sNT in 46 pediatric ALL patients treated in the PLAT-02 trial. While most of the CRS data come from CD19 CAR-T in the setting of B cell malignancy, NCI has reported similar levels of CRS with its CD22 CAR-T in the setting of B cell malignancy.

Neurotoxicity has also been observed for the CD19:CD3 bispecific T cell engager, BLINCYTO suggesting that neurotoxicity is not simply due to the CAR-T, but is secondary to CD19. Most of the data on CAR-T safety has come from acute lymphocytic leukemia (ALL); but in non-Hodgkin lymphoma (NHL), rates of sCRS appear lower, but rates of sNT remain problematic. The etiology of sNT remains unclear, but Jensen and colleagues at SCRI have established a primate model for sNT, noting that the alpha4 beta1 integrin may play a role. Interestingly, NCI has not reported sNT as a toxicity associated with CD19, further suggesting that CD19 may be important.

Note that with each successive update from the AxiCel ZUMA-1 trial, KITE reports lower rates of sCRS and sNT, suggesting that as sites and physicians gain experience, these events can be more successfully managed. In part this is due to more aggressive use of ACTEMRA and steroids as reports by several investigators support KITE's observations from ZUMA-1 that aggressive use of ACTEMRA can blunt the severity of CRS and that neither ACTEMRA, nor steroid use diminishes CD19 CAR-T product efficacy.

A recent update from NCI's pediatric CAR-T experience identified left ventricular systolic dysfunction as a new grade 3 event of concern related or possibly related to CD19 CAR-T therapy, as noted in Exhibit 12. SCRI has also reported left ventricular dysfunction as a toxicity in its pediatric ALL trials of JCAR107.

Exhibit 12. Non-Neurologic Grade 3 or 4 Toxicity Possibly Related to CD19 CAR-T

Event	Grade	N(%)
Fever	3	18 (38%)
Febrile neutropenia	3	11 (23%)
Cytokine release syndrome	3	5 (11%)
	4	2 (4%)
Hypotension	3	4 (9%)
	4	2 (4%)
LV systolic dysfunction	3	4 (9%)

Source: NCI

As noted by the FDA at the 2016 American Society of Gene and Cell Therapy, with 105 INDs for engineered T cells in the United States being monitored by more than 10 medical reviewers, the FDA is working with sponsors to allow sharing of safety data within the FDA. This, in turn, would allow the agency to more effectively and comprehensively characterize the safety profile of engineered T cell products. With a focus on CD19 CAR-T products in the setting of refractory disease, the FDA notes that the products have:

- Potential to be curative
- Complex in-vivo activity
- Substantial and complex safety concerns
- Complex manufacturing processes that relate to safety issues
- Registration studies ongoing

Because the trials, including registration trials, are small by agency standards, the FDA believes that the ability to compare across products in a centralized fashion will allow the FDA to inform sponsors of safety issues early in product development. For CD19 CAR-T products the agency has flagged:

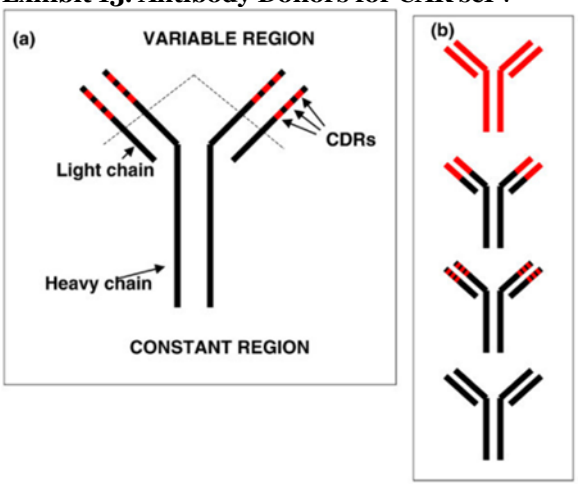
- Cytokine release syndrome(CRS)
- Neurological complication
- Intracranial hemorrhage
- Potential for second malignancy

The FDA has approached and received positive feedback from 3 commercial sponsors of CD19 CAR-T products.

CAR-T immunogenicity

Another aspect of safety relates to product immunogenicity, which may result in reduced efficacy if the CAR is recognized by the recipient's immune system and cleared. Immunogenicity likely limits the ability for repeat CAR-T dosing. For the most part, ACT/CAR therapy is practiced as a single dose administered over 1 or 2 infusions. Nearly all CARs that have entered the clinic have used antibody-derived single chain fragment variable (scFv) region constructs that contain murine sequences, which would be expected to be immunogenic; antibodies have traditionally been made in mice, although in state of the art antibody manufacture, this is not the case. As shown in Exhibit 13, the murine (mouse derived) content of the antibody from which the scFv of the CAR is derived can vary with a fully human antibody as the donor likely to be less immunogenic than a murine/chimeric antibody.

Exhibit 13. Antibody Donors for CAR scFv



Source: cancer.gov, red indicates murine sequences and black human sequences

However, lymphodepletion prior to CAR-T dosing has the effect of delaying the ability of the host to mount an anti-CAR immune response until the host's immune system recovers. **Some investigators have reported that anti-CAR responses have developed and do correlate with reduced efficacy, while others have not correlated an anti-CAR response to a reduction in efficacy.** Although the importance of anti-CAR responses remains an open question, CAR-T therapies are designed to kick start an immune response and any non-self-antigens are likely to become targets for host immune responses.

While the majority of CAR-T products have used DNA vectors to allow for durable and long-term CAR expression, concerns over the potential for off-target toxicity have led some to evaluate RNA-based CAR vectors. RNA vectors allow only temporary expression of the CAR, a construct U Penn describes as "biodegradable," with CAR expression lasting a few days.

U Penn's first in man RNA mesothelin-targeted CAR-T trial dosed four subjects without prior lymphodepletion: 1 with pancreatic cancer and 3 with mesothelioma. These patients received 21 infusions, of which 20 occurred safely. One subject received two doses of the meso-RNA CAR-T, $1x10^8$ T cells at week 1 and $1x10^9$ T cells at week 2, before enrolling into an extended dosing cohort and within minutes of the 3rd infusion, suffered anaphylactic shock. Review of the case suggested that the delay between doses led to a priming of the patient's immune system to the murine sequence in the mesothelin-targeting scFv, leading to an IgE mediated anaphylactic event upon re-challenge with the meso-RNA CAR. As noted by Maus and colleagues, use of fully human sequences would be expected to reduce risk for an immunogenic reaction, Maus *et al.,* Cancer Immunol Res. 2013 Jul; 1: 26–31. U Penn is also conducting a trial combining CD19 CAR-T with a mesothelin CAR-T on the basis that the CD19 CAR-T will remove B cells that would otherwise target the mesothelin CAR-T, reducing its persistence, NCT02465983. Several CAR companies including JUNO, KITE, and Novartis (NVS) have developed CD19 CARs with fully human sequences that are likely to be less immunogenic than their murine counterparts, and these agents have entered the clinic.

Several CARs using human scFv sequences in place of murine sequences have entered or are scheduled to enter the clinic. In addition to reduced immunogenicity some investigators have reported that the efficiency of CAR expression is increased with human sequences when compared to murine versions at the same multiplicity of (lentiviral) infection. As reported by De-Gang Song and colleagues, Oncotarget. 2015 Aug 28; 6(25): 21533–21546, use of human scFv sequences increased C4 folate receptor-alpha (αFR) CAR expression, allowing for a reduction in the number of viruses required for product manufacture. De-Gong Song

notes that use of fewer viruses offers potential cost savings and presents the opportunity to use a less potent human CAR to increase the therapeutic window. Such an approach may allow for the same level of efficacy against a tumor expressing levels of αFR above a high threshold, while causing less toxicity against healthy tissue expressing αFR below the threshold required for CAR-T activation. Companies developing CAR-T therapies report screening up to 100 different scFvs to achieve the desired characteristics, including the target threshold required for CAR-T activation.

At the recent 2016 American Society of Clinical Oncology (ASCO) & American Society of Hematology (ASH) meetings, U Penn and NVS presented data for CTL119, a CD19 CAR-T using a humanized scFv in patients failing prior CAR-T. In U Penn's original pediatric series with CTL019, there was a 93% overall response rate (ORR), but a 22% rate of early B cell recovery (less than 6 months). With 15% of patients having a CD19+ve relapse, some went onto receive a transplant. In the face of poor CAR-T persistence or where the risk of relapse was felt to be high, U Penn re-treated 18 pediatric patients, including 3 patients with CD19+ve relapse from the NCT01626495 (CHP959) clinical trial. Response to reinfusion was uncommon, and with U Penn concerned about an immune response to the murine scFv, a humanized CD19 CAR was developed. U Penn opened a trial enrolling patients who had previously received a CD19 CAR and either not responded, relapsed after CAR-T therapy, or exhibited B cell recovery indicative of CAR-T loss. At ASH, data from 36 pediatric ALL patients were reported – approximately 60% had received a prior transplant, 10% had primary refractory disease, 20% were in 1st relapse, and 65% were in 2nd relapse or greater; however, 50% of patients had negative minimal residual disease at the time of enrollment, but were at high risk for relapse, including some patients with B cell recovery; 14 patients had received treatment with CTL019 and all patients were lymphodepleted prior to CRL119**. U Penn reported a 100% complete response rate in 22 CAR-T-naïve patients and 8/12 (57%) in patients who had received a prior CAR-T, including 2/5 MRD-ve, but B cell +ve patients and 6/19 with CD19+ve disease.** Relapses were observed in 3/22 CAR-T-naïve patients, 2 of which were CD19 –ve, and 3/8 responders in the CAR-T experienced cohort. With 7-10 months of follow-up, relapse-free survival was 75-83% in the CAR-exposed and CAR-naïve cohorts. Durable B cell aplasia was observed in 2/8 CAR-exposed subjects and 14/22 CAR-naïve subjects. U Penn noted that CTL119 toxicity was very similar to CTL019. **U Penn notes that while the humanized CD19 CAR can produce responses in patients failing a murine CD19 CAR, immunogenicity may still be problematic and contribute to suboptimal persistence.**

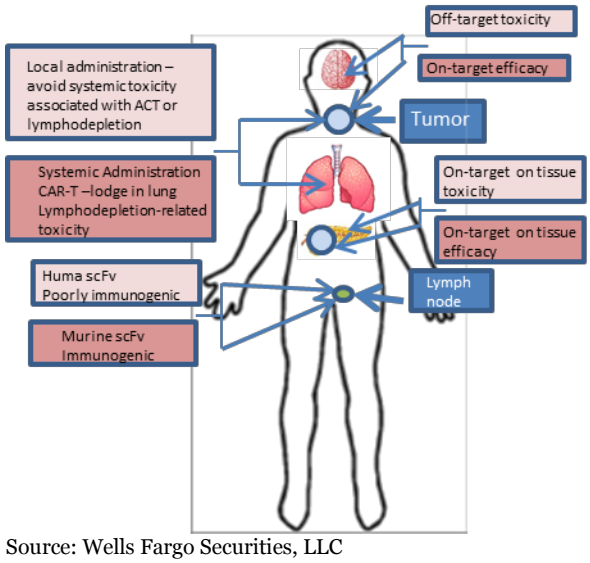
On-target off-tumor effects- CAR-T

Choice of tumor antigen for the CAR has important implications for safety as antigens that may also be expressed on critical tissues require more careful targeting than those on dispensable tissues. Persistence of CD19 CAR-T cells for 5 years or more in some patients is associated with B cell aplasia due to on-target CD19 targeting of a continuous supply of healthy B cell precursors by the CAR and patients require chronic immunoglobulin replacement therapy. In the case of targeting tumor antigens, persistence may be shorter as there is hopefully not a continuously renewing source of tumor antigen unlike CD19 on B cell precursors. While B cell aplasia is an unwanted but survivable side effect of CD19 CAR-T, on-target but off tumor toxicity affecting critical organs is unacceptable.

In 2010, the National Cancer Institute (NCI) published a case report from the first patient dosed on a CAR-T trial targeting HER2. **Within 15 minutes of CAR-T infusion, the patient developed respiratory distress associated with a pulmonary infiltrate, and after 5 days, the patient died.** NCI speculated that the CAR-T cells recognized low levels of HER2 in the epithelial lining of the lung, leading to T cell activation, Morgan *et al*., Mol Ther 2010 Apr;18(4):843-51. More recently, HER-2 CAR-T trials have been conducted safely, including a recently report from Baylor, Ahmed *et al.,* JCO May 20, 2015 vol. 33 no. 15 1688-1696. The HER2 experience is not unique, and while not fatal, targeting carbonic anhydrase IX (CAX9) with a CAR in renal cell carcinoma was associated with on-target off–tumor effects on normal epithelial cells of normal biliary ducts, leading to immune-mediated cholangitis, Lamers *et al*., Mol Ther 2013;21:904–12.

Recently, HER2 CAR-T products have been tested safely in the clinic, and this likely reflects an issue of potency. Several researchers have shown that by tailoring the affinity of the scFv, only cells with a high target density will be killed, while those healthy cells with a low density will be spared.

Exhibit 14. Summary of selected ACT toxicities



On vs off target	Neurotoxicity is an off target toxicity associated with CD19 CAR-T as was titin cross reactivity with MAGE-A3
On target toxicity vs efficacy	Loss of normal B cells or prostate tissue is on-target toxicity while loss of malignant B cells/prostate tissue is an on-target efficacy
Immunogenicity – human vs murine	Human sequences are less immunogenic than murine ones and may increase persistence
Local vs systemic administration	ACT lodge in pulmonary circulation for ~4h following systemic administration leading to greater risk for off-tissue toxicity vs local administration where ACT are retained at the tumor site

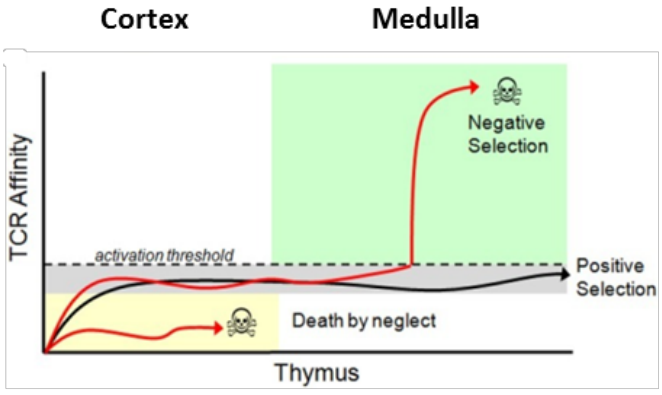
Source: Wells Fargo Securities, LLC

Engineered TCRs – Natural vs Affinity Matured TCR Therapeutics

Thymic selection of TCRs targeting self-peptides produces T cells with a potency that is suboptimal in comparison to that which can be achieved against the same target with TCR engineering. Development of high potency affinity matured TCRs targeting self-peptides specific to cancer offers an opportunity to optimize T cell-based therapy. However, the absence of thymic selection removes the critical safety step of negative selection.

Recall that MHC class II molecules present foreign antigens to the immune system, while MHC class I presents internal self-peptides mostly derived from self, but could include those derived from intracellular pathogen such as viruses. **The goal of T cell education is to delete, by negative selection in the thymus, T cells that recognize self with high avidity that could result in autoimmune disease, but allow T cells recognizing foreign peptides by positive selection in the thymus to mature**; 99% of T cells entering the thymus are eliminated by this process, which is termed central tolerance. In Exhibit 15, the process of central tolerance is summarized. In the cortex of the thymus, immature T cells that are unable to bind to self HLA do not receive any costimulation and die by neglect. T cells that recognize HLA class I downregulate CD4, to become CD8+ve, while those recognizing class II downregulate CD8 to become CD4+ve. These immature T cells move to the medulla, where central tolerance is determined by interactions with thymic dendritic cells that present peptides. If these interactions are too strong due to high TCR affinity, these self-reactive T cells are deleted by negative selection.

Exhibit 15. Positive and Negative Selection in the Thymus



Type of tolerance	Mechanism	Site of Action
Central tolerance	Deletion editing	Thymus
Antigen segregation	Physical barrier	Peripheral organ
Peripheral anergy	Cellular inactivation by weak signal without costumulus	Secondary lymphoid tissue
Regulatory T cell	Suppression by cytokines	Secondary lymphoid tissue & site of inflammation
Activation induced cell death	Apoptosis	Secondary lymphoid tissue & site of inflammation

Source: NIH.gov and Wells Fargo Securities, LLC

Because central tolerance is imperfect, there are several other mechanisms, listed in the table of Exhibit 15, that help limit damage from recognition of self. Certain organs, such as the eye, ovary, testis, and brain, for example, are sites of immune privilege where either physical separation, or absence of, MHC expression are designed to prevent targeting organ-specific proteins. Absence of a strong second signal blunts an immune response that may occur if the T cell recognizes an antigen being presented by an antigen-presenting cell such as a B cell, macrophage, or dendritic cell in a secondary lymphoid organ such as a lymph node; checkpoint blockade is a mechanism that depletes the supply of or blunts the stimulus from costimulation. Regulatory T cells specifically blunt the activation of CD4 or CD8 T cells, while antigen induced cell death or AICD occurs when a T cell is stimulated in a repetitive fashion.

On-Target on-tissue effects- eTCR

In 2009, NCI published a report of a clinical trial that evaluated an affinity enhanced eTCR, following on from a previous trial using the native TCR without affinity enhancement, Johnson *et al.,* Blood 114,3: 535-548. In the first trial, NCI isolated tumor infiltrating lymphocytes (TIL) from a melanoma patient that recognized the MART-1 melanoma antigen, cloned the TCR, and engineered T cells to express the MART-1 TCR, treating 31 patients. Disappointingly, only 4 (13%) achieved an objective clinical response compared to response rates of 50% or more with unselected TIL in melanoma, suggesting that the TCR did not have a high enough affinity to trigger T cell activation.

By avidity enhancing the MART-1 TCR, NCI developed a more potent eTCR product that was tested in 36 patients. The response rate more than doubled, to 30%; however, 40% of patients suffered eye and ear toxicity secondary to MART-1-induced melanocyte damage; in the original MART-1 TCR trial, no patient exhibited any eye or ear toxicity. In this case, binding of the TCR was strong enough to trigger T cell activation, but this occurred on both tumor and healthy MART1 +ve tissues.

Outside of melanoma, NCI has also reported significant toxicity, with a TCR targeting carcinoembryonic antigen, CEA, an antigen that is over-expressed in many epithelial cancers, including colorectal cancer, Parhurst *et al.,* Molecular Therapy, 2011 Mar; 19(3): 620–626. NCI derived a TCR to amino acids 691-699 of CEA using its human HLA-A*0201 transgenic mouse and then increased the avidity using a single amino acid substitution. Three patients were treated, with one patient achieving a partial response (PR) 4 months post CEA TCR treatment, one patient achieving a 17% reduction in metastatic disease to the lung at month 2, and a third patient showing no response. Any clinical benefit was not durable, with progressive disease noted at months 5 and 6 in the two patients exhibiting a response.

NCI notes that hematologic recovery following lympho-depletion was similar to that seen in its other trials; however, grade 2 or 3 diarrhea started between days 5 and 8, and lasted for 2 weeks before resolving by weeks 4-6. Colonoscopy revealed inflammatory colitis in all three patients, caused by the eTCR targeting CEA on healthy tissue.

Off-Target off-tissue effects- eTCR

By substituting amino acids in the TCR to increase binding affinity to the target antigen, the potential for an unexpected cross-reaction with an unrelated peptide increases and became fatally apparent in some of the first eTCR trials.

Both NCI and ADAP have described early clinical trials targeting MAGE-A3 that led to fatalities, due to cross-reaction between the eTCR and, in one case, a totally unrelated protein, and in the second case, a related protein. In 2013, Linette and colleagues describe two fatalities that occurred due to an unexpected cross-reaction between an avidity matured MAGE-3 HLA-A*01 restricted TCR and the unrelated cardiac protein titin, Linette *et al.,* Blood. 2013 Aug 8; 122(6): 863–871. MAGE-A3 is a cancer testis antigen and is rarely expressed on normal tissue, and the parental TCR for this product was isolated from a melanoma patient who exhibited no cardiac toxicity, likely due to the natural process of thymic selection for a low-affinity TCR, which would not be capable of causing damage to tissues expressing low levels of the MAGE-A3 antigen. Following MAGE-A3 eTCR treatment, a MAGE-A3 positive melanoma patient died 4 days after receiving the TCR and a MAGE-A3 positive myeloma patient died 5 days after receiving the TCR.

What is particularly instructive about this case is the fact that the eTCR did not recognize any of 38 normal cardiac-derived primary cells, including 10 confirmed as HLA-A*01. However, cardiomyocytes derived from induced pluripotent stem cells, iCells, which in culture exhibit a mixture of spontaneously electrically active atrial, nodal, and ventricular-like myocytes with typical cardiac biochemical, electrophysiological, and mechanical characteristics, were robustly targeted by the eTCR to MAGE-A3, but not the parental wild type MAGE-A3 TCR. Titin expression required a multicellular configuration, suggesting that panels of cell lines may not be sufficient to screen for cross-reactivity and organoid cultures can add an additional layer of safety.

Local versus systemic administration

Because of the potential for systemic toxicity and acknowledging that systemically delivered CAR-T can become physically affected in pulmonary circulation for several hours following administration as observed in the HER2 CAR patient treated at the NCI, **some investigators are evaluating local CAR-T administration.** Another advantage of local delivery is that it obviates the need for the use of systemic chemotherapy to achieve lymphodepletion, which in itself causes toxicity..

- **John Maher at Kings College in the United Kingdom is developing a CAR-T therapy for use in head and neck cancer, NCT01818323.** Maher is focused on epidermal growth factor receptor (EGFR or erbB) found in head and neck cancers and is using the promiscuous erbB ligand, T1E as the antigen-binding domain of the CAR since it binds to all 8 erbB homo or heterodimers, including those incorporating EGFR and HER2. At the recent 2017 AACR, data were presented for 10 patients treated at one of 3 dose levels- 1, 2, or $3x10^7$. Two patients were ineligible for analysis due to death or before treatment, or before the day 43 analysis. At dose level 1, 1 patient achieved stable disease (SD) and 2 patients had progressive disease at d43. At dose level 2, 1 patient died, with one each SD and PD. At dose level 3, all 3 patients had SD. Toxicity associated with the CAR-T was limited to grade 2 fever and chills at dose level 3, but no CAR-T were detected systemically. Maher has also evaluated intraperitoneal (ovarian) and intra-pleural (mesothelioma) dosing in animal models as other opportunities for local administration; however, some evidence of CRS in animals may reflect that these approaches present similar challenges to systemic dosing.

- **Local delivery to the CNS is also being evaluated at Mustang Therapeutics; a City of Hope spin-out recently updated its experience with a locally administered CAR-T therapy for glioblastoma**. A phase 1 trial of a 2nd generation IL-13R2α was recently initiated in 36 patients (NCT02208362) evaluating 3 weekly dose-escalated intratumoral infusions of IL-13BBZ/CD19t; the CD19t refers to a truncated CD19 that can be used to selected cells expressing the CAR. The trial includes 3 strata – intratumoral administration in resectable disease, intra-tumoral administration in unresectable disease and intra-cavitary for multifocal presentation of disease. In a recent update, City of Hope (COH) reported that dose level 1: $2x10^6$, $10x10^6$ & $10x10^6$ for the 3 consecutive doses, was well tolerated with no grade 3 adverse events, no CRS or neurotoxicity, and the trial is to proceed to dose level 2 evaluating 5 fold higher doses. City of Hope (COH) reported that 3 of 5 patients in the resectable cohort have SD following resection and CAR-T therapy. Interestingly, observations for control of metastatic disease in one patient following an initial response to intratumoral injection led to intrathecal administration, leading to control for 7.5 months, Brown *et al*. N Engl J Med 2016;375:2561-9.

- In advanced hepatocellular carcinoma, chemotherapy is often delivery locally via transcatheter arterial infusion (TAI). **Chinese company Shanghai GeneChem Co recently initiated two phase 1 trials delivering a glypican3 targeting CAR-T therapy using TAI for hepatocellular carcinoma and a mesothelin CAR-T in pancreatic cancer.**

Transfecting the Wrong Cell – What Can Happen When a CAR Gets Expressed in an ALL Blast

At the June 21, 2016, Recombinant DNA Advisory Committee (RAC) meeting, U Penn presented data showing that approximately 9 months after receiving CD19 CAR-T therapy for ALL, a patient relapsed from a CR with CD19 CAR+ve ALL. **Following detailed analyses, U Penn concluded that this CAR+ve ALL blast was created during the manufacturing process, and despite displaying CD19, was not eliminated by CD19+ve CAR-T+ve T cells.** Detailed analysis of other CAR-T products showed that the presence of a CAR+ve ALL blast in the product was not uncommon, but to date, CAR+ve ALL relapse has been observed in only a single patient out of 115 treated. Sequencing of the immunoglobulin (Ig) genes in the apheresis and bone marrow relapse product confirmed that the leukemia at relapse was present in the apheresis product. Evaluation of 18 patient products showed that a CAR+ve ALL contaminant was found in a second patient at relapse. This patient relapsed on day 62 post CAR-T with a CD19-ve ALL with the CAR+ve ALL cells making up less than 1% of the leukemic burden. Deep Ig sequencing of these 18 patients showed that in an additional 5, CAR+ve ALL cells could be found making up a median burden of contamination of 0.18%.

The CD19 CAR-T process in the setting of CD19+ve B cell malignancy is considered self-purifying. Why CD19 +ve ALL cells survive the manufacturing process is unclear; however, U Penn has some data suggesting that the CD19 CAR masks CD19 protecting the cell from CD19 CAR-T attack. In the case of the index patient, the CAR was inserted at two sites in the ALL cell's genome, neuropilin 1 and a carboxylase gene, PCCA, and it is unclear if the aggressiveness of the ALL changed due to the CAR insertion.

In summary - Current Applications of Adoptive Cellular Therapy (ACT) :

- TILs that are derived from a patient's tumor, expanded in the laboratory, and returned to the patient following lymphodepletion;
- eTCR using an affinity matured TCR against a tumor associated antigen presented by HLA to re-program autologous cells in HLA-matched patients, and
- CAR-T using a CAR against a tumor associated antigen to re-program autologous cells in an HLA-independent manner.

Key features of these three approaches are summarized in Exhibit 16. TILs represent the simplest approach, wherein the T cells are removed from the patient, i.e., their origin is autologous. The precise target of the TIL is unknown, but once expanded in the laboratory, the T cell population is assumed to contain T cells that target an HLA presented antigen on the tumor via a tumor antigen-specific TCR. Patients receive lymphodepletion therapy prior to receipt of the TIL product, which takes 4-6 weeks to manufacture and contains 10^{10} -10^{11} T cells, and despite the large number T cells, TIL-related toxicities are uncommon.

By comparison, both eTCR and CAR products are typically engineered using viruses to transfer DNA encoding for the recombinant tumor targeting proteins. The major difference is the way the engineered T cells target tumors. In the case of eTCRs, a specific tumor peptide presented by HLA is targeted, while CARs target a cell surface tumor protein without the requirement for presentation by HLA. Both CAR and eTCR products are typically autologous, and while the CAR can be used in all patients irrespective of their HLA subtype, each TCR can only be used in an HLA matched patient. As with TIL, CAR-T and eTCR products are administered after lymphodepletion, although as an adjunct to a stem cell transplant, this step is unnecessary since the stem cell transplant patient receives lymphodepleting or myeloablative therapy as part of the transplant procedure. At least one locally administered CAR-T product has been doses without lymphodepletion. Initial ACT trials used post ACT IL-2, and while LBIO continues to dose IL-2 following TIL, most CAR-T and eTCR trials no longer use post-infusion IL-2.

Both TIL and eTCR products have been extensively tested in solid tumors, while most experience for CAR products has been in hematologic malignancy. Doses of eTCR products tend to be higher than those for CAR products requiring additional manufacturing time, and while CAR manufacturing time lines are currently around 1 week, the time from apheresis to infusion is 2 weeks. Manufacturing time lines for TIL are 4-6 weeks. In contrast to the benign safety profile exhibited by TIL, product-related fatalities have been reported for both eTCR and CAR products. In the case of eTCR products, the fatalities have been related to cross-reactivity to off-target peptides on critical structures including heart and brain and for CARs, fatalities have largely been due to cytokine release syndrome (CRS) or cerebral edema associated with on-target CAR activity, and in the case of a HER2 targeting CAR, a case of on-target cardiopulmonary toxicity.

Exhibit 16. Key features of ACT circa 2015

1st generation	TIL	eTCR	CAR
Target known	No	Yes	Yes
Viral transduction	No	Yes	Yes
Engineered	No	Yes	Yes
Polyclonal	Yes	No	No
Cell surface target	No	No	Yes
MHC required	Yes	Yes	No
Autologous	Yes	Yes	Yes
Lymphodepletion	Yes	Yes	Yes
Solid tumors	Yes	Yes	No
Stem cell derived	No	No	No
Manufacturing time	4-6 weeks	2-3 weeks	2 weeks
Dose	10^{10}-15^{11}	10^{9}-10^{10}	10^{6}-10^{7}
Safety	None	Off-target fatalities reported	sCRS Neurotoxicity (CD19)

Source: Wells Fargo Securities, LLC

ACT Product Composition – how important is it?

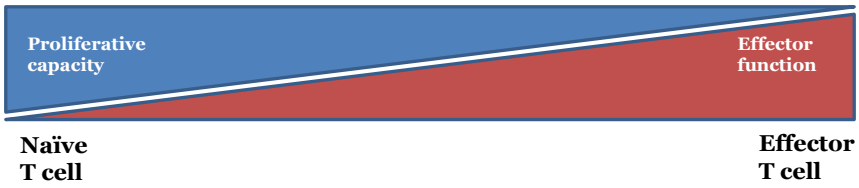
Given the broad repertoire of T cell phenotypes and their varying capability for expansion, persistence, target killing, and establishment of immunologic memory, there is increasing scrutiny on the composition and quality of T cell products. **ACT products have been extensively characterized, and there is considerable interpatient heterogeneity with respect to T cell composition** that reflects many factors starting before apheresis right through manufacturing to host factors following ACT infusion. The specific disease is not unsurprisingly important, and in chronic lymphocytic leukemia (CLL), for example, U Penn recently noted that patients receiving IMBRUVICA immediately prior to CD19 CAR-T therapy responded better than those not receiving IMBRUVICA prior to the CAR-T. U Penn showed that one potential explanation is that IMBRUVICA improves T cell function in part by down regulation of PD-1, Fraietta *et al.,* Blood. 2016 Mar 3;127(9):1117-27.

Following administration, the ACT T- cells need to expand and persist long enough to clear the tumor. T cells come in 2 major varieties distinguished by the presence of different molecules on their cell surface, namely CD8 and CD4. CD8+ve T cells are highly cytolytic, leading to a variety of associated names in the literature, including killer T cells, or cytotoxic T lymphocytes (CTL). CD4+ve T cells perform a helper function to other immune cells, but when the transcription factor, FOXP3 is expressed, form regulatory T cells (T_{REG}) that inhibit the activity of cytotoxic T-lymphocytes (CTL).

In almost all ACT trials, considerable interpatient product heterogeneity with respect to CD4/8 and central memory to effector memory ratios has been observed, and during the CAR-T manufacturing, several groups have observed a change in the nature of the population T cell phenotype toward one dominated by effector T cells. While these T cells are capable of killing tumor, they lack the ability to establish immunological memory. As noted in Exhibit 17, T-cell subsets can be differentiated and sorted based on the presence of absence of key cell surface markers. Traditionally, IL-2 has been used to expand T cells in culture, but use of IL-15 or IL-21 instead of IL-2 suppresses differentiation of naïve to effector T cells.

Exhibit 17. T cell Phenotypes

Phenotype	Cell surface CD markers
Naïve T cells	CD45RA+, CCR7+. CD27+, CD28+
Stem Central memory	CD45RA+, CCR7+. CD27+, CD28+
Central memory	CD45RA-, CCR7+. CD27+, CD28+
Effector memory	CD45RA-, CCR7-, CD27+/-, CD28 +/-
Effector T cell	CD45RA+/-, CCR7-. CD27-, CD28-



Source: Wells Fargo Securities, LLC

CAR groups such as those at Seattle Children's Research Institute/Fred Hutchinson Cancer Research Center (SCRI/FHCRC) have used a defined subset of T cells in their CAR-T products. These groups manufacture two separate CAR-T products for each patient, including 91) a CD4 product, and (2) a CD8 product, so that each patient receives a 50/50 split of CD4 and CD8 CAR-T cells. The 1:1 ratio of CD4 to CD8 cells is empiric, and given that CD8 CAR-T become predominant, perhaps due to improved persistence, SCRI is planning to evaluate a higher proportion of CD4+ve T cells to compensate for its less robust expansion following dosing.

Unlike effector memory T cells, which are non-engrafting, stem central memory T cells engraft and can produce both effector T cells and effector memory T cells. The FHCRC/SCRI and City of Hope (CoH) groups have tried to isolate just central memory CD8+ve cells from the apheresis product for CAR-T manufacture, but in Seattle at least, this is currently too onerous for routine practice.

In contrast to the Seattle experience, the NCI has reported that based on their clinical data they have no bias toward either central of effector memory T cells in the CAR-T product. However as reported recently by NCI impressive pre-clinical data for T_{SCM} has prompted them to modify an ongoing CD19 CAR-T clinical trial to evaluate a T_{SCM} CD19 CAR-T product.

The NCI recently reported on their approach to generate T stem central memory (T_{SCM}) starting from naïve CD45RA/CCR7 T cells, Sabatino *et al.,* Blood, 28 July 2016, Vol (128; no. 4). In their work NCI activated a large numbers of naïve T cells with their standard CD3/CD28 protocol but with the addition of IL-7, IL-21 and a glycogen-synthase-3β inhibitor and sequential magnetic bead separation for CD8+ve then CD62L+ve and finally CD45RA+ve T_{SCM}. Based on data from 6 donors, NCI achieved a median of 1.5 $x10^8$ +/- $0.6x10^8$ T_{SCM} cells from an apheresis product containing 3.8-8 $x10^9$ cells. NCI choose these reagents to:

- Provide key instruction signals for TSCM formation – IL-7
- Inhibit effector T cell differentiation – IL-21
- Activate β catenin/WNT activity to maintain appropriate transcription factor expression - glycogen-synthase-3β inhibitor

While NCI has a humanized CD19 CAR in clinical testing, the cells were transfected with their standard FMC63 CAR. NCI was surprised by the 73% transfection efficiency achieved using its standard retroviral protocol, given the dogma that retroviruses only transfect replicating cells and NCI's avoidance of T cell replication as part of the T_{SCM} production process. NCI confirmed that its induced T_{SCM} cells exhibited the same transcriptional profile as native T_{SCM} cells, supporting the fidelity of its process. While cytokine production by T_{SCM} CD19 CAR-T was similar to standard CAR-T and in animal models persistence also similar, NCI suggested that its T_{SCM} product exhibited improved metabolic fitness; NCI used CD28 as the costimulatory domain.

Anti-leukemic activity of the T_{SCM} was superior to standard CAR-T, as evidenced by increased tumor control and longer overall survival. Interestingly, NCI noted that the T_{SCM} cells were poor producers of IL-6, the key mediator of cytokine release syndrome, following leukemic cell challenge and based on the totality of its observations, NCI has modified an ongoing protocol, NCT01087294, to test a T_{SCM} CAR. As noted, NCI continues to use CD28 as a co-stimulatory domain for CD19 CARs; however, T cell memory phenotype can also be influenced by CAR architecture. U Penn researchers suggest that inclusion of 4-1BB in the CAR architecture promoted the outgrowth of long-lasting CD8(+) central memory T cells that had significantly enhanced respiratory capacity, increased anaerobic fatty acid oxidation, and enhanced mitochondrial biogenesis, as noted in Exhibit 18.

Exhibit 18. Costimulation and Metabolic Programming

	CD28	41BB
Persistence	↓	↑
Memory	Effector	Central
Fatty acid metabolism	↓	↑
Mitochondrial biogenesis	↓	↑
Metabolism	Glycolytic	Oxidative

Source: Wells Fargo Securities, LLC

In contrast, CAR T cells with CD28 domains favor the generation of short-lived effector memory cells with a genetic signature consistent with enhanced aerobic glycolysis and in head-head animal studies, **CD28 CAR-T tend to eliminate tumors faster than 41BB CAR-T cells,** Zhao *et al.,* Cancer Cell, 28, 415–428, 2015. Metabolic reprogramming by choice of co-stimulation domain requires further research, in particular, in terms of how it affects CD4 CAR-T, as well as the potential impact on other immune cells in the tumor microenvironment.

Further, in considering the treatment of solid tumors where enhanced anaerobic capability may be more resistant to low oxygen levels found in tumors, 41BB may be preferred as in addition to enhanced anaerobic ability vs. CD28, 41BB protects CAR-T from an exhausted phenotype, potentially increasing persistence. To illustrate this point, MSKCC researchers recently published data suggesting that at low CAR-T dose levels, 41BB CAR-T retained its cytotoxic and cytokine secretion functions longer than CD28 CAR T cells in the setting of immune-induced PD-1, Cherkassy *et al.,* J Clin Invest. 2016 Aug 1;126(8):3130-44.

Persistence – Likely critical for success in solid tumors

In stating the obvious, ACT therapeutic T-cells need to persist long enough to eliminate the last tumor cell, but also to establish immunologic memory to combat relapse and future metastatic spread. In the most mature ACT data, NCI researchers noted that 12 months after administering a TIL product in a melanoma patient with a complete response (CR) ongoing for 3 years, 4 of 5 T cell clones that were shown to identify the patient's tumor were present after 12 months, leading NCI to suggest that persistence was required to maintain the ongoing CR, Cancer Immunol Res. 2016 Jun 16. pii: canimm.0215.2015.

While several factors likely influence persistence, T cells need to see antigens to expand, and several strategies have been and continue to be evaluated to support ACT expansion and persistence, including the use of vaccines. T cells are programmed to rapidly expand in number in the presence of a specific immunologic threat and then just as rapidly, numbers contract. In the setting of bulky disease, scientists assume there is sufficient antigen present to ensure that an initial rapid proliferation of CAR-T occurs, and in acute lymphocytic leukemia (ALL), at least there is a strong correlation of efficacy to peak CD19 CAR-T numbers 1-2 weeks after infusion.

In the setting of minimal residual disease (MRD) there is concern that insufficient tumor will lead to sub-optimal expansion of CAR-T, which is insufficient to clear the residual tumor. In an attempt to counter this, some academics have selected T cells from donors that recognize common viral pathogens such as cytomegalovirus (CMV) or Epstein Barr virus (EBV) as a source for ACT. In this case, T cells can be stimulated either by its endogenous TCR recognizing viral peptides, or by the CAR or eTCR. Baylor has conducted several trials with viral-specific CAR-T, although initial results were disappointing. Baylor continues to conduct trials with bispecific T cells, including a host TCR targeting a virus and a CAR:

- A GD2 CAR-T in sarcoma patients who have previously received a varicella vaccine or have suffered the chicken pox. Varicella zoster-specific T cells are transformed with the GD2 CAR and the CAR-T is administered with a varicella vaccine, NCT01953900.

- A tri-virus-specific T cell was developed by culturing patient T cells with virally infected irradiated donor-derived antigen presentation cells, which were then transfected with a CD19 CAR, NCT00840853.

FHCRC has also conducted trials of an eTCR using virus specific T cells, again allowing for viral reactivation post-transplant to help stimulate the engineered T cells, in this case a WT-1 eTCR for AML. In this setting, the T cells are doing double duty, preventing clinical viremia and targeting leukemia. Instead of relying on host-provided antigens, City of Hope is planning to combine a CMV-specific CD19 CAR-T with a CMV vaccine to allow for vaccination as a way to support CD-19 CAR-T persistence.

Even in the context of CD19 CAR-T therapy for ALL, SCRI researchers have identified a population of patients that show suboptimal response using 15% CD19+ve cell involvement in the bone marrow as cut-off. Patients with less than 15% CD19+ve cell involvement in the bone marrow at baseline showed inferior outcomes to those with more than 15% CD19+ve involvement, prompting the researchers to propose the use of a CD19+ve vaccine in patients with less than 15% CD19+ve involvement. Specifically, for these patients, the researchers plan to engineer an autologous CD19-expressing T cell in addition to the CD19+ve CAR-T; in the context of SCRI's trials, each low CD19 patient requires three products to be manufactured – CD4 CAR-T, CD8 CAR-T and CD19+ve T cell. Manufacturing 3 products for a patient is acceptable as a scientific experiment, but for practical reasons, a viable ACT product needs to be engineered to include both tumor targeting and enhanced survival capability in the tumor microenvironment (TME).

Target Loss Limits Persistence

Loss of target from the cancer cell short circuits T cell activation, leading to premature loss of ACT T cells. For CD19, splice variants, point mutations, and deletions have been described that lack the domain targeted by the current CD19 CAR-T products; complete lack of CD19 has not been described, reinforcing how critical CD19 signaling must be to B cell biology. Recently, German researchers described a novel mechanism of resistance to CD19 bi-specific product BLINCYTO, Binder *et al.,* Blood 2017 129:100-104. In short, Binder and colleagues describe a mechanism for CD19 resistance that does not invoke any changes to CD19 itself, but rather, loss of a protein that co-localizes ith CD19 on the cell surface.

Looking to the Future of ACT

The flexibility of the ACT platform has created an endless list of possible solutions for optimization. Given the potency of ACT and potential for unwanted toxicity, a variety of technologies are being deployed that aim to improve safety. Suicide switches kill the transplanted cells, and activation switches and rheostats allow CARs or TCRs to be switched on and off under the control of a separately administered compound, while targeting more than one antigen on the tumor, which should help differentiate tumor from healthy tissue to increase the safety window.

T cells do not act in a vacuum and require chemical signals, which may be in short supply in the tumor microenvironment, as well as interaction with other cells, including immune cells to provide critical signals. Modifying ACT to become more autonomous in order to provide their own homeostatic signals such as IL-15 or IL-12 can also leverage switch/rheostat technology. The tumor microenvironment presents a host of challenges, including proteins and small molecules that inhibit T cell and immune cell function, limited supply of nutrients and cofactors that are required to support immune cell function, and a hypoxic environment favoring anaerobic metabolism. Countering the TME can take the form of co-administering ACT with checkpoint inhibitors or drugs to modulate the availability of nutrients such as glutaminase, arginase, or indoleamine 2,3-dioxygenase (IDO) inhibitors. **Cells can also be genetically engineered to block or more creatively re-direct negative signals from the TME such as a chimeric receptor that combines the extracellular domain of PD1 with the intracellular domain of CD28, so that instead of PD-L1 on a tumor leading to T cell suppression, it leads to T cell activation.**

With respect to PD-1, outside of Hodgkin lymphoma, the role of PD-1 inhibition in hematologic malignancy has yet to be defined; however, with data suggesting that PD-L1 can limit the activity of CAR-T therapy, anecdotal data are emerging that suggest PD-1 inhibition can be combined with CAR-T. At the recent ASCO meeting, U Penn presented a case report of a young DLBCL patient who progressed 78 days after CTL019 with a large lesion. Four months later the patient received OPDIVO, and three months after that, the large lesion had become necrotic. On the basis of this, U Penn reported promising data in another patient for sequential use of a PD 1 inhibitor after CTL019.

With synthetic biology offering endless options, in our view, a challenge will be how to select the optimal strategy without performing thousands of experiments. Researchers at Seattle Children presented an in-vivo competitive strategy they are developing to rapidly test the myriad of options at the recent AACR. Mice received 4 different CD19+ve solid tumors and a panel of CD19+ve CAR-T each with a different engineered function. At days 7 and 14, for example, the SCRI researchers can ask which of the double-positive CAR-T cell pre-dominated in each different tumor and whether there was a change between days 7 and 14.

Exhibit 19. Summary of Strategies to Create Next-Generation ACT Products

Feature	Description	Commentary
Suicide switch	In the event of ACT-mediated toxicity, genetic modification of the T cell to include a suicide switch would allow for control of ACT T-cell number	The rapidity of suicide gene-induced cell death may be important - minutes - hours for apoptosis induction vs hours-days for antibody mediated clearance The totality of suicide-induced cell death may be relevant - some vs all MolMed's HSV-tk is the first approved suicide switch for T cells BLCM's rimiducid inducible caspase 9 suicide switch is the most widely used and validated system
Boolean switch	Another strategy to reduce toxicity and increase specificity of tumor targeting	The concept of and AND, OR or NOT switches moves beyond targeting of a single antigen. If a target is shared between a tumor and a healthy cell adding, a second target that is also found on the tumor but not on the same healthy cell utilizes allows for use of an AND gate to activate a CAR for example. The use of antigen A or B allows for tumor heterogeneity and may protect against antigen escape - targeting CD19 OR CD20 for example. The NOT gate ensures that when a cancer cell and a healthy cell express the same target but the healthy cell expresses a target that is not found on the tumor cell the CAR is inactivated by the NOT gate to protect the healthy cell
Activation switches and rheostats	A separate way of controlling the expression of a CAR or another gene is to include a ligand activated switch controlling its expression	The expression of the target gene, whether a CAR, TCR or cytokine for example is controlled by a ligand. ZIOP's activator veledimex is used to control the expression of IL-12. BLCM's rimiducid is can also be used to control CAR activation. In addition to an on/off conformation, adjusting doses of activators can be used to vary the level of gene activation or protein activity. A different concept allows for a universal CAR design which is inactive until a second component is added to target and activate the CAR .
Protecting against Activation Induced Cell Death (AICD)	Repeat stimulation of the TCR can lead to AICD; recall the intracellular signaling of a CAR is the same as a TCR	The use of programmed death - 1 inhibitors with ACT can be supported by many lines of evidence including their ability to reduce AICD
Chimeric receptors	The tumor microenvironment contains many immuno-inhibitory components	Creation of a designer chimeric receptor can redirect the dominant negative signal in the TME to a positive one by fusing the extracellular domain of the negative signal's receptor to the intracellular signaling domain of an immuno-stimulatory pathway. As an example the PD-1 extracellular domain can be linked to the intracellular domain of CD28, a classic 2nd signal for T cell activation
Protecting against the TME	Co-expressing enzymes in gene-modified ACT cells	Reactive oxygen species levels increase in the immunosuppressive microenvironment - expression of catalase along side a CAR showed improved protection against ROS and the secreted catalase produced a by-stander effect.
Short circuiting the TME	By uncoupling immuno-suppressive signals in the TME or redirecting them, the immunosuppressive TME can be blunted	Expression of regulator subunit I anchoring disruptor (RIAD) can be used to uncouple the immunosuppressive TME in ACT cells, while co-expression of CD40L can be used to extend the duration of time ACT cells interact with other immune cells. Low oxygen is found in the TME, by linking CAR expression to low oxygen not only is the TME subverted, but CAR activation was restricted to the hypoxic TME.
Moving beyond proteins	Tumors frequently show differential post-translational modification of proteins	While proteins may be shared between healthy and diseased cells, targeting glycosylation patterns unique to cancer has been shown to provide specificity
Off- the-shelf	The vast majority of ACT approaches use patient derived autologous T cell as their source material - moving to donor cell has several advantages but presents several challenges	By using off-the-shelf OTS, donor derived cells ACT products can be pre-manufactured and stored on site rather than manufactured to order. Donor T cells are inherently "healthier" than those from cancer patients as they are not under the influence of a tumor and by definition these products are not contaminated by tumor. However, donor cells need to be engineered to prevent them targeting the recipient and also for increased persistence engineered to prevent their targeting by the host. Induced pluripotent stem cells offer an opportunity to engineer cells at a single cell level in order to derive a uniform population that can be subsequently engineered for ACT.

Source: Wells Fargo Securities, LLC

The Potential Role of Suicide switches to Turn off CAR-T and eTCR Therapeutics

With patients' deaths due to severe product-associated toxicity, the concept of a kill switch to rapidly deactivate and kill the ACT T cell has been proposed. Currently there are three common approaches, which include viral thymidine kinase (tk), inducible caspase 9 (iC9), and antibody-mediated clearance. Conditional approval of MolMed's ZALMOXIS in Europe validates the use of ganciclovir to trigger tk mediated cell death in the setting of GvHD.

Induced caspase 9 (iC9) has also been validated in the clinic as a suicide signal for donor lymphocytes used in the setting of allo-SCT where the potential for severe or fatal GvHD exists, Zhou *et al.,* Blood, 25 June 2015, v. 125, No. 26, 4103-4113 . The use of iC9 requires genetic modification of target cells with a virus, analogous to modification of cells for a CAR or eTCR, and at the time for deployment, activation with the IV administered ligand, rimiducid. Originally developed at Baylor, BLCM has secured a GMP supply of rimiducid for commercialization of iC9; however, several academic groups are deploying iC9 in their eTCR and CAR T-cell products. Caspase mediated killing has been shown to be very efficient, with rapid 90% clearance of target cells within 30 minutes. Note that killing occurs without triggering an inflammatory response, which may be relevant in situations where the toxicity has an inflammatory component such as CRS.

Another approach is to genetically engineer T cells to display a non-functioning protein that can be targeted by a commercial antibody such as ERBITUX or RITUXAN targeting truncated EGFR and an altered CD20 respectively. To date, no experience had been reported in the clinic for using an antibody to terminate toxicity, and one concern is that antibody-mediated cell death would exacerbate ongoing inflammation such as that associated with CRS. In the pre-clinical setting, data suggest clearance of T cells takes hours to days, which may not be optimal in the setting of acute toxicity. Koneru and colleagues recently reported that the addition of ERBITUX to a CAR-T expressing a truncated EGF receptor (EGFRt) led to greater than 60% cell death in vitro after 4 hours, Journal of Translational Medicine (2015) 13:102, while a second report noted that EGFRt CAR-T cells were cleared 4-6 days after ERBITUX in mice, Wang *et al.,* . Blood. 2011;118:1255–63. While such strategies may not be optimal for cell killing, these altered proteins can be used to aid manufacturing as their expression can be used to isolate cells that have been transformed with both the altered protein and a CAR or eTCR, assuming both elements occur in tandem on the vector.

Deployment switches for Boolean activation

In order to broaden the therapeutic window for ACT, conditionally activated CARs would require "and/or" signals, for example, the expression of both tumor-associated antigen 1 (TAA1) AND TAA2, alternatively activation requires TAA1, but presence of TAA2, signifying a healthy cell shuts off expression – a NOT gate.

Researchers at the University of California and Seattle Children's published data for a CD19-OR-CD20 CAR using a single chain bispecific CAR, Zah *et al.,* Cancer Immunol Res. 2016 Apr 8. The researchers optimized structural parameters for efficient dual antigen recognition, allowing for equally efficient killing by recognition of CD20 or CD19. The researchers believe that use of a bi-specific single-chain CAR recognizing two antigens on the target may prevent antigen escape, which has been observed for CD19 targeting CARs. The authors do not discuss potential immunogenicity, but clearly, such novel constructs have the potential to be immunogenic, which could limit their persistence. German researchers have published a similar concept, but one that incorporates a CAR and a TCR targeting different melanoma antigens, Uslu *et al.,* Exp Dermatol 2016 Jun 1. doi: 10.1111. At the recent AACR, the NCI described the challenges it faced creating a dual CD19 CD22 CAR. While CD led to robust CAR activity, CD22 was more fickle, leading NCI to develop a bispecific construct that looped back on itself. Thus, instead of a CD19-CD19-CD22-CD22, the best design turned out to be CD22-CD19-CD19-CD22, where a hairpin between CD19 allowed for the formation of a functional CD22 binder. NCI and Stanford intend to begin clinical evaluation of this CAR shortly, with a biscistronic allowing for individual expression of separate CD19 and CD22 CARs from a single vector expected to enter the clinic in 18-24 months as a more optimal configuration. Interestingly, NCI chose to use 41BB as the costimulatory domain for the CD22 CAR-T, noting better persistence.

On-board cytokines and chemokines

Just as the 2nd generation CAR-T added a CD28 domain to the CAR architecture, rather than rely on its activation by ligating CD80 on the target cell, several other cytokines that are important in T cell biology are also in short supply in the tumor microenvironment (TME).

The interleukin, IL-15 is an important cytokine for T cells and NK cells that is a member of the IL-2 family, but unlike IL-2, it does not stimulate T_{REG} development. While soluble IL-15 has some activity, membrane-tethered IL-15 is considerably more active as it forms a synapse between two cells.

Altor Bioscience is developing a tethered IL-15 for systemic administration, and has entered an ACT collaboration with NK-U.S. MD Anderson has developed and received Recombinant Advisory Committee (RAC) approval to evaluate a tethered IL-15 engineered into ACT CAR –T. ZIOPHARM (ZIOP) is now responsible for development of the IL-15 CAR-T and, according to management, this is a high-priority program at the company. In the case of ZIOPHARM, the tethered IL-15 is introduced on a separate plasmid as part of the company's Sleeping Beauty transfection technology, and pre-clinical data show that approximately 50% of CAR+ve T cells also express tethered IL-15. MSKCC has also published data for the inclusion of a tethered IL-15 into T cells drives expansion of a central memory phenotype, which is desirable for establishing long-term memory. MDACC recently initiated clinical development of a CAR-NK that incorporates an untethered IL-15 and BLCM's iC9. Similarly MSKCC and JUNO have been working on "armored" CARs, which produce biologically relevant proteins in the TME. JCAR020 is currently in phase 1 testing in ovarian cancer, where it targets MUC16 and secretes low levels of IL-12, while a CD19 CAR-T that also presents 41BB ligand is scheduled to enter the clinic shortly.

Chemokines help homing, and noting that mesothelioma cells express high levels of the chemokine, CCl2, researchers at U Penn have incorporated the CCR2 receptor into mesothelin expressing CAR-T cells to improve their homing to mesothelin positive tumors, Moon *et al.*, Clin Cancer Res. 2011 Jul 15;17(14):4719-30.

Activation switches and rheostats

While the aforementioned iC9 suicide switch induces T- cell death by rimiducid-mediated homodimerization of an engineered caspase 9, other proteins can be configured to be controlled by switches. For example, Bellicum (BLCM) has configured the costimulatory domain of a CAR to allow for rimiducid control. Without rimiducid, the CAR fails to signal effectively; but with dosing of rimiducid, the co-stimulatory domain is activated with consequent full CAR activation. However, rimiducid is dosed intravenously (IV) and while once weekly IV may be acceptable, an oral agent is more convenient and potentially allows for greater control. At the recent AACR, BLCM presented pre-clinical data for dual control using rimiducid for activation, but a modified iC9 that used rapamycin instead of rimiducid to activate a suicide switch.

The most advanced oral switch in development is ZIOPHARM's (ZIOP) rheostat, which is a synthetic nuclear receptor that initiates transcription only in the presence of the oral activator veledimex. In the current iteration, the oral activator veledimex is used to initiate production of IL-12, a potent immune stimulating cytokine in the oncolytic virus, AdRTShIL12. IL-12 has been shown to counter the hostile tumor microenvironment through multiple mechanisms, including reactivation of anergic TILs, inhibition of T_{REG}-mediated suppression of CTLs, recruitment of NK cells to the tumor site, and inhibition of IL-10 and transforming growth factor beta (TGF-β) secretion by tumor-associated macrophages (TAMs). NCI is currently preparing to evaluate veledimex-controlled IL-12 production in TIL.

Previously, NCI evaluated the nuclear factor of activated T cell (NFAT) promoter to control IL-12 production by TIL on the basis that activation of the NFAT promoter occurs downstream of TCR activation and thus, a TIL encountering tumor would be able to produce IL-12 locally, Zhang *et al.*, Clin Cancer Res. 2015 May 15;21(10):2278-88. **Unfortunately, severe IL-12 mediated toxicity was observed in 2 clinical trials that were closed prematurely.** In the CAR-T field, MSKCC recently published the design of an IL-12 secreting CAR, Koneru *et al.,* Journal of Translational Medicine (2015) 13:102, following positive pre-clinical data suggesting that compared to the non-IL-12 version, the IL-12 CAR exhibited increased persistence of T cells leading to improved overall survival. Interestingly, in its pre-clinical studies, MSKCC showed that using a CD19 CAR-T model, incorporation of autocrine IL-12 or IL-18 production eliminated the need for lymphodepleting chemotherapy and that CD4 production of IL-12 was critical as prior studies with CD8 T cells and systemic IL-12 was unable to remove the requirement for lymphodepletion. From a safety perspective, MSKCC expects its construct to produce lower levels of IL-12 than observed in the NCI studies using an NFAT promoter, and clinical development of a MUC16 targeting IL-12 CAR-T began recently.

Both Bellicum (BLCM) and ZIOPHARM (ZIOP) are using intracellular switches. The same concept can be applied to extracellular domains, and both Bluebird Bio (BLUE) and Cellectis (CLLS) have developed extracellular switches using the same synthetic biology dimerization concept as BLCM.

At the 2016 American Society of Gene and Cell Therapy (ASGCT) and 2017 AACR meetings, BLUE described DARIC, a novel dimerization strategy for controlling CAR functions. By physically separating the ligand binding and signaling domains of a CAR and adding the rapamycin binding motif, FKBP12 to the target binding half of the CAR and the FKBP12 binding protein (FRB) to the signaling half of the CAR, full activity occurs only when rapamycin brings the two halves of the CAR together via dimerization of FKBP12 with FRB.

BLUE envisages DARIC as enabling a next-generation controllable CAR, outlining CD19 targeting as benefiting from such an approach, where stoppage of rapamycin leads to loss of CAR activity, allowing recovery of normal B cells after clearance of tumor. While rapamycin is a licensed product simplifying its use with BLUE's CAR technology, it's used as an immunosuppressive agent and thus, is not an ideal agent for use with an immune-based therapy. BLUE is evaluating if rapamycin can be used at a dose that works with DARIC, but is not immunosuppressive. BLUE's alternative is to follow in BLCM's path by using another of Ariad Pharmaceutical's (ARIA) portfolio of FKBP12 binding rapalogs that do not bind to the native human FKBP12, but bind only to a mutant FKBP12 used in DARIC. Cellectis (CLLS) has described a very similar system that uses a rapalog to activate the CAR.

In an extension of the DARIC concept, incorporation of FKB12 binding protein (FRB) into the external domain of a standard CAR allows for switching target selection. In DARIC Plugin, an alternate CAR target binding domain modified with FKBP12 can bind to the FRB-CAR in the presence of pre-bound rapamycin, effectively redirecting the CAR's specificity. **BLUE has demonstrated that a B cell maturation antigen (BCMA) CAR-T could become a CD19 CAR-T using DARIC Plugin to bind a CD19 scFv to the BCMA CAR, allowing for functionality in the absence of BCMA, but presence of CD19.** Such a strategy may have utility in the setting of antigen loss by the tumor, allowing extension of CAR functionality without manufacture of a new product; we understand that BLUE is evaluating how to adapt DARIC Plugin for use with commercial antibodies.

The development of a universally generic CAR offers the potential to control both CAR activation and selection of target. Unum Therapeutics uses a CD16 motif on the CAR's extracellular domain instead of a standard scFv. CD16 bids to the Fc domain on one end of a monoclonal antibody, allowing the other end to bind to its specific target, creating a bridge between the CAR-T and the tumor cell. For example, administration of the universal CAR with RITUXAN would allow RITUXAN to bind to CD20 on a lymphoma cell and to the universal CAR, leading to CAR-T activation against the lymphoma cell. With such an approach there is competition between CD16 on the CAR and other immune cells. German academics recently described a similar universal CAR concept, the UniCAR, Cartellierii *et al.* Blood Cancer J. 2016 Aug 12. Instead of a scFv, the UniCAR recognizes a non-immunogenic 10 amino acid tag from the human nuclear autoantigen. CAR specificity is determined by a second component that carries both the 10 amino acid tag and the scFv. In the Carterllierii manuscript, the authors describe a proof of concept using CD33 and CD123 either alone or in combination to allow for simultaneous targeting.

Intelligent switches form the core technology for UCSF spin-out Cell Design Labs, which is building switches designed around synthetic Notch constructs developed in the laboratory of Wendell Lim. Activation of the Notch pathway requires engagement of a ligand to a receptor that leads to recruitment of proteases, releasing the internal domain that then migrates to the cell's nucleus, where it results in transcription of certain genes. Conservation of the proteolytic cleavage sites on Notch allow for the replacement of extracellular and intracellular domains to create synthetic Notch receptors.

In 2016, Lim and colleagues reported on the application of synNotch to CARs wherein transcription of the CAR is regulated by the presence of a triggering ligand that leads to release of a transcription domain from the Notch cleavage site, which binds to the CAR promoter, leading to CAR production, but only in the presence of the activating ligand. And in its absence, CAR expression decays from the cell surface over a period of hours. Lim, Cell. 2016 Feb. 11. In experimental models, Lim and colleagues have shown that using the model protein green fluorescent protein (GFP) that CD19 CAR production is produced only in the presence of co-expressed GFP, and that targeting of CD19+ve but GFP –ve bystander cells was not observed. KITE entered into a collaboration with CDL to use CDL's switch technology in CAR-T products, with a CLL1-targeting CAR-T for AML and a CD19 CAR-T in early development.

Switches and the Blood Brain Barrier

In the context of CD19 CAR-T, there is considerable neurological toxicity that is undesirable in settings where the brain is not a cancer site. The ability to restrict activation of CAR-T/ eTCR products outside of the BBB may have safety advantages, and while products activated in the periphery could cross the BBB, if the continued expression of the CAR/eTCR was dependent upon presence of the activating compound, if that were unable to cross the BBB, CAR/eTCR related neurotoxicity may be ameliorated. Currently the two most advanced switch technologies, ZIOP's veledimex and BLCM's rimiducid, are able to cross the BBB. In fact, ZIOP's current application of veledimex is in the setting of glioblastoma, where crossing the BBB is required. In the case of BLCM, rimiducid doses have been optimized for cell killing and BLCM has demonstrated using lumbar puncture that iC9+ve T cells in the CNS can be killed by systemic rimiducid. For both veledimex and rimiducid, it may be possible to define a dose that activates switches for CAR or eTCR expression/activity in the periphery, but does not reach the threshold required for activation in the CNS.

Protecting against activation induced cell death (AICD)

Activation-Induced Cell Death (AICD) can be a natural consequence of repeated stimulation of the TCR by an antigen, and in a healthy immune system, provides a back-up to avoid targeting self, as well as a mechanism for reducing T cell numbers after the immunological threat has passed

Clearly in the context of ACT, premature AICD could prevent the development of a curative immune response. Researchers from Baylor College of Medicine recently published an analysis of AICD, with a 3rd generation CD28-OX-40 CAR directed to the ganglioside antigen, GD2, a well-characterized target in neuroblastoma, Gargett *et al.,* Mol Ther. 2016 Mar 29. *In-vitro* this CAR-T exhibited immediate effector function without functional exhaustion against GD2+ve target cells. In long-term culture, cytokine production and CAR-T mediated cell killing capacity was observed to diminish, but this could be reversed by adding an inhibitor of the programmed death 1 (PD-1) pathway. Further analysis revealed that significant AICD was occurring with repeated antigen stimulation and this, too, could be blocked by the addition of a PD-1 inhibitor. Note that analysis of patient data from an ongoing GD2 CAR-T trial in melanoma, CARPETS, revealed deletion of CAR-T *in-vivo* suggestive of AICD. While dosing a CAR-T with a PD-1 inhibitor is feasible, it requires administration of two separate drugs. Engineering ACT T cells to secrete a PD-1 inhibitor would avoid the need to administer a second agent, and this concept is being evaluated by researchers at the Ningbo Cancer Hospital in China, who recently listed an EGFR-CAR-T trial in which the CAR-T cells have been modified to secrete an anti-PD-1 antibody, NCT02873390. In the United States, the use of clustered regularly interspaced short palindromic repeats (CRISPR) to delete PD-1 in T cells is scheduled to enter the clinic in 2017, in a project funded by the Parker Foundation through U Penn.

Chimeric receptors – redirecting negative signals from the TME

Several scientists have created chimeric receptors that combine the extracellular domain of a receptor to a ligand that is commonly found in the tumor microenvironment (TME) to the intracellular signaling domain of a receptor that has relevant biology to the cell.

While the options are perhaps endless, recently published examples come from the City of Hope and U Penn/NVS groups, where the PD-1 receptor extracellular domain is chimerized to CD28 intracellular domain (PD1CD28), resulting in a stimulation of T cell activity, rather than inhibition of T cell activity following binding of the PD1CD28 on the T cell to PD-L1 or PD-L2 on the tumor.

In the U Penn data, Liu *et al.*, Cancer Res; 76(6) March 15, 2016, the PD1CD28 DNA is inserted upstream of a 41BB/CD3ζ CAR (BBZ) that included either an scFv for the mesothelioma target SS1 (SS1BBZ), or for prostate cancer, PC3 (PC3BBZ). In pre-clinical studies, U Penn has shown that the SS1BBz/PD1CS28 T cell product was considerably more effective than the combination of an SS1BBz CAR-T with an anti-PD1 monoclonal antibody. In animal models, exposure of the SS1BBz/PD1CD28 product to tumor led to an increase in PD-L1 expression on the tumor surface, reflecting induction of both PD1 and PD1/CD28 on the CAR-T. The researchers theorize that the BBZ/PD1/CD28 receptor sequesters PD ligands, preventing them from binding to the native PD receptor, leading to further upregulation of PD-L1. Compared to non PD1/CD28 products, the SS1 or PC3BBz/PD1CD28 products exhibited both a faster onset of tumor regression and greater long-term control, reflecting an increase in peak T cell numbers, greater specific activity per cell, and greater persistence of activity. These phenodynamic changes reflect, in part, a difference in expression of other checkpoint inhibitors. In the presence of the anti-PD1 monoclonal, KEYTRUDA, CAR-T showed upregulation of LAG3, TIM3, and CEACAM1; however, this upregulation did not occur with the PD1CD28 CAR-T products. The Beijing Sanbo Brain hospital in China recently listed a chimeric switch receptor trial that uses PD-1 fused to a CD28 CAR, leading to T cell activation in the presence of PD-L1. In clinical trial NCT02937844, Dr. Lin and colleagues plan to treat 20 glioblastoma (GBM) patients using CAR-T doses ranging from $5x10^4$/kg to $1x10^7$/Kg, given as a 10%/30%/60% split dose following 3 days of fludarabine 25mg/m² and cyclophosphamide 250mg/m².

Coping with reactive oxygen species (ROS)

The tumor microenvironment leads to an increase in reactive oxygen species (ROS), which is associated with tumor promotion and impaired antitumor activity of T cells and other immune cells. Adding a catalase function to the engineered T-cell would allow metabolism of the ROS, hydrogen peroxide, and in 2015, Lightenberg and colleagues presented data for co-expression of catalase with a CAR, in a so-called CAT-CAR, J. Immunol. 196, 759–766. Compared to control CAR-T, CAT-CAR-T exhibited reduced oxidative stress with less ROS accumulation in both the basal state and upon activation, while maintaining their antitumor activity despite high hydrogen peroxide levels. Note that the CAT-CAR-T cells were able exert bystander protection of T cells that were not transfected.

Short circuiting the tumor microenvironment, TME

The tumor microenvironment is highly complex and heterogeneous, and three major strategies should be considered in seeking to address this environment in the context of adoptive cellular therapies (ACT) and CAR-T therapeutics in particular.

- In solid tumors, non-malignant cells in the tumor microenvironment contribute immunosuppressive factors such as prostaglandin E2 and adenosine in response to hypoxia. These factors bind to specific receptors on T cells, leading to dysfunction. While PGE2 and adenosine bind to different receptors, signaling occurs through a common pathway that is amenable to modulation. One strategy that is being evaluated at U Penn and other institutions is the **co-expression of regulator subunit I anchoring disruptor (RIAD), which selectively uncouples PGE2/adenosine-mediated inhibition of T cell function**. Co-expressing RIAD with a mesothelin CAR led to greater tumor suppression in a mesothelin expressing tumor model than a mesothelin CAR alone, Albelda *et al.*, Cancer Immunol Res. 2016 Apr 4.

- CD40 ligand is expressed on a broad array of immune cells, and with its cognate receptor, CD40 allows for autocrine and paracrine signaling between immune cells. On activated T cells, CD40L is expressed within 6 hours of activation and declines within 12-24 hours; clearly, **one strategy to extend the duration of CD40 signaling by CAR-T is to co-express CD40L with the CAR**. Curran and colleagues have reported on the co-expression of CD40L with a CD19 CAR, noting that this approach was successful in a CD19+ve tumor cell line that was resistant to a CD19 CAR-T, Mol Ther. 2015 Apr; 23(4): 769–778. The precise mechanisms for the reversal of resistance are not clear, but Curran and colleagues note that recombinant CD40L expression on T cells that do not express a CAR results in a decrease in PD1 expression and upregulation of the IL-21 receptor on the T cell, increased IL-12 secretion by dendritic cells, and increased expression of Class I HLA on CD40+ve tumor cells, all of which are supportive of a more functional immune response.

- Another TME-centric approach is to target the cancer-associated fibroblasts and specifically, fibroblast activation protein (FAP), which is expressed by more than 90% of cancer-associated fibroblasts in epithelial tumors, but is not expressed in benign tumors or normal quiescent adult tissues. U Penn, Baylor, and the NCI have all developed CARs targeting FAP. In the case of NCI, toxicity was noted due to cross reactivity with mesenchymal stem cell-like cells in bone marrow and muscle, and either genetic knockout, or use of the NCI CAR led to anemia, bone marrow hyperplasia, and cachexia.

- Low oxygen is commonly found in the TME, and CLLS has described a CAR activation system that incorporates a hypoxia inducible factor alpha (HIF-1a) domain. In the presence of low oxygen, the HIF1α motif is activated and the CAR then becomes active. In normoxic conditions the HIF1α motif is degraded, as is the CAR. CLLS believes that incorporating a HIF motif may reduce unwanted off-target toxicity.

- Recently, NCI has identified potassium as an immuno-inhibitory agent in the TME. In the low oxygen environment, necrosis leads to a release of intracellular potassium, which at high concentrations, impairs TCR signaling by T cells. In its research, over-expression of a potassium channel improves potassium efflux and increases in vitro and in-vivo T cell effector function, which in melanoma-bearing mice, led to improved tumor clearance and enhanced survival, Eli *et al.*, Nature. 2016 Sep 14.

- Replacing missing inhibitors was the focus of a recently publication from MSKCC, Boice *et al.* Cell. 2016 Sep 26. pii: S0092-8674(16)31083-2. Noting that germinal center lymphoma often displays HVEM resulting in B cell proliferation, HVEM-deficient lymphoma B cells also induce a tumor-supportive microenvironment marked by exacerbated lymphoid stromal activation. Administration of soluble HVEM binds to its cognate receptor on B cells, B and T lymphocyte attenuator, and BTLA restoring tumor suppression. MSKCC engineered a CD19 CAR-T to also continuously secrete soluble HVEM, noting enhanced therapeutic efficacy in xenograft models. MSKCC makes note of the concept of mini pharmacies for CAR-T engineered to secrete relevant proteins.

Other ways to increase T-cell Receptor (TCR) avidity

While increasing TCR affinity is a key strategy for eTCR approaches, modulating T cell fate after activation may also offer a strategy to increase T cell function. Recently published research from NIH identifies another pathway for controlling TCR avidity. Palmer and co-workers note that following stimulation of the TCR, a member of the suppressor of cytokine signaling (SOCS) family, Cish, binds to a component of the TCR signaling apparatus, leading it to be degraded by the proteasome. The authors conclude that Cish is a novel target that regulates the functional avidity of tumor-specific CD8+ve T cells and can be manipulated to improve adoptive cancer immunotherapy, Palmer *et al.,* J Exp Med. 2015 Nov 16;212(12):2095-113.

Focusing on Carbohydrates and Proteins as ACT Targets

Most of the focus of antigen discovery efforts has been for protein antigens; however, carbohydrates or carbohydrate lipid complexes such as gangliosides can also be targets, and clinical trials are ongoing against the ganglioside, GD2, and the ectodomain of mucin 16 (JCAR020). Carbohydrates can be fundamental to the cancer phenotype as they modulate cell adhesion and motility, which is critical to metastasis.

U Penn recently published data targeting carbohydrate structures that are found only on cancer cells that are associated with cell motility, cell adhesion, and metastatic potential. Specifically, Posey and colleagues, Immunity Volume 44, Issue 6, p1444–1454, 21 June 2016, have reported pre-clinical data for a CAR to the Tn glycoform of Mucin 1. In the absence of cancer, antibodies to Tn prevent expression; however, in the setting of cancer, immunological tolerance to Tn occurs. U Penn had previously shown that a recombinant antibody to Tn was safe in pre-clinical studies and leveraging this into U Penn's CAR platform was able to show efficacy in Tn+ve animal models of pancreatic cancer and leukemia. U Penn believes antigens such as Tn MUC1 represent neoantigens that can be targeted across several tumors.

Off the shelf cells for ACT

The need to manufacture patient-specific products delays the use of ACT, which has led to some patients not being able to receive product due to disease progression occurring during manufacturing. Delay in manufacturing also adds costs and complexity given the need to ship product to and from centralized manufacturing facilities. The concept of an off-the-shelf product using donor-derived T cells is attractive since it avoids the need for a patient-specific product and product can be stored at the site of use, ready for use without delay**. However, there are two key issues that need to be addressed since the T cells used are derived from a donor:**

- **Prevention of the donor cells from recognizing the recipient's healthy tissue as foreign** and thus, avoiding graft versus host disease (GvHD) is critical to safety and currently, the favored way to achieve this is to disable the native TCR, preventing it from interacting with HLA in the host. CLLS is the first company to move a native TCR-deficient ACT product into the clinic, UCART19, using its transcription activator-like effector nuclease (TALEN). JUNO and EDIT have reported on the use of CRISPR and CRISPR associated protein 9 to achieve the same end. While these companies have deleted components of the TCR, BLUE has described an elegant technique combining its megaTAL nuclease to disrupt the TCR with an AAV template for insertion of a CD19 CAR at the same site, in effect, killing 2 birds with 1 stone, i.e., expression of the CAR at the genomic site of the native TCR such that transfected T cells do not express a TCR, but express a CAR. Obviously, disruption of the donor TCR removes the possibility of a positive graft versus leukemia effect; however, the assumption of ACT companies is that the engineered CAR/TCR is a more targeted and effective strategy than relying on luck that one of the grafted T cells actually recognizes the tumor.

- **Preventing the host from recognizing the donor T cells as foreign, leading to their rejection prior to clearance of tumor.** This is more challenging given the need to modulate the donor T cell's expression of HLA in a way that prevents clearance by host T cells and NK cells. Currently, the lymphodepletion regimen used for ACT provides a window of around 30 days for host cells to avoid detection by host immune cells.

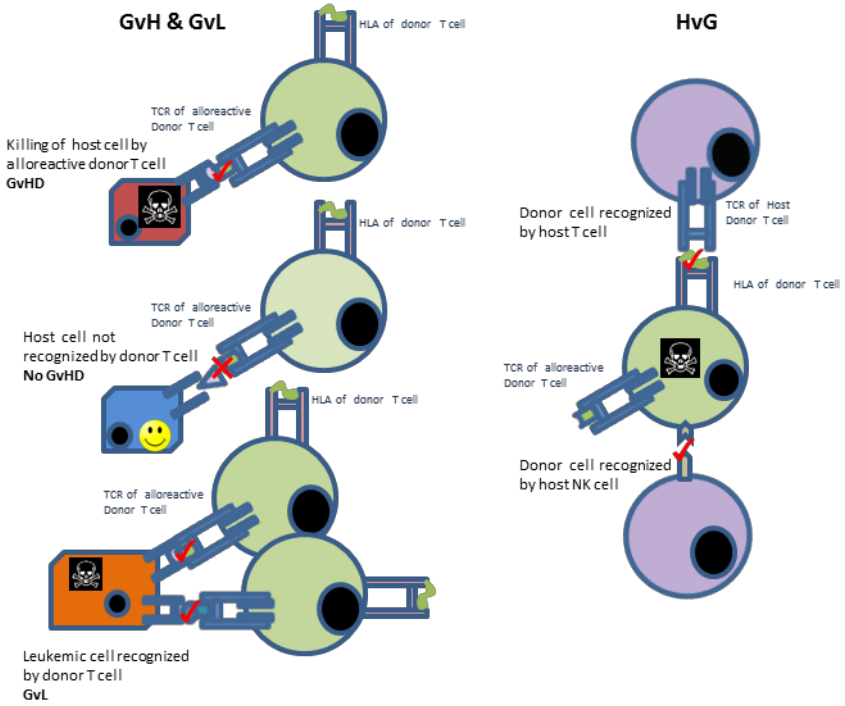
A different concept of off-the-shelf retains the individualized patient format, but invokes the need to manufacture product ahead of its scheduled use so that the product is ready if required. We understand that Lion Bio (LBIO) is interested in such an approach. The TIL process requires two stages for manufacture of product over 4-6 weeks, pre-rapid expansion protocol (REP), and REP and MDACC is banking partly manufactured products at the end of the pre-REP for patients who may benefit from TIL.

Graft versus Host Disease and Host versus Grafted reactions in the allogeneic setting

Allogeneic donor T cells can recognize both the host and the tumor, and this leads to an apparent paradox between the need to prevent the former, while encouraging the latter. As the transplant community has learned through trial and error, more rigorous removal of T cells from a graft reduces the risk for GvHD, but increases the risk for infection and disease relapse.

Exhibit 20 summarizes possible interactions between donor T cells in ACT graft and the host based on the native TCR and HLA, i.e., outside of an engineered TCR or CAR. In the top left of Exhibit 20, the allogeneic T cells recognize host healthy tissue as non-self, leading to donor T cell activation, resulting in tissue damage and acute GvHD. If the donor shows a perfect HLA match to the recipient, the donor T cells do not recognize the healthy tissue as non-self and remain inactivate; however, as noted in the bottom left panel, the goal of an allo-transplant is to provide donor T cells that recognize the cancer as non-self. In addition to donor T cells potentially recognizing the recipient as foreign, the reverse is also true, where the recipient can recognize the incoming donor T cells as foreign and remove them, as noted in the right panel of Exhibit 20. Lymphodepletion of the host prior to receipt of donor T cells provides a window for the donor T cells in the absence of an effective recipient immune system; however, as the recipient's immune system recovers, both T cells and NK cells have an opportunity to recognize and remove the donor T cells.

Exhibit 20. Balancing graft versus host, graft versus leukemia and host versus graft



Source: Wells Fargo Securities, LLC GvH – graft versus host, GvL – graft versus leukemia, HvG – host versus graft

In considering the challenges of GvHD, one strategy that has been adopted is to eliminate T cells with an α/β TCR from the graft, while leaving behind NK cells and γδT cells, which provide coverage against infection, although long-term control of disease remains a potential issue with removal of α/β TCR T cells. An alternate approach is genetic removal or silencing of the α/β TCR to prevent T cell targeting of healthy tissue, in the context of adding a CAR/eTCR to ensure tumor targeting. Several academics have developed T cells lacking a TCR, but first in the clinic is Cellectis (CLLS). At the 2015 American Society of Hematology (ASH) meeting, Cellectis (CLLS) presented data from a single patient treated with UCART19, a CD19 CAR-T that has the TCRα chain gene knocked out. In theory, because of the TCRα knockout, UCART19 T cells do not express a functional TCR and are unable to cause GvHD. An alternative approach to an off-the-shelf product is to use a cell type

that is less likely to cause damage, such as an NK or natural killer T (NKT) cell since these cells do not harbor an α/β TCR.

Avoiding a host versus graft response is also challenging and as CAR-T, as well as UCART T cells, express Class I HLA, they are subject to immune surveillance by both host T cells and innate immune cell such as NK cells. However, in the current paradigm for the use of lymphodepleting chemotherapy prior to receipt of ACT, host immune cells are removed. The role of lymphodepleting chemotherapy is primarily designed to eliminate competition for resources, but it also delays the ability of the host's immune system to target the graft. Lymphodepleting chemotherapy is not without its own side effects, and more subtle and targeted ways to control host immune response are being investigated. CLLS has also engineered UCART cells to be immune to specific agents that could be used both to treat the target disease, as well as delay host immune recovery. In addition to loss of the TCRα, CD52 has been knocked out in UCART19. CD52 is recognized by the monoclonal antibody CAMPATH, but without cell surface CD52, UCART19 is immune to CAMPATH. Similarly, CLLS has shown data for inactivation of deoxycytidine kinase to provide protection from fludarabine clofarabine or cytarabine, and removal of the glucocorticoid receptor provides protection against steroids.

Developers of eTCR products are also pursuing off-the-shelf strategies and in 4Q15, eTCR leader Adaptimmune (ADAP) entered into a collaboration with Universal Cells aimed at developing an off-the-shelf T cell for eTCR. Universal Cells has developed strategies to remove Class 1 and Class II HLA on the T cell, while adding HLAE to provide a protective signal against NK cell attack. For conserved antigens such as the cancer testis antigen NY-ESO-1, a Universal Cells T cell would allow ADAP to develop an off-the-shelf version of the current patient-specific NY-ESO1 SPEAR. Use of a Universal Cells product would still be subject to HLA restriction, however, as each ADAP TCR is specific for only one HLA haplotype.

Gene editing and nuclease Soup

While a detailed review of gene editing is beyond the scope of this review, it is relevant to note that there are several technologies gene editing that include the following:

- Zinc finger nucleases – Sangamo (SGMO)
- Transcription activator-like effector nucleases (TALEN) – Cellectis (CLLS)
- megaTALs – combining in a single molecule a TALEN with homing endonuclease – Bluebird (BLUE)
- Clustered, regularly interspaced short palindromic repeats (CRISPR) with CRISPR associated protein 9, (Cas9) – JUNO, NVS

Each of these technologies is a variation on a theme; but basically, an enzyme that cuts DNA is targeted by a DNA-binding molecule that recognizes a specific DNA sequence. Once both strands of DNA have been cut, non-homologous end joining (NHEJ) tries to repair the DNA, but this is an error-prone procedure and the end result is often gene disruption. Key to success is avoidance of off-target activity in order to reduce non-desired gene disruption. Zinc-finger nuclease has been validated by Sangamo (SGMO) as a way to knock out the HIV co-receptor, CCR5 in CD4 T cells. Each zinc finger motif binds to 3 nucleotides and for specificity, 3-6 motifs are used to bind to each strand. The Zn finger proteins are bound to the Fok1 nuclease, which is only activated after DNA binding allows dimerization.

TALENS utilize a DNA-binding protein armed with a nuclease to achieve double-stranded DNA breaks at pre-specified locations in a gene. The DNA-binding protein, or TALE, is comprised of 33 amino acids where specificity for a specific binding site is defined by a pair of amino acids. CLLS has interrogated the binding of amino-acid pairs to each of the 4 nucleotides that comprise DNA to develop designer TALEs that target genes of interest. As with the Zinc finger nuclease approach, two different TALEs recognize different strands of the DNA and dimerization of a nuclease Fok1 results in precise double-strand DNA cleavage. The megaTALs combine a TALE with a different class of DNA active enzyme, the homing endonuclease. MegaTALs offer the option to specifically target a single strand of the DNA, allowing removal of bases that contain, for example, a mutation. BLUE and Precision Biotechnology have developed megaTAL-based technologies.

The three systems described above use proteins to target specific DNA sequences, CRISPR-Cas9 uses a guide RNA to target a DNA endonuclease to the desired location of DNA. The ability to use RNA as a guide gives the CRISPR system greater flexibility, but specificity requires improvement before it matches that of other systems.

U Penn has published on the potential use of CRISPR for generating universal CAR-T cells, Ren *et al.*, Clin Cancer Res. 2016 Nov 4. Ren and colleagues used multiplexed electroporation of CAS9/guide RNAs to disrupt

endogenous TCR, beta-2-microglobulin (B2M), to disrupt Class I HLA and PD-1 combined with lentiviral transfection with a prostate stem cell antigen (PSCA) or CD19 CAR. U Penn notes that CRISPR can efficiently disrupt the TCR, especially with sequential electroporation. However, following single or double electroporation, a 12-fold and 8-fold increase in cell number was observed over 9 days, which represents at least a 70% reduction in expansion observed for non-electroporated T cells. Transfection of a CD19 CAR into the TCR-depleted T cells suggested that no decrease in T cell effector function was observed following TCR deletion. Eliminating HLA in addition to TCR resulted in a double negative population efficiency of 80% and when co-electroporated, the efficiency was 65%. Positive selection of CD3 allowed purification of CD3-ve T cells. Challenging the TCR/HLA^{NEG} T cells with HLA-matched tumor cells showed no recognition of the tumor cells; however, alloreactivity was observed against PBMC, but not purified T cells, suggesting that NK cells in the PBMC were able to recognize the TCR/HLA^{NEG} T cells as foreign. There was also an increase in HLA Class II expression by activated T cells, suggesting that host cell-mediated rejection could occur despite loss of HLA Class I. In an irradiated animal model, 4/5 mice developed lethal GvHD within 2 months of infusion of non-manipulated T cells. In contrast, none of the mice receiving TCR^{NEG} T cells or TCR/HLA^{NEG} T cells developed GvHD. Adding a CD19 CAR to TCR^{NEG} T cells or TCR/HLA^{NEG} T cells followed by cell number expansion resulted in a product that U Penn describes as consistent with a central memory phenotype. U Penn notes that similar overall survival rates in the setting of a CD19+ve tumor, nalm6 were observed between manipulated CD19-CAR-T and TCR^{NEG} or TCR/HLA^{NEG} CD19 CAR-T cells.

The benefit of PD-1 abrogation was evaluated using a prostate stem cell antigen (PSCA) CAR where, in the setting of a prostate tumor model, the PD-1^{NEG} CAR-T cells significantly enhanced the anti-tumor activity versus a control PSCA CAR-T. Notably, more than 90% of the prostate tumors gained PD-L1 expression after PSCA CAR-T treatment. In the setting of a TCR/HLA^{NEG} CD19 CAR-T disruption of PD-1 produced a more robust response than PD-1 wild type CD19 CAR-T in the setting of a nalm6-PDL1 model. Triple disruption of TCR/HLA and PD-1 could be achieved in 65% of cells.

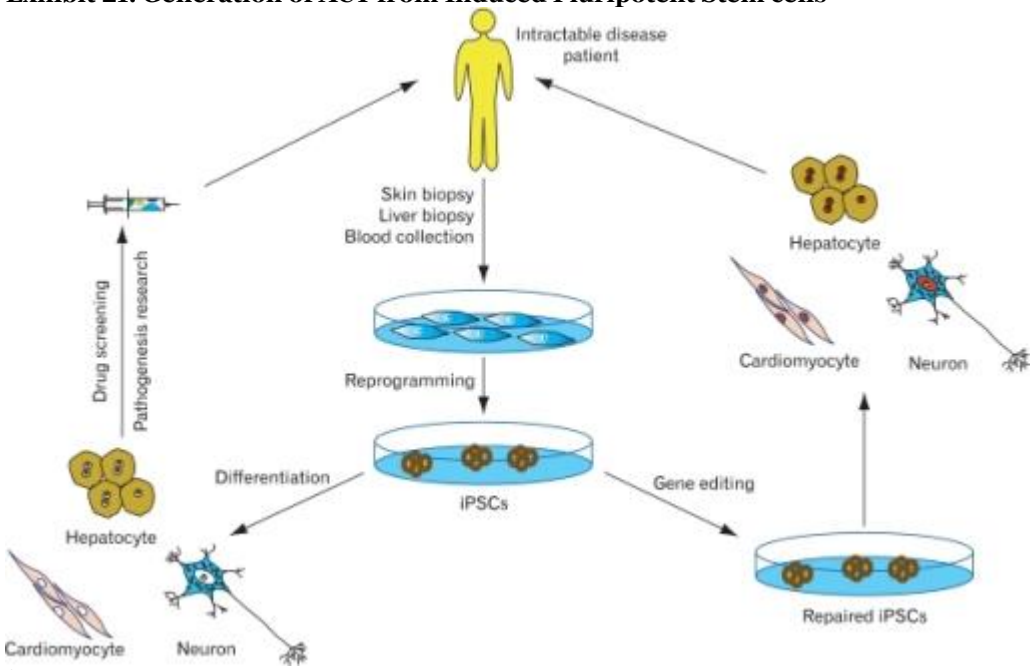
At the June 2016 recombinant DNA advisory committee (RAC), the Parker Foundation, a non-profit organization funding research at U Penn, MD Anderson and UCSF presented the first clinical trial proposal for use of CRISPR-CAS9. The trial is to use electroporation of multiplex CRISPR to remove the TCRα/β chains and PD-1 from autologous T cells, which then receive U Penn's NY-ESO-1 TCR (NYCE cells), reviewed in more detail further on herein. In pre-clinical studies using multiplex CRISPR, U Penn estimates that approximately 85% of T cells lose their endogenous TCR and 50-80% loose PD-1. The NYCE proposal is very ambitious and, as noted by U Penn during the RAC, the manufacturing protocol is to produce up to 16 different T cell clones. U Penn proposes to dose patients with an unselected product on the assumption that a dominant clone will emerge, which the researchers assume will exhibit loss of endogenous TCRα/β chains and PD-1, and expresses the NY-ESO-1 TCR. U Penn believes that the expression level of NY-ESO-1 TCR is higher in the TCRα/β knock out T cells than in unmodified T cells and, taking this into account, U Penn is proposing a starting dose 1/10th of that used in the original NCI NY-ESO1 TCR protocol.

The first clinical evaluation of a CRISPR modified T cells is to be undertaken by Cell Biotech Co in China. NCT102867332 describes a randomized trial of autologous T cells, with PD-1 knockout via CRISPR Cas9 in 20 renal cell carcinoma patients. Patients in the experimental arm are to receive 2x10^{7} PD-1 knockout cells/Kg fractionated over 3 days (20% day 1, 30% day 2, and 50% day 3) following a single dose of cyclophosphamide, 20mg/kg day -3. Patients are to receive 2, 3, or 4 cycles, with IL-2 support following each cycle.

Induced Pluripotent Stem Cells, iPSC

By introducing all of the desired genetic changes into a single stem cell, a homogeneous and limitless population of functional T cells can be produced. As depicted in the cartoon in Exhibit 21, differentiated cells from a donor are reprogrammed into stem cells – induced pluripotent stem cells (iPSC). These can be differentiated into tissue-specific progenitors for tissue repair as an example or of more relevance to ACT, engineered at a single cell level to achieve the desired final product characteristics. FATE is a leader in the field of iPSC, and in its view, these individually engineered cells can be differentiated into progenitors of effector immune cells and stored in a traditional master and working cell bank format to ensure an inexhaustible supply of cells that can be further engineered for specific use.

Exhibit 21. Generation of ACT from Induced Pluripotent Stem cells



Source NIH.gov

As currently practiced, modification of bulk cells with viruses carrying CARs or eTCR is an uncontrolled event since vector insertion into the genome occurs randomly. In the earliest trials, secondary cancers have resulted from this random insertion, and while modern vectors are safer, regulators have demanded 15-year follow-up of all patients treated with gene therapy to allow for collection of late-emerging adverse. A targeted nuclease approach to ACT gene insertion allows for precise control over the site of gene insertion, but the need to serially modify T bulk cell to achieve the desired phenotype/genotype reduces efficiency and increases manufacturing the time line.

A more controlled approach to generation of a controlled ACT product was recently described by Sadelain and colleagues at the Memorial Sloan Kettering Cancer Center. Sadelain described the development of a T cell-derived iPSC using 4 transcription factors including OCT4, a technology that is controlled by FATE, and in 3Q16, FATE and MSKCC entered into a partnership to develop off-the-shelf iPSC-derived T cell therapies. The MSK iPSC derived T cells actually appear to be much more like a γδ T cell since they are either double negative for CD4 and CD8, or just possess a CD8α chain despite expressing the β chain of the TCR. MSK has used these iPSC-derived T cells as a host for a CD19 CAR and also described elimination of the T cell receptor, to avoid GvHD and HLA loci to avoid host versus T cell response, Themeli *et al.,* Nature Biotechnology, 31, 928-33, 2013. By using an iPSC, Sadelain and colleagues was able to use a nuclease strategy to precisely insert the CAR into the desired location prior to cell expansion; recall that viral insertion of the CAR is random. At the 2016 American Association of Cancer Research meeting, Sadelain and colleagues presented comparative data for an iPSC CD19 CD28 (19/28) costimulated CAR-T versus their standard retrovirally produced 19/28 CAR-T. The expression level of the CAR on the iPSC was lower than observed from the retroviral CAR, but CAR expression on the iPSC T cell was more homogeneous across the product. Notably, the efficacy of the iPSC CAR-T was significantly superior to the retroviral CAR-T, presumably due to more stable CAR expression from the selected genomic site.

Generation of iPSC from melanoma TIL was recently described by Saito and colleagues, Stem Cells Int. 2016;2016:8394960. Saito used a Sendai-virus to transduce melanoma TIL with 4 transcription factors, including OCT4 leading to the production of iPSC that displayed stem cell-like morphology, stem cell-specific surface antigens, and pluripotency. Saito notes that T cells differentiated from melanoma-TIL iPSC, retained the same TCR repertoire as the original TIL, leading Saito to note that iPSC-derived TIL could obviate the problems associated with poor persistence with TIL given the self-renewal capability of iPSC. As Saito and colleagues note, there are several challenges to deriving iPSC from TIL, including an approximate 2-month time line to derive iPSC, reprogramming efficiency, and the patient-specific nature of such a product, although Saito notes that the iPSC could be used on an HLA-matched patient; however, this would only be relevant for TCRs that do not target patient-specific neoantigens.

In addition to FATE's collaboration with MSKCC for iPSC T cell therapies, the company has a longer standing Collaboration with the University of Minnesota of iPSC-derived NK cell therapies; the iNK collaboration was recently expanded to cover the intellectual property for a non-cleavable CD16 and chimeric antigen receptors. The first iNK product is to incorporate a non-cleavable CD16, which FATE has shown in pre-clinical models. It endows these iNK with superior antibody-dependent cellular cytotoxicity characteristics when the CD16 iNk are compared to conventional NK cells mixed with antibodies such as ERBITUX, RITUXAN, or HERCEPTIN. FATE intends to seek regulatory guidance in 2017 for moving the iNK program into the clinic. KITE is also developing off-the-shelf allogeneic T-cell therapies from renewable pluripotent stem cells through a collaboration with the University of California, Los Angeles (UCLA). Specifically, scientists at UCLA have developed an artificial thymic organoid cell culture system that replicates the human thyroid and can be used to differentiate T cells from iPSC in a reproducible and consistent manner.

Non-viral transfection and patient-specific neoantigens

Use of viruses to modify bulk populations of cells with patient specific neoantigens is likely not feasible in the current regulatory environment and non-viral gene transfer technologies are likely needed. The majority of CAR and eTCR products rely on the use of a virus to infect target cells with the desired DNA, which then integrates randomly into the genome of the cell. This is a complex multi-step process that requires the development in essence of a separate biological product, the virus with all of the attendant costs and controls of manufacturing. In the case where a single virus stock is to be used on many patients, the costs can be diluted; but for a patient-specific neoantigen approach, the cost and 6-month plus time line are prohibitive, in our view.

Detailed analyses of cancer cell genomes have revealed a spectrum of somatic mutations, i.e., specific to the tumor, with some tumors being highly mutated, while others having a lower mutation burden. One of the most highly mutated tumors is cutaneous melanoma, and sequencing of melanoma tumor neoantigens presented on the tumor by HLA reveals that while certain proteins are frequently mutated, each tumor present unique peptide sequences and each patient has a unique neoantigen profile. On the assumption that the most effective way to target a tumor is with an engineered T cell that potently recognizes a tumor-specific antigen, each patient must be treated with a unique product. Steve Rosenberg at the National Cancer Institute has undertaken such a task and the time of neoantigen identification, to sequencing, to product infusion was approximately 8 months. The primary reason for the length of development time was manufacture of the viral vector encoding the neoantigen eTCR. Currently the workhorses for CAR and eTCR products are two members of the retroviridae - gamma retrovirus and lentivirus. Both are RNA viruses and both have the ability to retrotranscribe RNA into circular DNA, which can then be stably integrated into the genome of the host cell. In ACT applications, these viruses do not contain viral RNA; rather, they contain RNA encoding the desired CAR or TCR. These viruses are manufactured by a packaging cell line that in the case of lentiviruses, is transfected with 2 or 3 plasmids containing RNA for the viral capsid packaging proteins and the ACT plasmid. Because the packaging plasmids can be maintained in a packaging cell line suitable for retroviruses, retroviral transfection requires just the ACT plasmid. The major difference between lentiviruses and retroviruses is that for the retroviral nucleic acid to enter the cell's nucleus, the nuclear membrane must be disrupted as part of the mitotic cell division process, while in lentiviruses, the pre-integration complex can be transported across an intact nuclear membrane. Put simply, retroviruses can only replicate in a dividing cell, while lentiviruses can replicate in both quiescent and dividing cells. As their name suggests, non-viral transfection systems avoid the need to manufacture a virus, and therefore, offer time saving. This may be the critical enabling step for delivery of neoantigen-specific therapies in a timely manner. ZIOP is using the sleeping beauty (SB) system that uses a fish-derived transposon and transposase to achieve integration of ACT DNA into the host genome.

Researchers from the NCI and ZIOP recently published on the use of SB to stably transform T cells to express patient-derived tumor neoantigen-specific TCRs, Deninger *et al.,* Molecular Therapy accepted article preview online 05 March 2016. The researchers built 3 unique mutation-reactive TCRs containing clinical-grade plasmids using murinized TCR (mTCR) αβ chain sequences that allowed for selection of cells that had been successfully transposed, resulting in expressed mTCR protein. The researchers co-electroporated the SB11 transposase and TCR containing transposons into peripheral blood lymphocytes from cancer patients. TCR expression could be detected on the following day, and these cells could be captured for enrichment by using an anti-murine B chain antibody. This antibody step enriched mTCR +ve cells from approximately 12% to 80%. Following selection, the mTCR+ve T cells were stimulated and the subject to a REP protocol. At the end of REP, day 22 stable expression of mTCR could be detected; however, the level of mTCR+ve T cells was lower than observed post enrichment, likely reflecting the fact that at enrichment both stably integrated and non-stably integrated, i.e., episomal mTCR expression was observed. While the dominant T cell phenotype at the time of electroporation was CD4, CD8+ve T cells dominated after REP likely due to the cytokine mix used to support the REP. The SB electroporation step itself resulted in an increase of CD45RA+ve cells, with a concomitant decrease in CD45RO+ve cells indicating a selection of naïve, rather than memory cells, and expression of these and other T cell markers remained relatively stable during REP. For each of the three

different TCR products, the final product comprised CD4+ve effector and memory T cells, and CD8+ve effector and minimally differentiated T cells. This latter population has been associated with the highest level of efficacy in models of ACT. Note that experience with TIL suggests a correlation between efficacy and CD27, and the majority of SB mTCR cells expressed CD27. CD27 is a co-stimulatory receptor that supports antigen-specific expansion of naive, but not effector T cells, and further CD27 is critical for the development of immunologic memory. The relative expression of the different mTCRs differed, likely reflecting the human variable regions since the murine regions were constant; and while this may affect efficacy, the authors noted that specific activity of the SB mTCRs was similar. The avidity of the SB transposed mTCRs was in the nM range, similar to that reported for virally transduced TCRs, and their avidity was similar to those displayed in the original TIL from which they were cloned. In functional assays, the researchers concluded that TCR-transposed T cells were highly functional and specific for mutated neoantigen and autologous tumor cells, as evidenced by the following:

- Surface expression of 41BB and other activation markers
- Secretion of cardinal proteins indicative of activated T cells such as IFNγ,
- Simultaneous production of multiple effector molecules, and
- Ability to lyse target cells

In early 2017, ZIOP/XON and the NCI announced a Cooperative Research and Development Agreement (CRADA) to formally develop and evaluate neoantigen-specific TCR therapies in patients with solid tumors using the aforementioned Deninger publication as a roadmap. By YE17, ZIOP intends to begin testing a point-of-care neoantigen-specific TCR therapy.

Sleeping Beauty is not the only non-viral transfection system in use, and at the recent American Society of Hematology meeting, German academics described the development of a CAR to the cluster of differentiation antigen 319 (CD319), also known as signaling lymphocyte activation molecule 7, the target of BMY's EMPLICITI and its intention to manufacture CAR-T in 72 hours using non-viral gene transfer, although details of the technology were not discussed. Australian researchers recently published on the use of the piggyBac non-viral gene transfer system to generate CD19 CAR-T cells, Ramanyake *et al*, Cytotherapy. 2015 Sep;17(9):1251-67. Like sleeping beauty, piggyBac is a transposon-based technology targeting an inverted repeat, AATT sequence for insertion. The piggyBac system was first described in the baculovirus, a virus that infects insect cells. Poseida Therapeutics is using piggyBac as a non-viral system to transfect T cells with CARs.

An alternative approach to non-viral gene transfer is the use of cell membrane disruption as is being pioneered by CellSqueeze (SQZ). By disrupting the cell membrane temporarily, foreign materials can be introduced into the cell, including nucleic acids or tumor antigens. SQZ signed a major agreement with Roche in 2015.

The state of the art for identifying and validating neoantigens involves several steps:

- Tumor is sequenced comparing whole exome and RNA sequences between healthy and tumor tissue to identify mutations;
- Algorithms predict which of these mutated peptides have properties to be displayed by MHC class I;
- Peptides are synthesized; and
- Peptide recognition is confirmed either by patient-derived TIL, or MHC tetramer.

According to Bob Schreiber, such a process at Washington University takes just 24 hours using a mouse tumor. In the context of ACT, the end product of such an analysis is an engineered TCR that recognizes the peptide. Alternatively, the peptides can be optimized for use in a vaccine, and Washington University and the University of Washington are collaborating on a soon to be launched pancreatic cancer trial using a patient-specific vaccine combined with a mesothelin eTCR. Schreiber is a founder of Neon Therapeutics, which is focused on patient-specific neoantigen approaches.

An alternative approach to identifying tumor-specific peptides was developed at PhosImmune, recently acquired by Agenus (AGEN). PhosImmune catalogued peptides that were aberrantly phosphorylated, presented by HLA and recognized by high-affinity TCRs that did not recognize the correctly phosphorylated peptide on normal healthy cells. Unlike the neoantigens, which are patient specific, the same aberrantly phosphorylated peptide can be found in the same tumor across multiple patients, suggesting that an HLA-matched off-the-shelf product may be possible. Separately, AGEN recently initiated phase 1 testing of a patient-specific neoantigen peptide vaccine. From tumor biopsy to vaccine dosing currently takes 8 weeks.

ACT Beyond αβT cells

As noted, the presence of a native αβTCR in T cells that have been used for ACT ensures that the necessary signaling apparatus is present for the introduced eTCR; but for off-the-shelf approaches, the native αβTCR can cause GvHD leading scientists to consider using immune cells other than αβ T cells. Immune cells such as NK and gamma delta, (γδ) T cells harbor receptors that in part, signal through CD3ζ,, which is a basic signaling component of CAR architecture, suggesting that these cells may be alternative hosts for CAR/eTCRs. CD3ζ CARs readily link to endogenous signaling pathways in NK cells and trigger cytolytic activity; however, NK cells have efficient antiviral defense mechanisms and so, use of retro or lentiviruses for CAR transduction is less efficient in NK cells than in T cells. In addition, culturing of NK cells to large numbers is not as reliable as culturing T cells and considerable interpatient variability exists and finally, cryopreservation is currently not feasible for an NK cell product.

The NK-92 cell line, originally derived from a patient with an NK lymphoma, offers several advantages over autologous NK cells. It has been approved for use in clinical trials and can be grown to large numbers reliably; and unlike NK cells, this cell line is readily transfected with virus. NantKwest is developing NK-92-based products, and with sibling-company, NantBioscience, recently entered into a Cooperative Research and Development Agreement with the National Cancer Institute to combine their proprietary recombinant NK technology with monoclonal antibodies for use as immunotherapy. Academic investigators are also developing NK-based CARs including Baylor, which has listed a phase 1 clinical trial for an NK-CAR to target GD2. Recall that as noted in the preceding text, Baylor has completed a GD2 CAR-T trial with a 2nd generation CAR and is conducting a trial with a 3rd generation CD28/OX-40 CAR; the NK CAR is also a 3rd generation CD28/OX40 CAR.

Gamma delta T cells constitute a population of T cells that possess qualities of αβ T cells, as well as NK cells. They arise from double-negative, CD8-, CD4- precursors. While the precise role of the γδ T cell is unclear, they are thought to be part of the first-line defense against pathogens at body surfaces, and to act at the interface between the innate and adaptive branches of the immune system. There are two major subsets defined by different δ chains. V δ1 are predominant in the intestine, where they respond to non-classical HLA molecules induced by stress. The V δ2 cells are predominant in tonsils, skin, and lymph nodes, where they serve as antigen-presenting cells and provide a memory function. The Chinese company, Beijing Doing Biomedical, is conducting a parallel trial with CD19 CARs, one expressed in an αβ T cells, and the other in a γδ T cell. Studies NCT02546739 and NCT02656147 are both phase 1, enrolling children and young adults with advanced B cell malignancies. With enrollment targets of 100 and 48, respectively, these studies may be able to provide insight into the relative merits of the two different T cell hosts for a CD19 CAR.

Convergence of innate and adaptive immunity

In an interesting convergence of adaptive and innate immunity, U Penn has described a novel CAR architecture that combines an scFv for antigen recognition with the cytoplasmic domain of a killer immunoglobulin-like receptor (KIR) from a an NK cell, which, in concert with activation motifs containing adaptor DAP12, forms an active CAR when transfected into T cells. U Penn has shown that a mesothelin-targeted KIR-CAR outperforms a 2nd generation mesothelin 41BB CAR in a mesothelioma model that is highly resistant to immunotherapy. U Penn asserts the superior performance to improved retention of the CAR and improved effector function of isolated tumor-infiltrating lymphocytes, and suggests that KIR-CARs may be particularly suited for ACT in immunotherapy-resistant solid tumors, Wang E *et al*., Cancer Immunol Res. 2015 Jul;3(7):815-26.

Convergence of eTCR and CARs

Because the affinity of antibodies can be an order of magnitude higher than even the most potent eTCRs, another potential development in ACT is the convergence of CAR and eTCR approaches with the development of antibodies that are specific for a peptide presented in the context of HLA. Unlike naturally occurring TCRs, these antibodies can show very high affinity, and this may be important as tumors downregulate HLA. The development of high-affinity antibodies to HLA/peptide complexes is not a trivial task, however, since antibodies typically bind to approximately 800 Angstoms (Å) of ligand and yet only 100-300 Å of a peptide bound by HLA is visible to the antibody; an Å is 0.1nM. An additional problem is posed by the fact that an estimated 65-85% of the peptide/HLA interface is comprised of HLA and thus, presents a problem of specificity since the antibody must only bind to the peptide/HLA and not to HLA alone to avoid off-target toxicity. Perhaps not unsurprisingly, early antibodies targeting peptides in the context of HLA have typically had low affinity, such as the HLA-1*1 –MAGE-A1 antibody described by Chames *et al.* in 2000, which had an affinity of 250nM. Recently, Cheung and colleagues at MSKCC described the development of an scFv that could bind to a specific Wilm's tumor antigen in the context of Class I HLA with an affinity of 2nM. Fusing the scFv to an antibody Fc region led to efficient NK cell mediated tumor killing in pre-clinical studies, while insertion of the scFv into a CAR allowed for transfection of T cells or NK cells with a potent 41BB-based CAR. The WT1-CAR-T was able to recognize and kill WT1+ve AML and breast cancer cell lines. In contrast, a parental low-affinity WT1/HLA scFv, when incorporated into a CAR, was unable to lyse target cells, Leukemia (2015), 1–10. MSKCC has also published on a TCR-mimic monoclonal antibody, which also binds WT-1, ESK-1 that has been used in a bispecific t cell engager (BiTE) format currently under development at Eureka Therapeutics, Dao *et al.*, Nat Biotechnol. 2015 Oct;33(10):1079-86.

ACT and bispecific antibodies

While ACT leverages the decades of investigation into antibodies, the attachment of antibody-based drugs to a generic linker on a T cell is also being investigated. The vast majority of the estimated 60 bispecific antibody formats in development are designed to redirect an immune cell to a cancer cell in-vivo; thus, AMGN's BLINCYTO redirects T cells via a CD3 binding domain to a CD19+ve cell, while AFMD's AFM13 redirects an NK cell via a CD16a binding domain to a CD30+ve cell. As a passive process, these approaches are beyond the scope of this review; however, Unum Therapeutics is developing CAR-T cells that have been modified with CD16, which binds to the Fc of the antibody, and a phase 1 trial combining the CD16 CAR with RITUXAN is ongoing. Noting the potential for such an approach to activate CAR-T cells by binding naturally occurring antibodies, scientists from the California Institute for Biomedical Research have published on the use of the inclusion of a yeast sequence, referred to as a peptide neo-epitope (PNE), into antibody fragments that bind to a universal switchable CAR-T (sCAR-T) that carries an ScFv CAR recognizing the yeast peptide, Rodgers *et al.*, Proc Natl Acad Sci U S A. 2016 Jan 26;113(4). In this publication, Rodgers and colleagues show that a CD19 sCAR-T plus a PNE-modified CD19 FMC63 antibody fragment has comparable activity in animal models to a standard FMC63-based CAR-T. Rogers notes that unlike a standard CAR-T, the PNE-CD19 can be titrated to precisely dial in the optimal dose and that as an antibody fragment, the half-life is short, suggesting an improved safety profile over existing CAR-T products. In addition, the sCAR-T platform allows for the ability to switch targets in the setting of resistance or presumably allows for multi-target strategies to blunt development of resistance. Rogers and colleagues are developing humanized PNE-CD19 antibody fragments for clinical trials to reduce the risk for antibody development by the host to PNE-CD19, which is to be dosed repeatedly. Of course, the downside to this approach is the need to develop PNE versions of targeting molecules in addition to the sCAR-T. Researchers at Purdue University have also reported on a similar concept for a universal CAR. By developing a small molecule activator that uses a common CAR binding domain and a tumor-specific binding domain, Purdue sees the ability to titrate CAR activity by does escalating the small molecule activator to modulate potential toxicity, but also the possibility of combining or changing small molecule activators to combat development of resistance and or treat heterogeneity.

Ex-vivo T cell arming

Researchers at the Karmanos have developed a strategy to arm autologous T cells with bispecific antibodies ex-vivo and then treat patients with multiple doses of the ACT product; recently the company Transtarget was formed to commercialize the technology. The Karmanos approach involves creation of bispecific antibodies by adding two different linkers to the Fc domains of commercial antibodies, a CD3 antibody for binding to the T cell and a tumor targeting antibody. The linker-derived antibodies are mixed together and, because of the use of two different linkers, only heteroconjugated bispecific antibodies are formed, i.e., a CD3-CD3 bispecific cannot form. Karmanos researchers have used Ortho Biotech's OKt3 as the CD3 donor and have published on the use of HERCEPTIN, RITUXAN, and ERBITUX as the tumor-targeting antibody. The bi-specific antibodies are mixed with autologous cells following apheresis, and administered repeatedly with IL-2 and granulocyte macrophage colony stimulating colony factor (GM-CSF) support; lymphodepleting chemotherapy was only used in the lymphoma setting as part of a transplant. Data for the HER2bi armed activated T cells (aATC) in

metastatic breast cancer were published recently in Clinical Cancer Research, Lum *et al.,* May 15, 2015 21; 2305.

Following apheresis, T cells were expanded with IL-2 and then combined with the bispecific construct at a dose of 50ng/10^6 cells. The trial was designed to test 4 dose levels, 5, 10 , 20, and 40x10^9 HER2bi aATC; however, the 40 x109 cell dose proved unfeasible and the highest technically feasible dose was declared at 20x10^9 cells. Eligible patients with a HER2 score of 0-3 received aATC twice weekly for 4 weeks with IL-2 (300,000 IU/m^2/day) and GM-CSF (250µg/m^2 twice weekly) starting 3 days before the first aATC dose and continuing 1 week after the 8th dose. The trial enrolled 23 patients, 11 (48%) with a HER2 score of 0, 2 each at a score of 1+ or 2+ and 7 with a score of 3+; 1 Her2 score was unknown. All patients except 1 had received prior chemotherapy/hormonal therapy (most ≥3 lines), and all of the HER2+3 patients except 1 received prior HERCEPTIN; most patients (73%) had visceral disease at baseline.

The ACT regimen was generally well tolerated, with grade 4 events rarely observed and grade 3 toxicity limited to chills, occurring in 60-70% of patients and a dose-dependent increase headache (25-75%); notably, one reason the toxicity profile of the Karmanos protocol differs from the CAR-T experience is the use of low-dose IL-2, which at approximately 7000 IU/Kg, is about 1,000-fold lower than the 750,000 IU/Kg used at NCI, for example. One patient at the 20 x109 aACT cell dose suffered a grade 4 headache, grade 3 hypertension and was removed from the study after the 3rd infusion. The patient suffered a subdural hematoma. Three additional patients were added without a DLT. One patient died of digoxin-related toxicity and upon autopsy, there were no myocardial T cell infiltrates. Several cytokine "flurries" were observed, but there were no cases of life-threatening cytokine storm and only 3 patients were hospitalized for cell-based toxicities. All toxicities resolved and all patients completed their scheduled doses. While OKT3 is a mouse antibody, no significant anti-mouse antibodies were observed following aACT.

Immune evaluation including target cytotoxicity was performed 4 times during the first 5 months and tumor evaluation was performed at week 14.5. At week 14.5, 1 patient had a partial response (PR), 11 stable disease (SD), and 9 progressive disease (PD); two patients had no evidence of disease at baseline. One patient entered the trial with PD and achieved a PR and 4 achieved a SD after entering with disease progression. Two patients entered with SD and were assessed as PD post aACT, while 7 had no change from SD and 7 had no change from PD; 1 patient had no evidence of disease at both enrollment and disease assessment. Median OS was 36.2 months, 57.4 for the HER2 3+ patients and 27.4 months for the HER2 0-2+ patients. Median time to tumor progression was 4.2 months, 7.9 and 3.7 months for the HER2 3+ and HER2 0-2+ patients, respectively. There was no correlation between aATC dose and overall survival (OS), time to tumor progression (TTP), nor target cytotoxicity and OS/TTP, although clear trends were observed for increased cytotoxicity correlating with stable disease versus progressive disease. Higher doses of aATC correlated with achieving SD vs PD, and SD correlated with improved OS and TTP. Compared to pre-treatment cytotoxicity, a statistically significant increase in T cell mediated cytotoxicity to the HER2 overexpressing SK-BR3 cell line was observed, and this correlated to a statistically significant increase in IFNγ production. Natural killer cell activity also increased following aACT, but the increases in cytotoxicity and IFNγ production was not statistically significant. Persistence of radio-labelled aATC suggested localization to lung, liver, and spleen immediately after administration and within 4 hours, to bone. After 24 hours, aATC cleared from lung, but persisted in spleen and bone for up to 4 days after infusion; and serial measurements in blood suggested clearance of 50% of aATC within 30 minutes, but there was evidence of radioactivity (1.1%) after 4 days post-infusion.

Lum and colleagues conclude that compared to published data for other agents in advanced metastatic breast cancer, a median OS of 36 months with aATC is encouraging. Following aATC, immunological correlates support an increase in cytotoxicity to HER2+ve target cells and the increase in cytotoxic lymphocytes was observed for up to 4 months.

NK cells can also be mixed ex-vivo with antibodies, and AFMD recently entered into an agreement with MDACC to evaluate pre-mixing NK cells with an AFMD TandAb. MDACC plans to initially evaluate AFM13, a CD16a:CD30 TandAb in the setting of AML with a goal of informing the development of a NK-CD16a:BCMA TandAb immediately following transplant in multiple myeloma.

Adoptive Cellular Therapy (ACT) of the future -Summary

Inevitably, we see a trend for increasing complexity on a cellular level as the challenges of treating cancer demand more enhancements of T cell function and increasing use of synthetic biology. In order for ACT to become widely adopted, safety needs to be improved to address two requirements: (1) the administration of ACT needs to move away from the bone marrow transplant (BMT) unit, and (2) the therapeutic window enhanced, particularly in solid tumors. The use of gate switches, suicide switches, and enablement of T cells to function and persist in the tumor microenvironment without the need for lymphodepletion is central to success, in our view.

Moving away from patient-derived cells is critical to enable an off-the shelf model, obviating the need for patient-specific batch manufacturing of CAR and eTCR products. We see highly engineered donor cells giving way to induced pluripotent stem cell (iPSC) technology where absolute certainty for the precise nature of the genetic modifications can be ascertained on an individual cell basis. These iPSC can be differentiated into the cell of choice, whether it be a α/β T cell, γ/δ T cell, or an NK cell. If successful treatment requires the development of neoantigen-specific therapy, and we believe it will, obviating the need for viral transfection should become a key enabling technology as the time line for product manufacture contracts from months to days.

Exhibit 22. Key features of future approaches in ACT

Next generation	TIL	eTCR	CAR
Target known	No/Yes	Yes	Yes
Viral transduction	No/Yes	Yes	No/Yes
Engineered	No/Yes	Yes	Yes
Polyclonal	Yes	No/Yes	No/Yes
Cell surface target	No	No	Yes
MHC required	Yes	Yes	No
Autologous	Yes	Yes/No	Yes/No
Lymphodepletion	Yes	Yes/No	Yes/No
Solid Tumors	Yes	Yes	Yes
Stem Cell derived	No	No/Yes	No/Yes
Manufacturing time	2-3 weeks	0-2 weeks	0-1 weeks
Dose	10^9 - 10^{10}	10^8 - 10^{10}	10^5-10^6
Safety	Mild	Mild-moderate	Mild - moderate

Source: Wells Fargo Securities, LLC

CD19 – Proof of Concept for Adoptive Cellular Therapy (ACT)

While there have been several proof-of-concept trials for adoptive cellular therapy (ACT), the level of evidence is highest for chimeric antigen receptors (CAR) T-cells targeting the cluster of differentiation antigen, CD19. CD19 is a pan-B cell marker that is found on normal healthy B cells, as well as B cell malignancies such as acute lymphocytic leukemia and non-Hodgkin lymphoma. CD19 has broader coverage across the B cell developmental lineage than that of D20 – the target for RITUXAN, which is not used to treat either acute leukemia. Successful targeting either of CD19, or CD20 would remove all B cells from circulation, leading to B-cell aplasia, which is reversed after differentiation of precursors into functional B cells.

CD19 CAR-T therapies entered clinical development in mid-2000s, with Memorial Sloan Kettering cancer center (MSKCC) initially testing a CD19 CAR-T, later to become JUNO's JCAR015, in chronic lymphocytic leukemia (CLL). However, it was emerging data in CD19 positive acute lymphocytic leukemia (ALL) that focused a spotlight on CAR-T therapy with dramatic data initially in the pediatric setting. To date, CAR-T therapy consists mostly of a single-use patient-specific product infused on day 1 or days 1 and 2 following completion of lymphodepleting chemotherapy. Recently, the off-shelf CD19 CAR-T product, UCART19 developed by CLLS entered clinical development. UCART19 is differentiated from other CD19CAR-T products in the clinic by virtue of being manufactured from donor and not patient-specific T cells.

What is the Bar for Success of ACT in Hematologic Malignancy? Considering endpoints

CD19 CART products are initially being developed in refractory patients where there is no standard of care. In such patients, an overall response rate of more than 20% with a reasonable duration of response has been considered adequate by the FDA. In historical trials of relapsed/refractory patients, complete responses and responses lasting more than 12 months are rarely observed. In this context, the achievement of a 36% CR rate 6 months after CD19 CAR-T AxiCel in KITE's ZUMA-1 DLBCL trial and an 82% response rate reported by NVS in the CTL019 ELIANA pediatric ALL trial could be considered practice changing. With the FDA accepting the CTL019 BLA for accelerated review and KITE completing the AxiCel BLA 1Q17, we expect the FDA to complete its regulatory reviews in 3Q17.

CD19 CAR-T products will likely re-define the hurdle for overall and complete response rates, progression, and overall survival. The achievement of a CR in half or more of refractory patients enrolled in some CD19 CART trials has presented the opportunity to go beyond standard metrics for response assessment for B cell malignancy and look for evidence of residual disease using highly precise techniques. Such analyses are routine in chronic myeloid leukemia, where the advent of BCR-ABL tyrosine kinase inhibitors has redefined the treatment landscape. The use of flow cytometry allows for the detection of cells based on the presence of cell surface markers; thus, 8-color flow measures 8 such markers, and using this technique, 1 cancer cell in 10,000-100,000 normal cells can be detected. The more sensitive technique of DNA sequencing allows for the detection of 1 cancer cell in 1000,000 normal cells – an MRD-10^{-6}; MRD is also referred to as complete molecular remission (CMR).

In some hematologic malignancies, stem cell transplant is a mainstay of therapy, but achievement of the prerequisite complete response is so uncommon in the refractory setting that it is rarely an option. Data from CAR-T trials and in particular, those in ALL, have shown that a significant number of patients are able to achieve a CR and proceed to transplant, which is commonly their 2nd transplant. The concept of using CAR-T as a bridge to transplant is attractive to physicians and patients because of the chance for cure; however, the FDA does not accept "bridge to transplant" as an endpoint. In diseases such as ALL and if CAR-T prove effective AML, where CAR-T could be used as a bridge to transplant in the refractory disease setting, the relative contribution of the CAR-T pre-transplant and the transplant to outcome becomes complex. However, if CAR-T can drive a high rate of MRD-ve CRs, this should translate to improved overall survival with or without a transplant. We note two recent examples correlating achievement of a MRD/CMR with improved survival.

- MD Anderson recently published its analysis on the results of salvage therapy in the setting of relapsed/refractory ALL. Kantarjian and colleagues reported on 78 subjects who achieved a response to BLINCYTO (n=11), or an anti CD33 antibody drug conjugate without (n=41) or with multi-agent chemotherapy (n=26) used as either the first (n=46), or second (n=32) salvage regimen for advanced disease; Jabbour *et al.*, Cancer. 2016 Sep 7. As noted in Exhibit 24, achievement of MRD-ve status was associated with superior outcome with respect to event-free survival (EFS) and overall survival (OS). A 2-year OS rate of 66% was achieved in subjects receiving an allogeneic transplant after an MRD-ve state following salvage 1 treatment, and EFS was superior following a transplant in salvage 1 vs salvage 2.

Exhibit 24. Achievement of a Minimal Residual Disease Negative State is Associated with Improved Outcomes in Relapsed/Refractory ALL

	MRD-ve	MRD+ve
Salvage 1	57%	43%
EFS (months)	18	7
2yr EFS	46%	17%
OS (months)	27	9
2yr OS	52%	36%
Salvage 2	47%	53%

Source: Jabbour *et al.*, Cancer. 2016 Sep 7

- AMGN recently published data from the TOWER trial of BLINCYTO in the relapsed/refractory ALL setting; recall the trial was halted early due to increased overall survival(OS) in the BLINCYTO cohort compared to chemotherapy, Kantarjian *et al.,* N Engl J Med 2017; 376:836-847. Complete response (CR) rates also favored BLINCYTO at 34% vs. 16%, increasing to 44% vs 25% with the inclusion of a CR with incomplete or partial hematologic recovery. Note that complete molecular remission (CMR) was more common in the BLINCYTO recipients, with 76% of BLINCYTO CR patients achieving CMR versus 48% of chemotherapy patients with CMR, defined as less than 10^{-4} blasts at week 12. The median duration of remission was 7.3 months for BLINCYTO and 4.6 months for chemotherapy and median OS at the time of allo-SCT was 6.9 and 3.9 months, respectively.

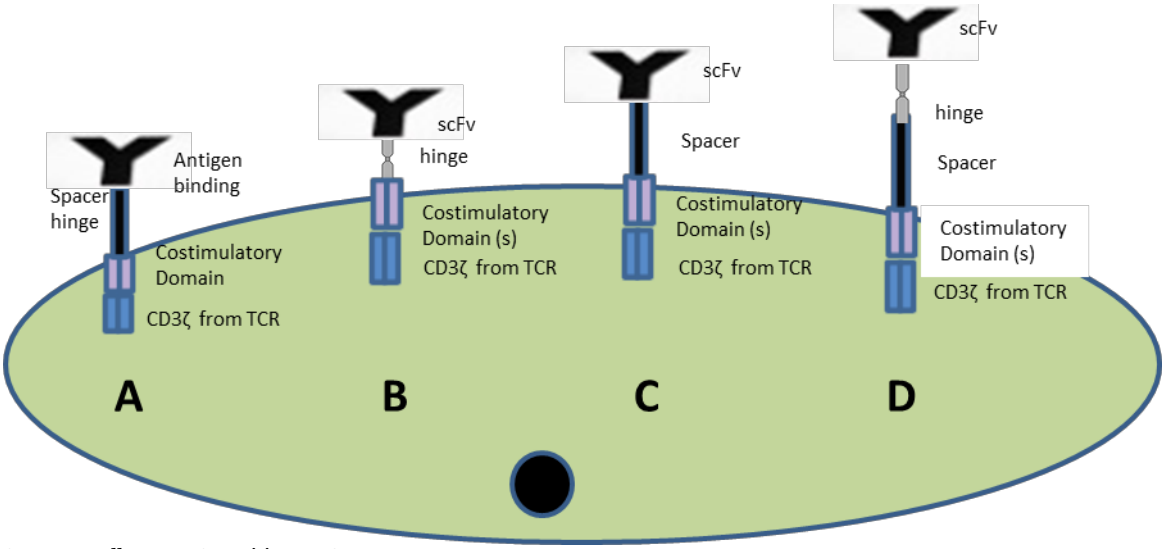
CD19 CART – Academic Roots

Several academic groups developed the first generation of clinical CD19 CAR-T therapies to CD19, choosing differences in CAR architecture with respect to the antigenic target for the single-chain fragment variable (scFv), hinge design connecting CD19 to the transmembrane domain of CD3, choice of costimulatory signal, and type of viral vector used for transfection.

With respect to CAR architecture, the two primordial designs have arisen, one from Memorial Sloan Kettering Cancer Center (MSKCC), which selected a CD19 SJ25C1 scFv with CD28 co-stimulation design, and one from St Jude's Children's Research Hospital (SJRH)/U Penn/FHCRC and SCRI choosing an FMC63 scFv with 41BB (CD137) co-stimulation. Clearly, linkage between the extracellular scFv and the intracellular signaling component of the co-stimulator is required, and this has produced additional diversity. Use of CD-28 as the co-stimulator allowed for the use of the CD28 transmembrane and part of the external domain, also referred to as the spacer, stalk and/or hinge. U Penn chose CD8 as the transmembrane domain and hinge; however, other investigators noted that the length of the linker between the scFv and the CAR-T cell surface modulated efficacy. Invoking the recreation of an antibody-like structure, the CH2-CH3 domains of an antibody could be inserted between the scFv and transmembrane domains to increase the distance between the cell surface and the scFv. Both IgG1 and IgG4 CH2-CH3 domains have been used with varying degrees of success, in part based on binding of Fc receptor positive cells to the IgG domains, which occurs with an IgG1 donor, but not IgG4.

At the site of a tumor, the attraction of macrophages and natural killer cells to the tumor would be desirable on a number of levels, including the possibility for antibody-dependent cytotoxicity (ADCC), as a result of an NK cell binding to the target cell vial the IgG domains of the CAR. However, in some animal models, binding of the CAR via its IgG domain outside of the tumor has led to T cell activation and activation-induced cell death (AICD), as well as T cell sequestering and inactivation by macrophages and NK cells away from the tumor. This has led some investigators to engineer the IgG1 to reduce Fc receptor binding to immune cells or replace it with IgG2 or IgG4 domains that do not bind Fc. The spacer/hinge region likely has to be individualized for each CAR since *a priori*, it is not well understood which combination works optimally. Cooper and colleagues reported that the spacer had a much greater impact on CAR efficacy than expected, noting that a CD8-derived spacer was more effective than an IgG4-derived spacer for their CD123 CAR, PLoS One. 2016 Aug 22;11(8):e0159477. Exhibit 25 illustrates the increasing complexity of CAR architecture.

Exhibit 25. CAR Architecture Optimization



Source: Wells Fargo Securities, LLC

The most advanced CD19 CART products are KITE's Axi-Cel, NVS's CTL019, and JUNO's JCAR017.

- Axi-Cel is derived from an NCI CAR-T program and in 1Q17, KITE completed a final analysis of 101 DLBCL patients enrolled in the U.S. ZUMA-1 trial. FDA review is expected to occur in 3Q17.
- Novartis's (NVS) CTL019 was developed at U Penn and NVS recently announced acceptance of the BLA for priority review by the FDA in the setting of pediatric ALL, with FDA review expected in 3Q17. NVS is completing a 2nd global trial of CTL019, JULIET for DLBCL, and expects to file an sBLA for DLBCL before YE17 and file a Marketing Authorization Application in Europe for both indications.
- JUNO recently terminated development of its most advanced CD19 CAR-T JCAR015 for adult ALL. JCAR015 was always intended as a placeholder for JCAR017, which JUNO considers a more optimized product. JUNO initiated a phase 1 dose escalation trial of JCAR017 in the setting of DLBCL in 4Q15, and following selection of a phase 2 dose in 1H17, plans to initiate a registration cohort with a goal of filing for approval in the United States in 2018. Development of JCAR017 outside of the United States is to be conducted by Celgene (CELG).

Summarized in the following table are the main components used in the CD19 CAR-T products that have been tested in the clinic.

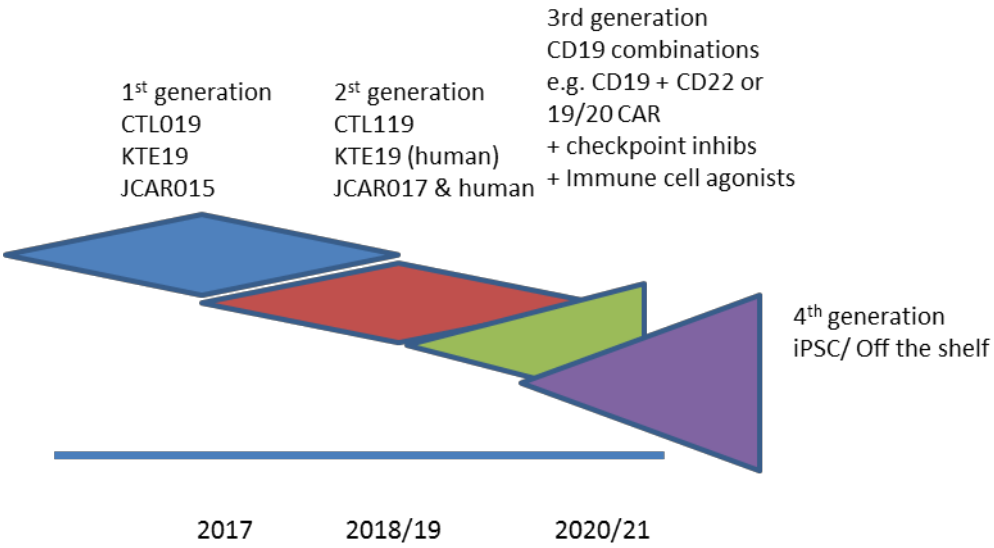
Exhibit 26. Comparative Structures of Selected CD19 CAR-T Products

Product	Sponsor	scFv	Spacer Domain	Signaling plus CD3ζ	Vector	Phase	Dose	Efficacy ALL - CR rate p- Pediatric
CTL019	NVS U Penn	FMC63	CD8a Hinge and transmembrane	41BB	Lentivirus	2	2-5x10^6/Kg	72%/ 82-93% (P)
KTE19 Axi-Cel	KITE NCI	FMC63	CD28	CD28	Retrovirus	2	2x10^6/Kg	60-67% (P)
JCAR015	JUNO MSKCC	SJ25C1	CD28	CD28	Retrovirus	2	1-3x10^6/Kg	78%
JCAR017	JUNO FHCRC/SCRI	FMC63	IgG4-Fc/CD28 hinge	41BB	Lentivirus	1/2	5x10^5-5x10^6/Kg	93% (P)
CD19 CAR	Baylor	FMC63	IgG1-Fc (CH2-CH3)/ CD4 hinge	CD28	Retrovirus	1	1x10^6-2x10^7/m^2	NR
CD19+ CAR-T	ZIOP MDACC	FMC63	IgG1-Fc (CH2-CH3)	CD28	Sleeping Beauty	1	10^6-10^8/m^2	NR

(P) – Pediatric patients, NR – not reported
Source: Wells Fargo Securities, LLC.

The following diagram provides a conceptual framework for what we expect to be a rapid evolution of the CD19 CAR-T space. Starting with today's murine containing scFv products, future products should feature fully human scFvs, optimized to particular T cell subsets and leveraging of synthetic biology to include suicide/control switches, logic gates, and an armamentarium of strategies that allow T cells to thrive in the hostile tumor microenvironment. For example, Bellicum's (BLCM) academic partner in Europe is expected to begin clinical development of a CD19CART containing BLCM's suicide switch in 2017, while ZIOP's tethered IL-15 is expected to enter the clinic in 2018. This 3rd wave of products is to address issues of resistance, with options ranging from dual CAR inhibition, to the addition of non-CAR-T products to increase CAR-T potency, persistency, and modulation of the immunosuppressive tumor microenvironment. While JUNO recently acquired a clinically validated adenosine receptor antagonist, which it intends to bring into the clinic in 2017, NVS is probably best positioned to combine CAR-T with an in-house portfolio of checkpoint inhibitors including PD1, LAG3, TIM3, and CSF1 inhibitors. However, NVS recently shuttered its cell and gene therapy unit to re-integrate those programs into its pharmaceuticals business, and recently made a comment that the current focus for the CART program is on manufacturing. A final chapter of the CAR-T evolution could be the movement away from autologous cells as a source, with adoption of immune cells engineered from induced pluripotent stem cells and the introduction of off-the-shelf T cells, which avoids the need for patient-specific manufacture. CLLS's partner Servier initiated clinical development of a 1st generation off-the-shelf CD19CART in Europe in 2016, and PFE expects to join the ongoing trials with U.S. sites in 2017.

Exhibit 27. Conceptual Rapid Succession of CD19 CAR-T Products



Source: Wells Fargo Securities, LLC

KITE/NCI –- CD28 murine CD19 CAR (Axi-Cel) - retrovirus

In relapsed/refractory (r/r) ALL, NCI has reported an intention to treat complete response rate of approximately 60-70%, 90% of which were free of disease by the most stringent measure – MRD-ve. Severe cytokine release syndrome (sCRS) was observed in approximately 30%. CAR-T persistence was 1-2 months, and leukemia-free and overall survival rates were approximately 10 months and 12-16 months, although post-CAR-T allogeneic transplant complicates analysis. NCI has also published initial experience for CAR-T treatment in aggressive NHL showing complete responses in 4/7 patients, noting that compared to their ALL experience, the CR rate is lower, but CRs are more durable. In addition, while sCRS rates in lymphoma are lower than seen in ALL, severe neurotoxicity (sNT) is more problematic. KITE recently completed the ZUMA-1 trial of Axi-Cel in aggressive NHL using NCI's CAR-T, but with a shortened manufacturing. Using KITE's process, NCI reported similar rates of sCRS, but higher rates of sNT. At the 2017 AACR meeting, KITE presented updated results from ZUMA-1 including 6-month follow-up data. KITE reported an overall response rate of 82%, with a complete response rate of 54%, including 82%/49% in DLBCL, and 83%/71% in transformed follicular or primary mediastinal B cell lymphoma.

National Cancer Institute (Nci) – All Experience

NCI has conducted several clinical trials with a murine-derived ScFv targeting CD19 paired with a CD28/CD3ζ co-stimulatory domain and using a retrovirus for T cell transduction. NCI has established a base non-myeloablative lymphodepletion regimen of fludarabine (25mg/m² days -4 to -2) /cyclophosphamide (900mg/m² d -2) (Flu/Cy) with or without post CAR-T infusion IL-2. NCI conducted a phase 1 dose escalation trial in 21 pediatric patients with a refractory B cell malignancy, including 20 with CD19+ve ALL; 6 patients had primary refractory disease and 8 had failed a prior stem cell transplant, Lee *et al.* Lancet 2015; 385: 517–28. NCI prepared the CAR-T with a retrovirus and manufactured autologous product using an 11-day manufacturing process. Transduction efficiency was reported at 66%, and 19 patients received their assigned CAR-T dose for a 90% feasibility score, i.e., 19/20 received a CAR-T product. Two patients received 3% and 16% of their planned doses of $1x10^6$ and $3x10^6$ CAR-T cells/kg, respectively, due to manufacturing difficulties. In a recent update, NCI has been able to rescue two failed CAR-T product runs with removal of monocytes, and the current success rate on one trial was reported as 96%; MSKCC has reported similar data on the removal of monocytes as a strategy to rescue failed product manufacture.

Based on the occurrence of severe grade 3 or 4 cytokine release syndrome, NCI reported an MTD of $1 x10^6$ CD19-CD28 CAR-T cells/kg. CAR-T doses of $1 x10^6$ or $3 x 10^6$ CAR-T were evaluated in the first 10 patients prior to expansion at $1x10^6$. Post infusion, CD19 CAR-T cells expanded 1-2.5 log over 14 days, but by 2 months post infusion, CAR-T were largely absent, coinciding with normal B cell recovery. Toxicities observed included grade 3-4 cytokine release syndrome (CRS) in 28% (n= 6, 3 grade-3 and 3 grade-4) and to combat CRS, ACTEMRA with or without steroids was used in 3 patients. Six patients reported mild neurotoxicity, including visual hallucinations or impaired speech (dysphasia). With respect to efficacy, NCI reported a 67% CR rate 28 days after CART-T administration, including 12/20 ALL patients who were MRD negative. Of these, 10 underwent allogeneic stem cell transplantation and remained MRD negative after 12 months. The two remaining patients relapsed at 3 and 5 months, respectively, which included relapse with CD19 negative ALL in one patient. The authors noted that 2 patients with active CNS lymphoma resolved all CNS disease after CAR-T therapy. With a median follow-up of 10 months, 51.6% of subjects remained alive and leukemia-free survival in the 12 MRD-ve patients was 79% at 5 months. NCI correlated the degree of CAR-T expansion with both efficacy and severity of CRS. In addition, subjects with severe CRS had demonstrated higher levels of CD8+ve CAR-T, as well as both CD4 and CD8 effector memory CAR-T. NCI recently completed enrollment to this trial and per KITE at the end of November 2015, 25/32 patients treated by NCI were responders, with 21/22 who achieved a CR still in remission.

A recent update from NCI described outcome in a separate doses escalation study enrolling 46 subjects (45 ALL, 1 DLBCL; 53 at ASH 2016 – 51 ALL and 2 DLBCL), all of whom had measurable disease and were considered high risk, including subjects with primary refractory disease (n=9), Philadelphia chromosome +ve (n=5), Downs Syndrome (n=3), and 5 with CNS involvement. Median age was 13 years (range 4-27) and median ALL disease burden was 30% marrow involvement (range 0.03-97%). NCI reported that 60% achieved a CR and 51% were negative for minimal residual disease; and with a median follow-up of 14.5 months, overall and leukemia-free survival rates were 61% and 40%, respectively, with NCI noting 3 patients relapsing with CD19-ve disease. Using a cut-off of 25% marrow blasts to identify high-burden disease, NCI noted that high disease burden correlated with inferior outcome, including a lower CR rate – 40% vs. 84%. In addition, NCI noted that increasing the intensity of the preparative lymphodepleting regimen either by increasing the doses of Flu/Cy (30mg/m²/d x 3 and 1200mg/m²/d x2), or use of alternate chemotherapy regimens did not increase CR rates. At the ASH 2016 meeting, NCI noted that with 2.2 years of follow-up, median OS was 13.3 months for patients receiving Flu/Cy vs. 5.5 months for non Flu/Cy. NCI also noted that allogeneic stem cell transplant after CAR-T correlated with improved outcome. In 21 subjects receiving an allo-SCT post CAR-T, 2 relapsed (9%) vs. 6/7 (86%) who did not receive an allo-SCT post CAR-T; the median time to an allo-SCT was 54 days. Long-term survival was noted in 65% of subjects receiving an allo-SCT, vs. 14% for those not receiving an allo-SCT; however, NCI also reported a 24% incidence of transplant-related mortality. NCI concludes that the risk for CD19-ve relapse is mitigated, but not eliminated, by allo-SCT consolidation post CAR-T. Of the 8 relapses noted, 5 were CD19 –ve, including 4/6 in the non allo-SCT and 1/2 in the allo-SCT cohorts.

With respect to toxicity, the introduction of a grade-driven algorithm reduced the rate of severe CRS with a 10% incidence of grade 3 and 4% of grade 4. No grade 4 neurotoxicity was observed and grade 3 neurotoxicity was observed rarely.

National Cancer Institute (NCI) – NHL Experience

In 2015, NCI published its initial experience of CD19 CAR-T in 11 patients with non-Hodgkin lymphoma, including 9 with aggressive disease. Of the 9 patients with aggressive NHL, 7 were evaluable, including 4 with diffuse large B cell lymphoma (DLBCL) and 3 with primary mediastinal B cell lymphoma (PMBCL), with 4 achieving a complete response, 2 a partial response, and 1 stable disease. Duration of response was 1 month for

SD and 1 PR, and 6 months for 1 CR, with the other responses ongoing at 6+-22+ months. Imaging studies showed convincingly that CART-19 was able to penetrate bulky diseased lymph nodes. One patient died 16 days after CART-19, but the death was not associated with CAR-T-related toxicity. Grade 3 or 4 hypotension was noted in 4 subjects, and all patients had elevations of interleukin 6 and/or IFNγ, and two patients received ACTEMRA for cytokine release syndrome. Neurological toxicities previously observed with CD19 CAR-T therapy in leukemia studies were observed in lymphoma patients, but three patients had different and unexpected neurological abnormalities. In addition to data for the initial cohort of 9 DLBCL patients, NCI has reported data for 5 low-grade lymphoma patients, i.e., 2 CR, 2 PR, and 1 not-evaluable (NE), and 7 CLL patients, i.e., 4 CR, 2 PR, and 1 NE, noting that, including DLBCL, only 1 of 11 patients achieving a CR has relapsed.

KITE is leveraging NCI's CD19 CAR- T experience in the ZUMA-series of trials in CD19+ve leukemia and lymphoma, but with an optimized and shorter manufacturing time line, which has been incorporated into NCI's trials. KITE has reported that the NCI data using its manufacturing process are very similar to the data generated by the NCI's original process, and response rates and response duration are summarized in Exhibit 28.

Exhibit 28. NCI CD19 CAR-T Efficacy

	Initial Response		Best Response	
All patients (n=41)	78%	34%	78%	54%
All aggressive NHL (n=27)	70%	33%	70%	48%
Using KITE process (n=13)	69%	46%	69%	54%

Duration	>5yr	>3yr	>2yr	>1yr
ORR	1	4	10	15
CR	N/A	3	9	12

Source: Company reports

Donor-derived Allo-CAR-T appears safe

Several of the ALL patients treated by NCI had received a prior allo-SCT, and so, while cells used to manufacture their CAR-T product came from the patient, their origin was from the donor of stem cells used in the allo-SCT. Given adequate demonstration of safety and lack of GvHD, NCI conducted a CD19 CART trial in allogeneic hematopoietic stem cell transplant (allo-HSCT) failures using the original donor as a T cell source for CAR-T manufacture.

Donor lymphocyte infusions (DLI) are commonly used following allo HSCT, with the same donor used as a source of cells for both transplant and DLI; however, their success in acute lymphocytic leukemia is poor, with reported CR rates ranging from 0% to 20%, and acute graft versus host disease (aGVHD) the main cause of early mortality, which is observed in 6-11% of subjects receiving a DLI. NCI set out to improve DLI by using the donor cells as a source for CAR-T manufacture. With CAR-T known to be far more potent at killing target cells than donor lymphocytes, and CAR-T leading to a heightened immune state, NCI conducted a dose escalation trial with a focus on safety and in particular, aGVHD. Unlike NCI's standard protocol, lymphodepleting chemotherapy was not administered ahead of the CAR-T infusion. CAR-T doses were escalated from 10^6/Kg to $5x10^6$/Kg in subjects with an unrelated donor (n=5) and 2 $x10^6$ to $10x10^6$ in subjects with a sibling donor (n=15). In total, 8 responses were noted, with 5 ongoing at the time of publication (3+ to 30+ months), and in addition, two patients exhibited long-term stable disease (24+ and 31+ months), although these patients entered the trial with SD and a PR, respectively. The therapy was particularly effective in ALL, with 4 of 5 subjects achieving a minimal residual disease negative complete response, although the response was durable for only 3 months in one patient and 5 months in the second. The NCI was encouraged by the absence of aGVHD, but noted that CAR-T doses were at or below the 10^7 threshold associated with aGVHD from DLI and in addition, the median time for onset of aGVHD generally occurs later than the time at which CAR-T remained in circulation.

Kite Pharma (KITE) leveraging NCI's experience- Axi-Cel BLA for aggressive NHL completed 1Q17

Kite Pharma (KITE) is leveraging the NCI's experience into commercializing CAR-T products and recently completed the goal to finish the rolling Axi-Cel BLA filing by the end of 1Q17 in the setting of relapsed/refractory diffuse large B cell lymphoma . Axi-Cel has both FDA Breakthrough Therapy Designation and EU PRIME designation for DLBCL. In 2017, KITE expects to file Axi-Cel for approval in Europe and enroll the first European patients into a ZUMA-1 extension trial.

For the CD19 CAR, Axi-Cel, KITE is using NCI 's CAR and retroviral vector, but the company has removed human serum albumen from the manufacturing process, moved to a closed manufacturing system, and streamlined production to establish a 6-8 day manufacturing time line. Adding in the time for apheresis, product shipping and release leads to an approximate 14d vein-vein turnaround for Axi-Cel. At the recent 2017 AACR meeting, KITE presented data from the ZUMA-1 trial that included 6 months follow-up data on 101 subjects, including 77 with DLBCL and 24 with transformed follicular lymphoma (tFL) or primary mediastinal B cell lymphoma (PMBCL). In the ZUMA-1 trial, KITE used a 3-day Flu (30mg/m^2)/Cy (500 mg/m^2) lymphodepletion regimen and infused a target of 2x10^6/Kg CD19 CAR-T, and no bridging therapy was allowed between apheresis and Axi-Cel infusion. One month after Axi-Cel, KITE observed an 82% ORR, including 82% and 71% for the DLBCL and tFL/PMBCL cohorts, respectively, and CR rates were 54% overall, including 49% and 71% for the DLBLC and tFL/PMBCL cohorts, respectively. With 6 months of follow-up, approximately 60% of patients who achieved a CR maintained that response.

Approximately 42% of subjects received ACTEMRA and 26% received steroids for toxicity. Rates of grade 3 or higher CRS were 13%, and grade 3 or higher, NT 28%. Three patients died proximal to Axi-Cel, one case of hemophagocytic lymphohistiocytosis (HLH) in a DLBCL patient and one cardiac arrest secondary to CRS in a tFL/PMBCL patient were deemed Axi-Cel related, while a pulmonary embolism was not.

Additional potential registration enabling KITE CD19 CART trials initiated include the following:

- ZUMA-2 phase 2 recruiting relapsed/refractory mantle cell lymphoma patients (n=70), data 2018;
- ZUMA-3 phase 1/2 recruiting relapsed/refractory CD19+ve adult ALL (n=75), data 2018;
- ZUMA- 4 phase 1/2 recruiting relapsed/refractory CD19+ve pediatric ALL (n=75), data 2018; and
- ZUMA-6 phase 1 /2 recruiting relapsed/refractory DLBCL combining Axi-Cel with Tecentriq, phase 1 data 2017, phase 2 data in 2Q18.

ZUMA 6 is the first of a wave of Axi-Cel trials that include ZUMA 5 in indolent NHL, ZUMA 8 in CLL and ZUMA 7 in an earlier less refractory group of DLBCL patients, with patient enrollment planned to start in 2017.

The University of Pennsylvania (Novartis) – 41BB murine CD19 CAR- lentivirus

The University of Pennsylvania (U-Penn)/ Children's hospital of Philadelphia (CHOP) has conducted several trials with a 41BB CD19 CAR-T in CD19+ve leukemia and lymphoma, CTL019. In r/r ALL CR rates of 80-90% have been reported decreasing to approximately 60% in aggressive lymphoma. As with NCI's experience, most CRs are MRD-ve, although CAR-T persistence is longer than reported by NCI with B cell aplasia, a sign of CAR-T persistence ongoing at 5 years in some patients. In r/r ALL, median relapse-free survival is approximately 60% at 12 months and 12-month median overall survival (OS) is close to 80%. As with NCI, approximately 30% of patients suffer sCRS. U Penn and CHOP have treated more than 200 patients with CTL019, and recently, NVS reported interim data from the first 50 relapsed/refractory pediatric ALL subjects enrolled into the 100-patient ELIANA trial. The interim analysis confirmed a high MRD-ve CR rate of approximately 80%, and 6-month relapse-free and overall survival rates of 60% and 80%, respectively. NVS reported a 50% rate of admission to the intensive care unit (ICU), which lasted a median of 8 days, and while 2 deaths were noted within 30 days of CAR-T administration, no fatalities due to cerebral edema have been reported. Based on these data, NVS filed a BLA and recently announced that the U.S. FDA has accepted the filing for accelerated review. NVS is also conducting a potentially registration-enabling trial of CTL019 in DLBCL, JULIET, with plans to submit a sBLA 2H17. Both ELIANA and JULIET are global clinical trials and for European approval, NVS plans to file for both pediatric ALL and DLBCL 2H17. CTL019 has both FDA Breakthrough Therapy Designation and EU PRIME designation for pediatric ALL. NVS conducted the ELIANA and JULIET studies at clinical sites in the United States, Europe, Australia, and Japan, shipping apheresis product to its U.S. manufacturing facility in Morris Plains New Jersey. NVS notes that this is the first Food and Drug Administration (FDA) Good Manufacturing Practices quality site for cell therapy production in the United States. NVS recently announced that the company has processed cells from more than 150 patients in its manufacturing facility. In 2017, NVS plans to add a German-based contract manufacturer for CTL019 supply.

ELIANA – Interim Data For Pediatric Acute Lymphoblastic Leukemia (All)

ELIANA is a global phase 2 trial being conducted at 25 sites and uses NVS's centralized manufacturing facility in New Jersey. The trial enrolled 100 subjects with relapsed/refractory ALL who were 3-21 years old at the time of diagnosis and were naïve to CD19 directed therapy including BLINCYTO. The primary endpoint was overall remission rate within 3 months of a single infusion of CTL019, which was stable for at least 4 weeks. Secondary endpoints included minimal residual disease status (MRD), duration of response, overall survival, CAR-T kinetics, and safety.

Prior to receipt of CTL019, subjects were treated with lymphodepleting chemotherapy, which comprised fludarabine 30mg/m^2/d x 4 and cyclophosphamide 500mg/m^2/d x 2. For subjects ≤50kg, the target CAR-T dose was 2-5x10^6/Kg, decreasing to 1-2.5x10^6/kg for subjects ≥50kg. As of the 2016 ASH presentation, NVS noted that of 98 patients screened, 67 had or were awaiting product infusion, as shown in Exhibit 29. In short, approximately 20% of subjects failed screening, and of those who passed screening, 80% received a CTL019 product with manufacturing failure and patient death, the primary reasons for non-receipt. During Q&A following presentation of the data, the presenter commented that the average time between enrollment and infusion of CTL019 was 41 days.

Exhibit 29. Patient Disposition and Baseline Characteristics in ELIANA

n	Stage	Comment
98	Screened	
81	Enrolled	5 are awaiting infusion
62	Infused	14 discontinued before infusion 6 died 5 product failures 3 adverse events
44	In follow-up	18 discontinued study follow-up 6 died 5 no-response/relapse 5 received new therapy while in CR 2 subject/guardian decision

Baseline Characteristic	Age
Age	12 (3-23)
Prior SCT	56%
Previous lines of therapy	3 (1-8)
Disease status:	
Primary refractory	10%
Chemo-refractory	11%
Relapsed disease	79%

Source: ASH and Wells Fargo Securities, LLC

Of those patients who received a product and with a median of 4.3 months of follow-up (0.4-14.6 months) approximately 10% of subjects died and 8% were either non-responders to or relapsed following CTL019. With respect to baseline characteristics, the average age was 12 years and approximately half of subjects enrolled had received a prior stem cell transplant (SCT). The median number of prior lines of therapy was 3 and the majority of patients, 80% had relapsed, rather than refractory, disease.

In the interim analysis, NVS reported that 41/50 subjects, 82% achieved clinical remission of CR/Cri, defined as less than 5% blasts in the bone marrow with or without recovery of neutrophils or platelets, and all 41 had a blast count of less than 0.01%, meeting the criterion of negative MRD. Compared to the null hypothesis of 20% CR/CRi and 15% MRD-ve, NVS reported a significance level of less than 0.0001 for CTL019 in ELIANA. Median 6-month overall survival was 89% (55/62), and 6-month relapse-free survival was 60%, 24/41.

Kinetics of CAR-T expansion suggested that in 3 non-responders, the peak of CAR-T expansion occurred later and was less robust, as defined by a lower peak and area under the curve over 28 days than observed in responders. With respect to safety, NVS reported that the majority of AE's occurred during the first 8 weeks following CTL019 administration, with serious adverse events (SAE) suspected to be CTL019-related occurring in 68% over the first 8 weeks, compared to 2% and 10% after week 8. Selected adverse events and AEs of special interest are summarized in Exhibit 30.

Exhibit 30. Selected Adverse Events from ELIANA

Event	All grades	Grade 3 %	Grade 4%
CRS	79%	21%	27%
Decreased appetite	39%	15%	2%
Pyrexia	37%	13%	3%
Hypotension	33%	11%	11%
AAT increase	31%	13%	5%
Hypokalemia	26%	13%	3%
Hypoxia	23%	11%	6%

Event	All grades	Grade 3 %	Grade 4 %
CRS	79%	21%	27%
Cytopenia >28d	37%	11%	19%
Infection	40%	23%	3%
Neuropsychiatric	45%	15%	0%
TLS	5%	5%	0%
Cerebral edema	0%	0%	0%

Source for both tables: ASH and Wells Fargo Securities, LLC – CRS – cytokine release syndrome, AAT – alanine aspartate transferase, TLS – tumor release syndrome

NVS noted that 2 deaths occurred within 30 days of CTL019 administration, one due to ALL and the other a cerebral hemorrhage. Of the 62 patients receiving CTL019, 29 were admitted to the ICU, all for CRS; 29/41 patients with CRS were admitted to the ICU (59%). The median time of onset for CRS was 3 days (1-22), with a median duration of 8 days (1-36). Of the 59% admitted to the ICU for CRS, the median duration of stay was also 8 days (1-36). Treatment for CRS included anti-cytokine therapy (51%), high-dose vasopressor support (33%), invasive ventilation (20%), and dialysis (12%).

Phase 1 Experience –Acute Lymphocytic Leukemia (All)

U-Penn reported data from a phase 1 pediatric ALL trial with CTL019 in the New England Journal of Medicine, Maude *et al.*, 2014 October 16; 371(16): 1507–1517. A 90% CR rate was noted (27/30), of which 22/27 were deemed MRD –ve 1-month after the CAR-T. Several patients had failed a prior transplant (n=18) and 3 the CD19/CD3 BiTE, BLINCYTO. U-Penn reported an 82% CR rate in subjects with high disease burden at baseline, including 2 out of 3 who had failed BLINCYTO. As with NCI, U Penn's CD19 CAR-T eradicated any evidence of CNS leukemia present at the time of treatment. Seven patients relapsed following a CR at 6 weeks to 8.5 months post CAR-T, an event that in:

- 3 coincided with premature loss of CAR-T between 2 weeks and 3 months;
- In 3 more patients, disease relapse occurred after recovery of normal B cells, which was observed 2-3 months post-CAR-T, and is indicative of CAR-T loss; and
- In the 7th, an MRD-ve state was never achieved.

In 3 patients, relapse was associated with appearance of CD19-ve disease. At 6 months, U-Penn reported event-free and overall survival rates of 67% and 78%, with 7 subjects dying after disease progression or relapse, including one patient succumbing to myelodysplastic syndrome while ALL free.

In the 27 responding patients, CAR-T expanded to make up approximately 40% of CD3+ve cells in peripheral blood (range 4.4-69.3%), while in the three non-responders, maximal CAR-T levels were 0.2%, 0.6%, and 8.2%. Unlike the NCI experience, U-Penn reported persistence of the CAR-T in the majority of patients, with a 68% probability for persistence at 6 months, and a correlation between persistence and longevity of clinical response. Persistence of the CAR-T also correlated with prolonged aplasia, with U Penn reporting a 73% probability of relapse-free B cell aplasia at 6 months; persistent B cell aplasia required intravenous immunoglobulin (IViG) as supportive care. The U Penn authors concluded that CAR-T persistence for 6 months is likely necessary for disease eradication in patients electing not to undergo a stem cell transplant. As with NCI, U-Penn observed cytokine release syndrome toxicity, and while CRS was mild to moderate in 22 patients, it was serious in 8, leading to the use of ACTEMRA. Severe CRS was associated with a high burden of disease at the time of CAR-T administration. Beyond CRS, U-Penn also reported transient neurologic dysfunction in 13 subjects.

Long-term follow-up data were presented at the recent 2016 ASCO meeting on the first 60 pediatric and young adult subjects treated at CHOP. Baseline characteristics and response are summarized in Exhibit 31.

Exhibit 31. Response to CTL019 is gated by disease burden

Patient Characteristic	N=60	Response
Median Age	11 (1.7-24)	ND
Post allo-SCT	39 (65%)	ND
Baseline ALL burden		93%
>5% blasts	32 (53%)	88%
0.01-5% blasts	12 (20%)	100%
<0.01% blasts	16 (27%)	100%

Source: ASCO and Wells Fargo Securities, LLC

Following CTL019, 56/60 achieved a CR (93%), and all patients with or without MRD at baseline achieved MRD negative status without further treatment by month 3. In the patients with bulky disease, 4 patients did not respond and 3 additional subjects remained MRD positive, of which 2 relapsed. Of the 32 subjects with greater than 5% blasts in their bone marrow, 23 had more than 50% marrow involvement and 6 failed to achieve an MRD-ve CR, for a MRD-ve CR rate of 83%. Patients with a history of CNS disease (n=16), and later in the protocol, patients with CNS disease at baseline (n=6) were enrolled. Of the 6 with active CNS disease, 5 achieved CNS remission and were MRD-ve. The remaining patient did not achieve a response in the bone marrow and was not assessed for change in CNS disease. Of the 15 assessable patients with CNS involvement at baseline, 11 remained in CR with 4 failures due to relapse in the bone marrow without CNS relapse. CTL019 was detected in the CSF of 98% of subjects tested and persisted out to 12 months, which was the time of the last analysis. Relapse-free survival at 12 and 24 months was 60% and 53.2%, respectively, and 24 patients remained in remission for 12 months or longer, 19 without further therapy. Seven patients proceeded to an allo-SCT, and one received a donor lymphocyte infusion; 2 of the SCT patients relapsed. With a median follow-

up of 15 months, 12- and 24-month overall survival was 79% and 61%. CRS was observed in 88% of subjects requiring medical management, with an IL-6 antagonist such as ACTEMRA in 28%.

U Penn's latest ALL clinical trial for CART19 is in the setting of detectable minimal residual disease following up-front treatment. Penn plans to enroll 24 subjects who receive a split CART19 dose of 10%/30%/60% of their target dose over 3 days, NCT02935543; the split dosing over 3 days has become a standard in U Penn CAR-T trials.

Phase 1 Experience - CLL

In a recent update, U Penn has now treated 206 patients with CTL019, including 50 patients with CLL, 114 with ALL, 89 of whom were pediatric, 31 with NHL, and 11 with multiple myeloma. In the early CLL experience, 14 patients were enrolled who were expected to live less than 2 years. Of these 14, 4 achieved a CR all ongoing at 15+ - 60+ months, 4 achieved a PR, and 6 had no response. Peak CAR-T expansion correlated with efficacy as those with a CR saw a greater than 3-log expansion of CAR-T, those with a PR a greater than 2-log expansion, and those with no response less than a 2-log expansion. CAR-T persistence may also be significant with B cell aplasia as a surrogate for CAR-T persistence, lasting up to 5.5 years. In this cohort there was no clear patient or disease characteristic that predicted response, but with a 2-log difference in cell dose, U Penn conducted a randomized phase 2 trial comparing 1-5x10^7 with 1-5 x10^8 CTL019 cells in 24 subjects with CR at 3 months the primary endpoint and a plan to enroll a further 8 patients at the chosen dose. In the randomized stage, 28 patients were enrolled, with 24 achieving their target dose, 11 at the high dose and 13 at the low dose. Lymphodepleting regimens use were primarily bendamustine (5/24), or cyclophosphamide plus fludarabine/pentostatin (18/24), and many patients had fludarabine refractory disease at baseline. Cytokine release syndrome was the most significant toxicity, and like CRS in ALL, it was reversible with use of ACTEMRA to neutralize IL-6. In CLL there was no relationship between CRS severity and CAR-T dose. The overall response rate was 42%, with a CR observed in 25%. There was a suggestion that responses were more robust at the higher cell dose, where a 54% response rate was observed, compared to 31% at the lower dose. An additional 6 patients have been enrolled at the higher dose and for all 17 patients at this dose, U Penn reports a 53% response rate, including a 35% CR rate in a population that had received 3 prior lines of therapy. Prior therapy included IMBURVICA in one-third, and almost half had a p53 deletion at the time of enrollment. Both PFS and OS are approximately 80%, and all relapse events occurred within 3 months.

With recent post-hoc data suggesting that patients receiving CTL019 on the background of IMBRUVICA show improved responses presumably due to improved T cell competence, U Penn has opened a trial of CTL019 in CLL patients that have a PR or SD on the background of IMBRUVICA, NCT02640209. In this 15-patient trial, IMBRUVICA dosing is to be continued as a background to CTL019. At the recent ASH meeting, U Penn also noted that in xeonograft models, IMBRUVICA reduced CRS associated with CD19 CAR-T therapy by inhibiting the production of pro-inflammatory cytokines - another reason for combining CD19 CAR-T therapy with IMBRUVICA.

Phase 1 Experience -NHL

In NHL, U Penn has reported on 43 subjects enrolled in a CTL019 trial, including 26 with DLBCL, 14 with follicular lymphoma, and 3 with mantle cell lymphoma. Of these, 13 patients did not receive a CAR-T product, i.e., 4 for progressive disease, 6 for production failure, and 3 withdrew their consent; and 30 patients received CTL019 doses of 1-5 x10^8 CAR-T. Production failure was due to too few cells harvested and/or poor expansion of cells from the apheresis product of heavily pre-treated patients. In this trial, choice of lymphodepleting regimen was individualized, rather than prescribed by the protocol. In 15 efficacy-eligible DLBCL patients, 7 responded at the 3-month post CAR-T assessment, including 3 with a CR. By month 6, 3 patients with a PR by computer tomography at month 3 converted to a CR, while 1 patient with a PR developed progressive disease at 6 months. Patients with PD at the three-month assessment had declared PD very early, and U Penn noted that response did not correlate with peak CAR-T levels. With a median follow-up of 17.3 months, median PFS was 3 months, with no progression events noted after 6 months.

At the recent ASH meeting, U Penn reported outcomes based on cell of origin - germinal center (GC) or non-germinal center (NGC) in DLBCL, or transformed follicular lymphoma (tFL), including double-hit lymphoma patients defined as those with both a *myc* translocation and a translocation of either BCL2, or BCL6. Three months following CTL019, U Penn reported an ORR of 52% (7/13) and CR rate of 38% (5/13). In the GC subset, ORR was 71% and CR 57%, while in the NGC, all responses were CR, 40%. All 3 tFL GC patients achieved a CR, as did both double-hit GC patients; and at a median follow-up of 23.3 months, none of the patients achieving a CR had relapsed.

In the follicular lymphoma cohort, 13/14 received a CTL019 product and at three months, 10 achieved a response, with 6 a CR and, repeating an observation from DLBCL, 3 patients improved from PR to CR with

additional follow-up. A patient with a PR at months 3, 6, and 9 experienced progressive disease by the month 12 assessment; one patient died in CR from a neurological cause. As with DLBCL, peak CAR-T did not correlate with response. Cytokine release syndrome was observed in the FL cohort, but it did not lead to hospitalization. In lymphoma, U Penn notes that TREANDA is an excellent lymphodepleting preparative regimen.

Fred Hutchinson Cancer Research Center/ Memorial Sloan Kettering Cancer Center/ Seattle Children's Research Institute (JUNO) –CD19/41BB/ lentivirus or CD19/CD28 /retrovirus

Juno has three distinct CD19 CAR-T products, one from each of the three academic collaborators. MSK's JCAR015 was the company's most advanced and basic product, and is most similar to KITE's Axi-Cel and NVS's CTL019, with data in r/r lymphoma and leukemia very similar to those reported by NCI and U Penn. SCRI's JCAR017 and FHCRC's JCAR014 are defined composition products that comprise 1:1 ratios of CD4 and CD8 T cells. JUNO believes that using a defined composition produces less variability in a CAR-T product, which may improve safety and or efficacy. While JCAR014 and JCAR017 are sibling products, JUNO has selected JCAR017 for development and JCAR014 as a test-bed for product development. JACR017 data in relapsed/refractory pediatric ALL suggest MRD-ve CR rates in excess of 90%, with an sCRS profile that may be superior to that reported for NCI and U Penn CAR-T products. In its latest data, JUNO's collaborator has reported a sCRS rate of 15%, which compares favorably to 30% reported by NCI and close to 50% reported in ELIANA. JUNO is currently completing a dose response trial of JCAR017 in relapsed/refractory DLBCL, with a goal to file a BLA in 2018.

While JCAR015 was on paper at least JUNO's most advanced CD19CART, repeated clinical holds have eroded the potential BLA filing time line advantage over JCAR017 to such an extent that JUNO recently elected to terminate development of JCAR015.

JCAR017 in DLBCL

At the 2016 ASH, investigators for the TRANCEND NHL-01 trial presented data on 28 subjects enrolled into the ongoing trial, including 90% with DLBCL. JUNO reported an ORR of 80%, including a 60% CR rate. JUNO noted that all 9 high-risk patients with double-hit (MYC and BCL2 n=2), triple-hit (MYC, BCL2, and BCL6 n=3), or double-expressor (MYC and BCL2 protein expression n= 4) lymphoma responded, with 7 achieving a CR. No grade 3/4 CRS was observed, and while a 33% rate of grade 3/4 NT was noted for JCAR017 given as a single dose, split dosing reduced the rate of sNT to 14%; at the recent 2017 AACR, SCRI lead investigator Mike Jensen noted that if SCRI used the U Penn/NCI/MSKCC definition of sCRS the grade 3/4 sCRS rate for the PLAT-02 pediatric ALL trial would be 0%.

FHCRC is using essentially the same CAR-T architecture and defined CD4:8 product mix, although it focused on central memory, rather than CD8 naïve used in JCAR017, to treated adults with refractory B cell malignancy including ALL. Per kilogram doses of $2x10^5$, $1x10^6$ or $2x10^6$ were evaluated; however, a patient death due to sCRS at $2x10^6$ /Kg established $1x10^6$ as the MTD. Of the 11 patients treated during dose escalation, 9 achieved an MRD-ve CR. Severe CRS and neurotoxicity were reported in 23% and 50% of subjects, respectively.

At the 2016 ASH meeting, FHCRC noted that 106 patients had been treated with JCAR014, including the following:

- IMBRUVICA refractory CLL – 69% ORR with 25% CR rate, with no progression events noted in patients clearing the index CLL clone by deep sequencing. Serious CRS and sNT were noted in 5 subjects (20%) each.

- Adult ALL – 32/34 achieved an MRD-ve CR, including all 22 receiving Flu/Cy lymphodepletion. Serious CRS/NT rates were 39% reducing to 21%/26% in subjects receiving Flu/CY lymphodepletion.

- NHL – 41 patients, including 30 with DLBCL, and 71% received Cy/Flu lymphodepletion; and at a JCAR014 dose of $2x10^6$ /Kg, JUNO reported overall and complete response rates of 81% and a 50%, respectively. Grade 3/4 sCRS was 10% and sNT 5%.

SCRI conducted a phase 1 dose escalation trial of a JCAR017 in pediatric ALL, PLAT-02, with planned doses of $5x10^5$/Kg, $1x10^6$/Kg, $5x10^6$/Kg, and $1x10^7$/Kg. Following cyclophosphamide lymphodepletion, later modified to Flu/Cy (n=14), SCRI observed an MRD negative CR in 40/45 (89%), including 26/28 patients who had relapsed post-transplant, and established $1x10^6$ as the MTD; in 14 subjects receiving Flu/Cy lymphodepletion, the MRD-ve CR rate was 100%. All responding patients showed robust CAR-T expansion 1-2 weeks after infusion and loss of CAR-T persistence correlated with relapse. Six- and 12-month OS were reported as 85% and 73%, and leukemia-free survival at 6 and 12 months, 66% and 50%. Loss of B cell aplasia after 8 weeks correlated with disease relapse, and in PLAT-03, subjects with low disease burden at baseline who are at

higher risk for loss of B cell aplasia are to receive a CD19+ve T cell vaccine over the first 6 months of treatment. Cytokine release syndrome was observed in the majority of patients and was severe in 27%; however, with instigation of aggressive CRS management, the rate of sCRS decreased to 15%. Severe neurotoxicity rates of 22-25% were reported before and after the instigation of CRS management. At the recent AACR meeting, lead SCRI investigator Mike Jensen noted that applying the commonly used U Penn definition for sCRS to the PLAT-02 trial would have led to a 0% incidence of sCRS.

JCAR015 in ALL

Memorial Sloan Kettering Cancer Center (MSKCC) has conducted a phase 1 dose escalation trial (1-3 $x10^6$ CAR-T/kg) of the CD19/CD28 CAR-T, JCAR015 in relapsed/refractory pediatric or adult ALL. Eligible patients received cyclophosphamide (1.5-3.0 g/m^2) lymphodepleting chemotherapy the day before CAR-T administration, which was split over 2 days using CAR-T doses of $1x10^6$/Kg on day 1 in subjects with 5% or more ALL blasts (morphologic disease) at baseline and $3x10^6$/kg on day 2 or on both days for those patients with less than 5% blasts (minimal disease). The most recent update at ASH covered 45 adult subjects, approximately half of whom had morphologic disease at baseline, with MSKCC reporting an 82% CR rate (37/45; 75% for morphologic disease at baseline and 90% for minimal disease at baseline). The MRD-ve rate was a 66% rate, and in 36 CR patients assessable for MRD, 30, or 83%, were MRD-ve.

Exhibit 32. Summary of Clinical Outcomes for JCAR015 Pediatric ALL Trial

	Number of Patients n=45
Overall CR Rate	37/45 (82%)
Morphologic disease	18/24 (75%)
Minimal disease	19/21 (90%)
Overall MRD-ve rate	30/36 (83%)
Median time to CT	21 days

Source: Company documents

Of the 37 patients achieving a CR, 18 have relapsed, including 4/12 who went on to receive an allo-HSCT. Another 3 subjects relapsed with CD19-ve disease. At relapse, MSKCC has re-treated 5 patients, two of whom had no response and 3 of which had a transient response. Median OS with a median follow-up of 5.9 months was reported as 9 months, ranging from 6 months in the MRD+ve cohort and not reached in the MRD-ve cohort; but notably, most patients eligible for transplant elected to receive one. Since not all patients elected to receive a post JCAR015 transplant, MSK has compared outcomes between the transplant and non-transplant cohorts for MRD –ve CR patients, noting median survival of not reached vs. 10.6 months (at ASH 2014, MSKCC reported median OS of 10.8 and 8.5 months, respectively). MSKCC suggests that survival is similar between the post JCAR015 transplant and non-transplant cohorts, and that CAR-T can move beyond a role as bridge to transplant to replacing transplant. As of ASH 2015, 13 of 37 patients remained in CR.

As with other CD19 CAR-T products, CRS was common, including severe in 11/46 (24%) patients and the FDA placed the JCAR015 trial on hold following two sCRS-related deaths. In total, 3/46 subjects suffered toxicity-related death. JCAR015 came off hold following several protocol changes excluding certain patients or prohibiting infusion on day 2 in the event of acute toxicity as examples; MSKCC delivers the CAR-T dose over 2 consecutive days. MSKCC has also reported neurological toxicity in 28% of subjects. With respect to persistence, MSKCC noted that CAR-T persistence exceeded 3 months in most patients.

MD Anderson (ZIOP) –CD19/CD28 with Sleeping Beauty

MD Anderson's approach to modifying T cells is to avoid the need for a virus to transport the CAR DNA into the T cell by using the "Sleeping Beauty" (SB) transposon. MD Anderson has conducted trials using a CD19 CAR-T in the post-transplant setting. While both patients with minimal residual and bulky disease have been treated, data for the MRD setting have been updated most recently. ZIOPHARM (ZIOP) and MD Anderson are conducting a trial with a revised CD19 CAR in bulky disease, which should allow more of a direct comparison to existing data from virally manufactured CAR-T products. Ultimately, Sleeping Beauty (SB) is destined for use for point-of-care eTCR, and ZIOP may use lentivirus for autologous CAR-T products.

At the 2015 American Society of Hematology (ASH) meeting, MDACC describing the use a CD19 CAR-T in the post-hematopoietic transplant setting. In one trial, lymphoma patients undergoing an autologous stem cell transplant received the CAR-T immediately after the transplant. In a second trial in acute lymphocytic leukemia, patients received the CAR-T approximately 2 months after the transplant. In both trials the preparative chemotherapy regimen used for transplant was considered sufficient to negate the need for lymphodepletion immediately prior to the CAR-T infusion. With up to 3 years follow-up in the autologous setting, ZIOP noted an impressive, 3-year progression-free survival (PFS) of 83%, compared to a relevant historical control of 49%, and that the approximately 200-day survival of the CAR-T cells was comparable with data for viral CD19 CAR-T products presented also at ASH 2015, as noted in Exhibit 33.

Exhibit 33. Summary of First-in-Man Data for the Sleeping Beauty CD19 CAR-T used as Consolidation Following Either an Autologous, or Allogeneic Hematopoietic Stem Cell Transplant

T-cell survival	Average # of days	Maximum days
Autologous	201*	360
Allogeneic	51	180

** Compares favorably versus 5 of 7 other CD19 studies presented at ASH*

Patient groups	Summary
Autologous	83%, 3-yr PFS compared with 49% historical controls
Allogeneic (all)	63%, 1-yr OS compared with 20-34% historical controls
Allogeneic (haploidentical)	75%, 1-yr OS and no GVHD despite large numbers of T cells infused

Source: Company documents

Data are less mature in the allogeneic setting; however, ZIOP noted that approximately twice as many subjects remained progression free or were alive at 12 months, as expected from historical controls. CAR-T durability was less in the allogeneic setting, likely reflecting that fact that delayed administration reduced the ability of the CAR-T to engraft.

A notable observation from the allogeneic study was that graft versus host disease (GvHD) was not exacerbated despite the high doses of activated T cells given, and this observation was especially noteworthy in the haploidentical setting.

These data were developed using a manufacturing time line of approximately 1 month; and in early 2017, ZIOP announced that the first patient receiving a SB CD19-CAR-T product produced in 2 weeks was dosed.

Adoptive Cellular Therapy (ACT) – A Selection of Who's Invested

Adaptimmune (ADAP) – eTCR leader

ADAP is the leading developer of eTCR therapies for solid tumors; sister company Immunocore is developing bi-specific T cell engagers using eTCRs. ADAP uses affinity maturation to achieve what it believes is the optimal function of the eTCR. Lead product, New York esophageal squamous cell carcinoma-1 (NY-ESO-1), SPEAR is partnered with GSK, (GSK3377794) and is being evaluated in a broad number of indications, but with accelerated development in synovial sarcoma. GSK recently selected PRAME as its second target under a 5-target deal with ADAP. ADAP is currently switching between an academic manufacturing process to one that should support initial commercial launch. ADAP plans to use the newer manufacturing process for a potentially registration-enabling trial of NY-ESO1 SPEAR in synovial sarcoma, which should be initiated by mid-2017. ADAP is also studying NY-ESO1 SPEAR in myxoid round cell liposarcoma, which, like synovial sarcoma, bares a hallmark translocation, leading to NY-ESO1 overexpression. In 2017, ADAP plans to initiate a trial of NY-ESO1 SPEAR in NY-ESO1+ve myeloma in combination with a PD-1 checkpoint inhibitor; in this setting, NY-ESO1 SPEAR also recognizes a LAGE1 peptide that can also be over-expressed in myeloma. NY-ESO1 SPEAR is being developed as an autologous product in selected subjects and specifically, NY-ESO1 SPEAR is restricted to HLA-*A2, which is found in approximately 40% of the Caucasian population. Beyond NY-ESO1 SPEAR, ADAP has IND's approved or planned for 3 other HLA-A*2 restricted eTCRs: MAGE A3/4, MAGE A10 and alpha-fetoprotein (AFP). The two MAGE targets are cancer testis antigens that are over-expressed across a broad range of solid tumors, and ADAP has entered into a collaboration with MD Anderson to help lead development of these products. AFP is a developmental antigen that is found in hepatocellular carcinoma. Most recently, ADAP entered into a collaboration with BLCM for co-development and co-commercialization of two products that are to include BLCM's GoTCR technology. GoTCR uses an inducible costimulatory domain to provide survival signals to T cells, which may increase their persistence as they locate their eTCR-defined target. ADAP and BLCM expect to select clinical candidates in 2017 and begin formal co-development in 2018.

Exhibit 34. Selected ADAP Clinical Trials

Product	Target	Autologous or allogeneic	Lead Indications	Status
NY-ESO-1 SPEAR	NY-ESO-1	Autologous	HLA-A2 NY-ESO-1 +ve synovial sarcoma n=65	Phase 1/2
MAGE A10 SPEAR	MAGE A10	Autologous	HLA-A*0201/*0206 MAGEA10 +ve NSCLC n=32	Phase 1/2
NY-ESO-1 SPEAR	NY-ESO-1	Autologous	HLA-A*0201/*0205/*206 NY-ESO-1 +ve NSCLC n=10	Phase 1/2
NY-ESO-1 SPEAR	NY-ESO-1	Autologous	HLA-A2+ve NY-ESO-1 or LAGE1 +ve myeloma n=26 HLA-A2+ve NY-ESO-1 or LAGE1 +ve myeloma n=10	Phase 1/2
MAGE A10 SPEAR	MAGE-A10	Autologous	HLA-A*0201/5 MAGE A10+ve H&N, urothelial or melanoma n=22	Phase 1
NY-ESO-1 SPEAR	NY-ESO-1	Autologous	HLA-A*0201/*0205/*206 NY-ESO-1 +ve myxoid/round cell liposarcoma n=15	Phase 1/2
NY-ESO-1 SPEAR	NY-ESO-1	Autologous	HLA-A*0201 NY-ESO-1 +ve ovarian cancer n=10	Phase 1/2
NY-ESO-1 SPEAR	NY-ESO-1	Autologous	HLA-A*0201 NY-ESO-1 +ve NSCLC n=10	Phase 1/2
NY-ESO-1 SPEAR	NY-ESO-1	Autologous	HLA-A*0201 NY-ESO-1 +ve myeloma n=6	Phase 1/2

Source: www.clinicaltrials.gov and Wells Fargo Securities, LLC

Amgen (AMGN) – KITE collaboration for CAR-T and neo-epitope strategy with ADXS

Amgen (AMGN) has a major collaboration with KITE on CAR-T products for as yet undisclosed targets in solid tumors and hematologic malignancy. The collaboration leverages AMGN's in-house cancer targeting expertise from bispecific antibodies (BiTE) products like BLINCYTO. KITE recently announced the first target from this collaboration, and AMGN is expected to declare its first target in 2017.

In 3Q16, AMGN announced a global agreement with Advaxis (ADXS) to develop and commercialize the latter's pre-clinical stage patient-specific immuno-therapy, ADXS-NEO. ADXS-NEO utilizes, ADXS's proprietary attenuated *Listeria monocytogenes* bacterial vector to activate the patient's immune system to unique patient-specific neo-epitopes, which are specified by the vector. Neo-epitopes are identified using ADXS's MINE or My Immunotherapy Neo-Epitopes technology. ADXS's MINE technology allows identification of patient-specific neo-epitopes and subsequent engineering of the bacterial vector over a period of days. AMGN made a $40MM up-front payment to ADXS and acquired an additional $25MM in ADXS equity. AMGN is to be responsible for clinical and commercial activities following demonstration of proof-of-concept (PoC). ADXS retains

responsibility for manufacturing, and following demonstration of proof-of-concept, is to receive development, regulatory, and sales milestones of up to $475MM and high-single-digit to low- to mid-double-digit royalties.

Astellas/Potenza – an antibody recognizing an HLA-expressed peptide

In 2017 Astellas plans to file an investigational new drug (IND) application for a TCR-like antibody, h8F1 targeting the PR1 antigen in acute myeloid leukemia (AML) in HLA-*A2 subjects. Separately, in collaboration with Potenza Therapeutics, the companies plan to file INDs to two new checkpoint inhibitors, including one targeting T_{REG}.

Atara Bio – allogeneic anti-viral ACT

Atara Bio (ATRA) uses donor-derived allogeneic T cells that target viruses or proteins associated with cancer as ACT. ATRA's lead product, ATA129 has U.S. FDA BTD in the setting of EBV-associated lymphoproliferative disease (EBV-LPD) following allo-HSCT that is refractory to RITUXAN. In late 2016, ATRA reached agreement with the FDA on the design of two-phase single-arm 3 trials, one in the setting of hematopoietic stem cell transplant, and the second in the setting of solid organ transplant. Following PRIME designation in Europe, ATRA intends to submit for Conditional Marketing Authorization in 2018 in the setting of RITUXAN-refractory EBV-LPD based on completed phase 1 and 2 trials and ongoing phase 3 trials. ATRA is also developing allogeneic off-the-shelf T cell products for CMV infection and WT-1+ve myeloma. ATRA's most recent development candidate is an EBV-targeting T cell that is to be evaluated in multiple sclerosis.

Aurora Biopharma- bispecific CAR-T

Aurora Biopharma is developing bispecific CAR-T therapies for solid tumors that target both a main antigen and a co-stimulatory antigen. Aurora has completed phase 1 testing of AU101 in sarcoma/osteosarcoma and as an intracranial therapy for glioblastoma, and the company is planning a phase 2b/3 trial in sarcoma/osteosarcoma and a phase 1b/2a intrathecal trial for recurrent GBM. Separately, AU105 has completed a phase 1b/2a trial in newly diagnosed GBM and the company is planning to start a phase 2b/3 trial in this setting and indication, for which the company is applying to the FDA for Breakthrough and Orphan Drug designations.

Autolus – Leveraging CAR-T pioneer Martin Pule

Autolus was founded on technology from one the of CAR-T pioneers, Dr Martin Pule, who has been credited with helping to develop the iCAS9 system being commercialized by Bellicum (BLCM), a 3rd generation GD2 targeting CAR for neuroblastoma and the truncated CD20 kill switch being used by Cellectis (CLLS). Autolus appointed former Micromet CEO Christian Itin as CEO 1Q16.

In 2017, Autolus intends to initiate clinical development of two CAR-T programs:

- 2Q17 – targeting APRIL – in the setting of myeloma; APRIL recognizes both BCMA and TACI
- 2H17 – targeting TRBC1 in the setting of TRBC1+ve T cell leukemia

Bellicum (BLCM) – Emerging full-service ACT player with best-in-class suicide switch technology and unique co-stimulatory platform

BLCM's lead program is an engineered T cell product for use following an α/β depleted haploidentical allogeneic hematopoietic stem cell transplant (haplo-SCT). BPX-501 is engineered with an inducible caspase 9 suicide switch, which is activated following IV administration of the small molecule, rimiducid. BPX-501 is intended to increase the speed of immune recovery following an α/β depleted haplo-SCT, leading to reductions in infection rate and length of hospitalization, while allowing for control of graft versus host disease (GvHD). GvHD is a potentially fatal complication of a standard haplo-SCT and there is no standard of care for GvHD prevention. Rimiducid leads to significant BPX-501 cell death minutes after dosing and in clinical trials, rimiducid administration has led to resolution of GvHD within 24 hours. BLCM has received guidance from European regulators that could allow a regulatory filing for BPX-501 in the setting of malignant and non-malignant disease as early as 1H18. In the United States, BLCM is finalizing plans with the FDA for BPX-501 registration, which the company expects to require pivotal trials and result in a 12-18 month lag for product approval.

BLCM has leveraged the technology used in BPX-501, referred to as CaspaCIDe into the ACT space:

- CIDeCAR/TCRs use CaspaCIDe in a CAR-T or eTCR product to allow for rimiducid-induced removal in the event of toxicity. BLCM's CAR-T products also incorporate the novel co-stimulatory domain referred to as MC, instead of CD28 or 41BB.

- GoCAR-T / Go-TCRs uses rimiducid to activate the MC costimulatory switch, which provides survival signals to the T cell. This may be especially beneficial in the setting of solid tumors, where T cells may face challenges penetrating the tumor. Stimulation of MC in this setting may extend the window of opportunity for a CAR-T or eTCR to find its target.

In 1Q17, BLCM initiated phase 1 testing of the first GoCAR product, BPX-601, targeting prostate stem cell antigen (PSCA) in pancreatic cancer, as well as the first CaspaCIDe TCR targeting PRAME, BPX-701. BLCM collaborated with the University of Leiden to obtain a naturally occurring TCR against PRAME. In the United States, BLCM plans to evaluate BPX-701 in AML, while in Europe, Leiden is to lead a trial in uveal melanoma, a disease without a standard of care that could allow for a fast-market opportunity.

In2016, BLCM elected not to file an IND for BPX-401, a CaspCIDe CAR-T targeting CD19. Instead, BLCM entered into a collaboration with the Ospedale Pediatrico Bambino Gesù (OPBG) in Italy. OPBG plans to conduct investigator-led trials of the CD19 CaspaCIDe product as part of a broader collaboration, which should see OPBG manufacture BPX-701 for the uveal melanoma trial, as well as conduct a pediatric neuroblastoma trial with a GD2 CAR-T. OPBG has been the lead site for the potentially registration-enabling BPX-501 trial.

Exhibit 35. Selected BLCM Clinical Trials

Product	Target	Autologous or allogeneic	Lead Indications	Status
BPX-601	PSCA	Autolgous	Non-resectable relapsed/refractory pancreatic cancer (n=30)	Phase 1

Product	Target	Autologous or allogeneic	Novel Features	Lead Indications	Status
BPX-701	PRAME	Autologous	Suicide CideCAR	relapsed/refractory PRAME +ve acute myeloid leukemia or myelodyspalstic syndrome n=36	Phase 1

Source: www.clinical trials.gov and Wells Fargo Securities, LLC

Bluebird Bio (BLUE)- CAR-T effort led by NCI pioneer, CELG collaboration pared down to 1 target, collaborating with KITE on HPV eTCRs

Bluebird Bio – BLUE recruited Dr Morgan, an author on several NIH CAR-T and TCR manuscripts to lead their ACT program. BLUE and CELG are collaborating on developing 1st and 2nd generation CAR-T products to the B cell maturation antigen (BCMA) for use in myeloma. BLUE has also entered into an exclusive agreement with Viromed to develop and commercialize a CAR-T using a proprietary humanized antibody to an undisclosed target in solid tumors.

BLUE and CELG presented interim phase 1 data from an ongoing phase 1 trial of BCMA-targeting CAR-T bb2121 in relapsed/refractory myeloma. BLUE presented data from 3, 3-patient cohorts evaluating doses of $5x10^7$, $15x10^7$, and $45x10^7$ with all 6 patients at the two higher doses achieving a PR or better. In 2017, BLUE plans to file an IND for bb21217, a 2nd BCMA product that utilizes improved manufacturing.

BLUE has published on the use of nucleases to target CAR integration into a specific site, including the TCRα chain potentially killing 2 birds with one stone, namely introduction of a BCMA CAR, and creation of an allogeneic CAR-T product. BLUE also has a collaboration with KITE to utilize BLUE's lentiviral gene-editing technology to improve T cell function for developing 2nd generation TCR products against HPV-16 E6.

Exhibit 36. Selected BLUE Clinical Trials

Product	Target	Autologous or allogeneic	Lead Indications	Status
bb2121	BCMA	Autologous	Relpased myeloma (n=50)	Phase 1
bb21217	BCMA	Autologous	BLUE intends to initiate a phase 1 trial of bb21217 in 2017	IND

Source: company documents, www.clinical trials.gov and Wells Fargo Securities, LLC

CARsgen

CARsgen is a Chinese CAR-T company that is completing phase 1 CAR-T trials in China targeting EGFR and PGC3. Following completion of a $30MM series B financing, the company is planning to file an IND with the FDA to initiate clinical testing of the EGFR and GPC3 CAR-T products in the United States.

Exhibit 37. CARsgen Clinical Trial

Product	Target	Autologous or allogeneic	Lead Indications	Status
CAR-GPC3	Glypican-3	Autologous	CARsgen is conducting a phase 1 single center trial in China in relapsed/refractory squamous lung cacner n=20	Phase 1

Source: www.clinical trials.gov and Wells Fargo Securities, LLC

Celgene (CELG) –Replaced BLUE as major CAR-T partner with JUNO

CELG has a collaboration with BLUE for 1st and 2nd generation CAR-T products targeting BCMA. More important is CELG's 10-year Master Research and Collaboration Agreement with JUNO that gives CELG rights to exercise a global, ex-China, co-development/co-commercialization option on several programs including JUNO's CD19 and CD22 CAR-T candidates. CELG recently executed its right to license the CD19 program paying a $50 million fee and committing to a midteens percentage of sales from it territories.

CELG is also developing NK cell therapeutics independent of collaboration. PNK-007 is in clinical testing for both multiple myeloma, NCT02955550, and AML, NCT02781467.

Exhibit 38. CELG ACT Clinical Trials

Product	Target	Autologous or allogeneic	Lead Indications	Status
PNK-007	None	Allogeneic NK	Post autologous stem cell transplant in myeloma n=40	Phase 1
PNK-007	None	Allogeneic NK	Relapsed/refractory acute myeloid leukemia n=40	Phase 1

Source: www.clinical trials.gov and Wells Fargo Securities, LLC

Cell Design Labs

Cell Design Labs was spun out of UCSF in 2016 to develop a synthetic Notch switch originating in the laboratory of Wendell Lim. Lim has also published on the use of small molecule rapalog switches and in 2Q16, KITE announced a research collaboration and license agreement with Cell Design Labs (CDL) to develop next-generation, precision-controlled CAR-T products utilizing CDL's small molecule switching technology with a focus on certain targets relevant for AML. KITE has an exclusive option to broaden the agreement to other B cell malignancies.

By 2H18, CDL intends to have 1-2 internal ACT programs in the clinic and 1 new partnership.

Cellectis – (CLLS) Leader in off-the-shelf technology and next-generation T cell engineering for CAR-T

Cellectis (CLLS) has used a gene-editing technology to engineer a universal T cell from healthy donors that can be used as an off-the-shelf product. Specifically, CLLS uses transcription activator-like effector nucleases (TALEN) to delete targeted sequences in genes including the T cell receptor alpha gene (TRAC). Absent TRAC, the universal T cells are unable to assemble a functional TCR and are unable to cause GvHD. TCR knockout cells can be used for CAR-T therapy with or without further modification. CLLS also incorporates a proprietary CD20-based kill switch allowing for rituximab-based CAR-T removal. CLLS has major collaborations with Servier and Pfizer (PFE) for the CD19 CAR-T, UCART19.

CLLS's lead program , UCART19, an off-the-shelf CD19 CAR-T, entered clinical testing in the United States in December 2015 and is being developed by Servier ex-United States and PFE in the United States. Wholly owned CAR-T cell targets in development include CD123, CS1, CD38, and CD22. The core to UCART is knocking out of the TCR alpha chain on T-cells, preventing the transplanted T cells from targeting host tissue causing graft versus host disease (GvHD). CLLS's UCART123 has open INDs for AML and blastic plasmacytoid dendritic cell neoplasm (BPDCN), with the first patient expected in 2Q17. CLLS plans to file INDs for UCARTCS1 and UCART22 in 2017, and UCART38 in 2018. UCARTCS1 and UCART38 are to be developed in multiple myeloma and UCART22 for B cell malignancy.

Additional gene knockouts are being targeted such as deoxycytidine kinase (DCK) or CD52 conferring resistance to standard-of-care chemotherapy namely cytarabine/ fludarabine and alemtuzumab. UCART123 is

being developed in two versions, one with a TCR knockout, and version 2 with knockouts to both TCRα and DCK. CLLS plans to conduct UCART123 trials initially at Weill Cornell and MD Anderson. With Weill, CLLS plans to evaluate both versions of UCART123 in AML, and with MD Anderson, a trial with version 1 is to be conducted in BPDCN. CLLS is also working with MD Anderson on the CS1 CAR; however, due to the presence of CS1 on T cells, CLLS's approach is to knock out CS1 in addition to the TCRα; a second version incorporates a PD1 KO.

Exhibit 39. Selected CLLS Clinical Trials

Company	Product	Target	Autologous or allogeneic	Novel Features	Lead Indications	Status
Servier/PFE	UCART19	CD19	Allogeneic	TCR KO CD20 kill	UCART19 is being developed in chronic lymphocytic leukemia and acute lymphoblastic leukemia	Phase 1
CLLS	UCART123	CD123	Allogeneic	TCR KO (DCK KO) CD20 kill	ALCLS plans to study the TCR KO in acute myeloid leukemia (AML) and blastic plasmacytoid dendritic cell neoplasm and a TCR/DCK KO in AML.	Phase 1
CLS	UCARTCS1	CS1	Allogeneic	TCR, CS1 (PD1) KO	ALCLS plans to study the TCR/CS1 dual KO in multiple myeloma with a 2nd generation UCART incorporating PD-1 KO	Pre-clinical

Source: www.clinical trials.gov and Wells Fargo Securities, LLC

CLLS has also developed a small molecule-inducible multichain IgE-based CAR, obviating the need for a suicide switch. Specially, ALCSL describes the use of rapamycin interacting with an FKBP12 domain that is engineered between the hinge region of the CAR and the ScFv. CLLS has also developed a CAR that is active only in the presence of low oxygen levels, such as are found in solid tumors, which may increase the therapeutic window for targets that are found on both tumor and heathy tissue.

Cellular Biomedicine Group

CBMG was established in China in 2009, where it conducts its research and operates a U.S.-compliant cGMP manufacturing facility. Cellular Biomedicine Group is traded on the NASDAQ and has a U.S. base in California. The company's clinical CAR-T, C-CAR011 targets CD19 and is in development for DLBCL and ALL. In pre-clinical development, the company lists anti-CD20 for CLL and NHL, bispecific CD19/CD20 for CLL and NHL, BiCAR (undisclosed) for AML, PD-1 knockout CAR (undisclosed) for hematologic and solid tumor, anti-CD30 CAR for HL, anti-CD22 CAR for hematologic malignancy, and an anti-EGFR CAR for solid tumors.

Exhibit 40. CMBG Clinical Trials

Product	Target	Autologous or allogeneic	Lead Indications	Status
C-CAR011	CD19	Autologous	CBMG is conducting a phase 1 single center trial in China in relapsed/refractory DLBCL n=15	Phase 1
C-CAR011	CD19	Autologous	CBMG is conducting a phase 1 single center trial in China in relapsed/refractory Adult ALL n=20	Phase 1

Source: www.clinical trials.gov and Wells Fargo Securities, LLC

Cell Therapy Catapult

Catapult Therapy has initiated clinical development of a WT-1 TCR in AML (NCT01621724) and a second trial in patients with either myelodysplastic syndrome, or AML (NCT02550535) who have failed a hypomethylation agent. Catapult recently reported that the first 3 patients in the 535 trial had been dosed successfully with a low dose of WT-1 TCR. Catapult's product is a mixture of CD4+ve and CD8+ve cells.

Exhibit 41. Cell Therapy Catapult Clinical Trial

Product	Target	Autologous or allogeneic	HLA	Lead Indications	Status
WT-1 TCR	WT-1	Autologous	HLA-A*02	Catapult is conducting 2 studies, one in AML and the other in AML or MDS patients who have failed hypomethylating therapy n=18	Phase 1/2

Source: www.clinical trials.gov and Wells Fargo Securities, LLC

Chimeric Therapeutics – academics spin-out to focus on one CAR-T target

Chimeric Therapeutics was spun out from the United Kingdom's Cancer Research Technology, University of Birmingham and Cell Therapy Catapult to develop a CAR-T targeting C-type lectin domain family 14-member A, CLEC14a a specific marker of tumor angiogenesis.

CytomX – Partnership with MDACC for Probody-NK CAR-T

CytomX has developed the Probody technology, which requires the presence of a tumor-associated enzyme to activate an antibody, thus preventing unwanted activation in heathy tissue. In late 2015, CytomX entered a collaboration with MDACC to develop Probody-enabled CARs for use in NK cells, ProCAR-NK. The partners aim to use targets that have traditionally been limited by toxicity, and CytomX retains an option to license therapeutics that demonstrate proof of concept.

Endocyte – Universal CAR-T

Endocyte announced a collaboration with Seattle Children's Research Institute to develop a universal CAR-T using ECYT's small molecule drug conjugate (SMDC) bi-specific platform. The universal CAR-T binds to fluorescein isothiocyanate (FITC), while the SMDC binds to a tumor target and presents FITC for engagement of the CAR-T. ECYT expects to complete pre-clinical evaluation of a lead CAR-T in 2H17.

Eureka Therapeutics – Reducing CRS

Eureka has developed the ARTEMIS platform, which provides an alternative to the standard CAR architecture with a proposed advantage of reducing T cell hyper-activation following contact with a tumor cell. Eureka believes this approach can reduce unwanted side effects such as cytokine release syndrome (CRS) without reducing potency. Eureka has isolated several antibodies to difficult to drug targets, including peptides presented by MHC, which are incorporated into the ARTEMIS structure. In 2017, the company intends to initiate clinical development of an alpha-fetoprotein (AFP) and a CD19 CAR-T. The AFP program uses a traditional CD28 CAR, while the CD19 program is to use ARTEMIS. Separately, Eureka has two collaborations with JUNO/MSKCC, one for MUC16 and one for myeloma targets including BCMA. In addition a WT-1 targeting program is partnered with NVS.

F1 Oncology -- Conditionally active CARs

F1 Oncology is a U.S.-based CAR-T company that has agreements with BioAlta for the use of their conditionally active biologic (CAB) platform on CAR-T. SunTerra Capital participated in F1's Series A financing and through a development and commercialization agreement with Shanghai SunTerra Biotechnology, F1 Oncology intends to being clinical testing of a first-in-class CAB-CAR-T in China in 2017.

Fate Therapeutics

FATE is a leader in the use of small molecule pharmacologic modulators to optimize cell function, and has a collaboration with JUNO in the field of T cells. FATE is also a pioneer in the field of iPSC technology and has academic collaborations with UC San Diego for iPSC-derived NK cells and MSKCC for iPSC-derived T cells and CAR-T therapeutics. FATE's lead program is FATE-NK100, an adaptive memory NK cell that is being developed in collaboration with the University of Minnesota, initially in AML, but with plans to move into solid tumors starting with ovarian cancer.

Exhibit 42. FATE ACT Clinical Trials

Product	Target	Autologous or allogeneic	Lead Indications	Status
FATE NK-100	NA	Allogeneic	FATE and the University of Minnesota are conducting a phase 1 trial of NK100 in AML	Phase 1

Source: www.clinical trials.gov and Wells Fargo Securities, LLC

Formula Pharmaceuticals - Using NKT cells

Formula Pharmaceuticals is a U.S.-based company that is using the NKT cell as the base for CAR expression. NKT or cytokine-induced killer (CIK) cells are part of the innate immune system and offer a high-quality source of cells. Formula intends to collect PBMC from HLA matched or haploidentical donors and use non-viral transfection to introduce the CAR. The lead CIK-CAR targets CD19 and is in pre-IND testing, with intended use following stem cell transplant in ALL and NHL. In pre-clinical proof-of-concept studies are CIK-CAR.CD33 (AML) and CIK-CAR.PSMA (solid tumors).

GlaxoSmithKline (GSK) – Broad immunotherapy aspirations with investments in eTCR (ADAP), CAR-T (Miltenyi), and internal checkpoint program

GSK has two major collaborations in the field of adoptive cellular therapy (ACT) including (1) engineered TCRs with Adaptimmune (ADAP) and 2) CAR-T with Miltenyi Biotec. The first product in the ADAP collaboration is for an affinity-enhanced TCR to NY-ESO1, which is currently in several signal-seeking studies in solid tumors with a potentially registration-enabling trial in NY-ESO1+ve synovial sarcoma in HLA-A*02 patients, expected to begin mid-17. GSK and ADAP recently announced that GSK has selected Preferentially Expressed Antigen in Melanoma (PRAME) as the second of 5 targets.

The Miltenyi collaboration combines GSK's pre-clinical CAR-T candidates with Miltenyi's cell engineering capabilities.

iCell Gene Therapeutics -- Leader in T cell lymphoma

iCell Gene Therapeutics has a portfolio of pre-clinical autologous CAR-T therapies led by ICG122, a CAR-T targeting CD4 in the setting of T cell lymphoma. ICG122 has an Orphan Drug Designation for T cell lymphoma. ICG124 is a CAR-T targeting CD5 that is to be developed in the setting of T cell ALL.

Juno Therapeutics (JUNO) – One of two emerging ACT powerhouses founded on ACT technologies, collaborations with CELG, FATE, and EDIT

JUNO was founded with integration of adoptive cellular therapy (ACT) technology from the Fred Hutchinson Cancer Research Center (FHCRC), Memorial Sloan Kettering Cancer Center (MSKCC), and Seattle Children's Research Institute (SCRI). Since its founding, JUNO has made several acquisitions and entered into several collaborations, including a 10-year Master Research and Collaboration Agreement with Celgene (CELG) that gives CELG rights to exercise a global, ex-China, co-development/co-commercialization option on several programs including JUNO's CD19 and CD22 CAR-T candidates. CELG can also develop CAR-T /TCR products independently of JUNO within the collaboration with JUNO, retaining an option to co-develop and co-commercialize. CELG paid JUNO an up-front fee of $150.2MM and if CELG exercises options for both CD19 and CD22 during the initial opt-in window, an additional fee of $100MM would be paid; CELG recently opted to license the CD19 program paying a $50MM license fee to JUNO. Other notable collaborations include the following:

- Opus Bio – CD22 CAR-T
- FATE – small molecule modulators of engineered T cell products
- EDIT – gene-editing technology

Following several patient deaths and 2 clinical holds from the FDA, JUNO recently terminated development of the CD19 CAR-T product, JCAR015, in favor of focusing on its defined composition CD19 CAR-T products, led by JCAR017. JCAR017 was developed by SCRI and differs from JCAR015 in three major ways:

- JCAR017 uses CD137 (41BB) as the co-stimulatory domain vs. CD28;
- JCAR017 is a defined 1:1 mixture of CD4 and CD8 cells, i.e., it comprises two discretely manufactured products; and
- JCAR017 incorporates a truncated EGFR that binds cetuximab, which could be administered if rapid CAR-T removal was required.

JUNO is currently conducting a dose escalation trial of JCAR017 in DLBCL with a goal to initiate a registration enabling cohort once the recommended phase 2 dose has been determined. JUNO intends to initiate several other studies of JCAR017 in B cell malignancy in 2017, as well as continue and initiate clinical trials with other CD19 CAR-T products, including a fully human CD19, a 41BB ligand armored CD19 and JCAR014 with programmed death 1 inhibitor durvalumab and the combination of a CD19 CAR-T with adenosine 2. JCAR018 targets CD22 and is also being developed for B cell malignancy, where it may have a role in the setting of CD19 antigen loss following a CD19 CAR-T or as a stand-alone therapy; and JUNO believes that co-targeting CD19 and CD22 may reduce the risk for post-CAR-T relapse. Most recently, MSKCC has initiated development of a BCMA CAR-T, and JUNO expects FHCRC to initiate testing of a different BCMA CAR-T, also in 2017.

In solid tumors, JUNO collaborator SCRI has initiated clinical development of a L1CAM CAR-T for pediatric neuroblastoma (JCAR023), while MSKCC has initiated phase 1 testing of an IL-12 armored CAR targeting MUC16 in ovarian cancer (JCAR020). Separately JUNO has initiated clinical development of an ROR-1 CAR-T,

which may have use in both liquid and solid tumors. Most recently, JUNO has entered into a collaboration with Peter Macallum in Australia to evaluate a Lewis Y CAR-T in lung cancer. Also in 2017, JUNO intends to initiate clinical development of a CAR-T targeting IL-13R for glioblastoma.

Exhibit 43. JUNO and collaborator CAR ACT Trials

Company	Product	Target	Autologous or allogeneic	Lead Indications
JUNO	JCAR017	CD19	Autologous	Relapsed/refractory aggressive NHL with focus on diffuse large B cell lymphoma (n=144)
SCRI	JCAR017	CD19	Autologous	Relapsed/refractory pediatric acute lymphoblastic leukemia (PLAT-02 n=80) Relapsed/refractory pediatric acute lymphoblastic leukemia (PLAT-01 n=18)
NCI	JCAR018	CD22	Autologous	Relapsed/refractory adult/pediatric CD22+ve acute lymphoblastic leukemia including a CD1
SCRI	JCAR023	L1CAM	Autologous	Recurrent/refractory neuroblastoma (ENCIT-1 n=80)
FHCRC	JCAR014	CD19	Autologous	Relapsed/refractory B cell malignancy (n=125)
FHCRC	JCAR014 Durvalumab	CD19 PD-1	Autologous	Relapsed/refractory NHL (n=42)
JUNO	JCAR024	ROR-1	Autologous	ROR1+ve CLL, MCL in Cohort A or ROR1+ve NSCLC or TNBC in Cohort B
MSKCC	BCMA	BCMA	Autologous	Myeloma relapsed/refractory to IMiD/PI n=24
MSKCC	JCAR020	MUC16	Autologous IL-12 armed	3rd line + MUC16+ve ovarian cancer n=30

Source: company documents, www.clinical trials.gov and Wells Fargo Securities, LLC

Moving to eTCR therapeutics, JUNO is planning to file and IND for a Wilm's tumor 1 engineered TCR based on research being conducted at FHCRC. FHCRC is conducting a WT-1 TCR trial in hematologic malignancy and recently initiated a trial in WT-1+ve NSCLC/mesothelioma. In 2017, JUNO plans to file and IND of an eTCR for use in HPV+ve tumors.

Exhibit 44. JUNO and collaborator TCR ACT Trials

Product	Target	Autologous or allogeneic	HLA	Lead Indications	Status
JTCR016	WT-1		A*02		
WT1-TCRc4	WT-1	Autologous	A*02	HLA-A*0201 WT-1+ve 2nd line NSCLC	Phase 1/2
WT1	WT-1	Allogeneic	A*02	High risk or relapsed AML, MDS or CML	Phase 1/2

Source: company documents, www.clinical trials.gov and Wells Fargo Securities, LLC

Kite Pharmaceuticals (KITE) - One of two emerging ACT powerhouses founded on ACT technologies, acquired a European footprint with T cell factory acquisition, collaborations with AMGN and BLUE

KITE Pharmaceuticals was formed through key collaborations with the National Cancer Institute (NCI) to acquire CAR-T and TCR products and related technology. Axi-Cel is the first KITE-manufactured CAR-T product and the first CAR-T to report data from a potentially registration-enabling multicenter trial using centralized manufacturing. KITE intends to complete a BLA for Axi-Cel to treat relapsed/refractory aggressive NHL in 1Q17. In late 2016, KITE initiated a clinical trial with the company's first eTCR targeting MAGE A3/4.

KITE's lead program, Axi-Cel, is a CD19 targeting CAR-T that replicates the structure of the CAR developed by NCI, but uses KITE's manufacturing method and supply chain. Following a positive outcome in the ZUMA-1 trial, KITE expects to complete a rolling BLA 1Q17, with approval in the United States before YE17. The ZUMA-1 trial is being conducted in patients with aggressive relapsed or refractory lymphomas such as diffuse large B cell lymphoma (DLBCL), primary mediastinal B-cell lymphoma, or transformed follicular lymphoma. KITE intends to expand the Axi-Cel label with data from the ongoing ZUMA 2-4 trials, which include relapsed refractor mantle cell lymphoma (MCL), and both adult and pediatric acute lymphocytic leukemia (ALL). The company also recently initiated ZUMA-6 trial evaluates the combination of Axi-Cel with TECENTRIQ.

Exhibit 45. KITE and Collaborator CAR ACT Trials

Company	Product	Target	Autologous or allogeneic	Lead Indications	Status	Registration timeline
KITE	Axi-Cel	CD19	Autologous	Relapsed/refractory aggressive lymphoma (ZUMA-1, n=112) Relapsed/refractory mantle cell lymphoma (ZUMA-2, n=70) Relapsed/refractory adult acute lymphoblastic leukemia (ALL - ZUMA-3, =75) Relapsed/refractory pediatric ALL (ZUMA-4, n=75) Relapsed/refractory aggressive lymphoma combined with TECENTRIQ (ZUMA-6, n=31)	Phase 2 Phase 2 Phase 2 Phase 1	2017
NCI	CD19 CART - fully human	CD19	Autologous	Relapsed/Refractory B cell lymphoma Cohort 1 - No prior allo-HSCT Cohort 2 - prior allo-HSCT	Phase 1	

Source: www.clinical trials.gov and Wells Fargo Securities, LLC

In 2017, KITE intends to study Axi-Cel in chronic lymphocytic leukemia, follicular lymphoma and in 2nd line DLBCL. KITE is also intending to file an IND for the BCMA-targeting CAR-T, KITE-585 in 2017 and the CLL1 targeting CAR-T, KITE-796 in 2018. KITE-796 contains CDL's switch technology. KITE's partner NCI is testing a fully human CAR-T in the clinic, and KITE is developing a 3rd generation CD19 CART that also includes the CDL switch. Moving to engineered TCR therapies, KITE's partner NCI is testing two eTCR therapies against cancer testis antigen MAGE A3 and one against HPV+ve cervical cancer.

Exhibit 46. KITE and Collaborator TCR ACT Trials

Company	Product	Target	Autologous or allogeneic	Cellular Characteristics	HLA	Lead Indications	Status
NCI	MAGE A3	MAGE-A3	Autologous	CD4/8 mix	A*-01	MAGE-A3 solid tumors	Phase 1
NCI	MAGE A3	MAGE-A3	Autologous	CD4	DP0401/402	MAGE-A3 melanoma or other solid tumors	Phase 1
KITE	KITE-718	MAGE-A3/A6	Autologous	CD4/8 mix	A*02	MAGE-A3/A6 solid tumors	Phase 1
NCI	HPV-16 E7	HPV-16 E6/E7	Autologous	CD4/8 mix	A*02	HPV-16 E7 +ve cervical or head and neck tumors	Phase 1

Source: www.clinical trials.gov & Wells Fargo Securities, LLC

KITE has initiated clinical development of a NCI's Class II MAGEA3 eTCR. But unlike NCI, is transducing both CD4 and CD8 T cells. For HPV, KITE intends to take an HLA franchise approach, developing 4 or 5 products that are to target 90% of U.S. patients.

LBIO – leading the development of TIL

LBIO is the leading company in the commercial-scale development of TIL. LBIO has increased manufacturing capacity and development capabilities, and recently expanded the ongoing LN-144 TIL trial for melanoma. LBIO recently listed the first of two planned trials for LN-145, a TIL product derived from head and neck cancer, with a second trial in cervical cancer scheduled to start later in 2017. In 2016, LBIO entered into a collaboration with the Karolinska Institute, which is to lead the clinical development of TIL in the setting of glioblastoma and pancreatic cancer.

Exhibit 47. LBIO and Collaborator TIL ACT Trials

Product	Autologous or allogeneic	Lead Indications	Status
LN-144	Autologous	LBIO is conducting a TIL trial in recurrent/metastatic squamous cell carcinoma of the head and neck (n=47)	Phase 2
LN-145	Autologous	LBIO is conducting a TIL trial in metastatic melanoma (n=40) including comparison of fresh vs frozen product and re-treatment of patients relapsing with a new lesion using a new TIL product	Phase 2
TIL + YERVOY	Autologous	Moffitt is conducting a trial of TIL on the background of YERVOY (n=13)	Phase 1
TIL + OPDIVO	Autologous	Moffitt is conducting a trial of TIL on the background of OPDIVO, and evaluating the use of 4-1BBL during TIL manufacture (n=12)	Phase 1
TIL + KEYTRUDA	Autologous	NCI is conducting a trial of "young" TIL in a basket of solid tumors on the background of KEYTRUDA (n=290)	Phase 2

Source: www.clinical trials.gov and Wells Fargo Securities, LLC

Leucid Bio – Academic spin-out evaluating locally administered CAR-T for solid tumors

Leucid Bio is a Kings College London company that has formed around another CAR-T pioneer, John Maher. Maher has developed a CAR for targeting epidermal growth factor receptor (EGFR or erbB) members found in head and neck cancers. The promiscuous erbB ligand T1E binds to all 8 erbB homo or heterodimers, including EGFR and HER2. By combining T1E with CD28, Maher has developed a 2nd generation CAR-T that is being investigated in the clinic as a locally administered therapy for H&N cancer on the assumption that local administration reduces the potential for toxicity.

Exhibit 48. Leucid Bio/King's College London T4 ACT Trial

Product	Target	Autologous or allogeneic	CAR Design	Novel Features	Lead Indications	Status
T4	ErbB	Autologous	CD3ζCD28	IL-4	BioLucid is conducting a phase 1/2 trial of locally administered T4 in head and neck cancer n=30. Maher incorporates IL-4 receptor to drive CAR-T cells in the presence of tumor-associated IL-4.	Phase 1

Source: www.clinical trials.gov and Wells Fargo Securities, LLC

Mustang Therapeutics – academic spin-out

Mustang Therapeutics was formed to develop CAR-T therapeutics from City of Hope. The companies lead CAR-T therapeutic uses an IL-13 ligand, and not an scFv, to target IL-13 receptor 2α in patients with glioblastoma (GBM). A first generation intra-tumorally administered CD28 product showed evidence of safety and bioactivity, and a second generation 41BB CAR-T, hinge optimized version with 10-fold greater potency is being evaluated in an ongoing phase 1 trial. Mustang's 2nd product, MB-102 is a CD28 CAR-T targeting CD123 in AML/BPDCN.

Exhibit 49. Mustang Therapeutics CAR ACT Trials

Product	Target	Autologous or allogeneic	CAR Design	Novel Features	Lead Indications	Status
MB-101	IL13Rα2	Autologous	CD3ζ41BB	CD19t Hinge optimized	Mustang/CoH is conducting a phase 1 trial of a 2nd generation IL-13R2α specific CAR-T in 100 glioblastoma patients evaluating 3 weekly dose-escalated intratumoral/intracavitary infusions	Phase 1
MB-102	CD123	Autologous	CD3ζCD28	EGFRt Hinge Optimized	Mustang/CoH is conducting a phase 1 trial of a 2nd generation CD123 specific CAR-T in 60 AML/BPDCN patients who have an allo-HSCT donor	Phase 1

Source: www.clinical trials.gov and Wells Fargo Securities, LLC

NantKwest – NK cell focus

NK-U.S. is a company focused on engineering an immortalized NK cell line, NK-92, or aNK that has been evaluated in the clinic. NK-U.S. is currently conducting a trial of aNK with the tethered IL-15 product, ALT-803 from Altor Biosciences. The haNK product engineers NK-92 to increase CD16 and IL-2 expression, while taNK is to endow aNK cells with CARs. NK-U.S. expects to initiate clinical testing of a HER2 taNK in glioblastoma in Europe and in breast cancer in the United States in 2017.

Exhibit 50. NantKwest ACT Trials

Product	Target	Autologous or allogeneic	Lead Indications	Status
aNK ALT-803	NA	Allogeneic	NK is conducting a trial of aNK with ALT-803 in advanced Merkel cell carcinoma (n=24)	Phase 2
haNK	CD16	Allogeneic	NK is conducting a trial of haNK (aNK engineered for enhnace IL-2 and CD16) advanced solid tumors (n=16)	Phase 1

Source: www.clinical trials.gov and Wells Fargo Securities, LLC

Novartis (NVS) – the only "Big Pharma" invested in ACT

NVS is the first company to file a BLA for a CAR-T product, CTL019, for pediatric ALL. NVS is also the first company to conduct global clinical trials for CAR-T; and following completion of an ongoing trial in DLBCL, plans to file a Marketing Authorization Application in Europe for both pediatric ALL and DLBCL, and an sBLA in the United States for DLBCL.

In 2012, NVS entered into a global license with U Penn to develop CAR-T for cancer. NVS and U Penn established a new R&D facility at U Penn, the Center of Advanced Cellular Therapies, and NVS established an autonomous cell and gene therapy unit. To manufacture CAR-T, NVS acquired one of DNDN's PROVENGE facilities. In 3Q16, NVS eliminated 120 positions from the cell and gene therapy unit and integrated the unit into the existing R&D structure. In recent comments, NVS stated that the company was increasing investment in manufacturing sciences for CAR-T and noted that potential future products included CTL119, a CD19 CAR-T with a human scFv, a BCMA CAR-T, CTL019 combinations including checkpoint inhibitors, CD123 CAR-T for AML, mesothelin CAR-T for solid tumors, and an EGFRvIII CAR-T for glioblastoma. In addition, NVS is developing next-generation CAR-T incorporating regulated CAR-T products, i.e., switch controlled, and CRISPR-edited products for allogeneic, i.e., off-the-shelf use.

OGD2 Pharma – Targeting GD2 for Glioblastoma

OGD2 Pharma was spun out of the University of Nantes to develop therapeutics based on that University's research in O-acetyl-GD2 ganglioside, a tumor-associated antigen. GD2 has been validated by the approved anti-GD2, dinituximab; however, GD2 is present on peripheral nerve and dinituximab therapy is associated with peripheral nerve toxicity. By contrast, O-acetyl GD2 is not found on peripheral nerves. OGD2 Pharma is developing antibodies, anti-body drug conjugates, and with Green Cross LabCell, NK cells transduced with an OGD2 CAR.

Poseida Therapeutics

Spun out of Transposagen, Poseida is using the non-viral piggyBac system for gene delivery into T cells. Poseida's leading CAR-T products targets BCMA and PSMA, and are in IND-enabling and preclinical testing, respectively. Poseida uses centyrins as an alternative to scFvs for the target binding domains of its CARTyrin products.

Potenza Therapeutics – Drew Pardoll Checkpoint Company

Potenza was founded by Drew Pardoll and closed a series A financing round in December 2014 to develop checkpoint inhibitors. In April 2015, the company entered into an exclusive research and collaboration agreement with Astellas to develop a portfolio of checkpoint inhibitors. In 2017, Potenza and Astellas plan to file INDs to two targets, including one against T_{REG}.

Regeneron – Emerging Immuno-Oncology (IO) Player with Adicet Bio Collaboration

In 3Q16, REGN announced a strategic collaboration with Adicet Bio, a private company established by ex-KITE Pharmaceuticals CEO Aya Jakobovits and OrbiMed-led investors, around the assets of an Israeli company Applied Immune Technology (AIT). Under the terms of the 5-year collaboration agreement, REGN made an initial $25MM investment and plans to contribute research funding to generate multiple clinical candidate ACT therapies including fully human chimeric antigen receptor (CAR) and T-cell receptor (TCR) products in the field of hematologic and solid tumors.

AIT developed a universal immune cell therapy (uICT) and the REGN collaboration is to focus on off-the-shelf ACT products. AIT's most advanced technologies including the development of TCR-Like antibodies that recognize disease-specific peptides in the context of the major histocompatibility complex and Epitarget™, a proprietary technology to identify and validate novel disease-specific peptide targets. To compliment AIT's mining of internal peptide targets for use in uICT ACT products, REGN can leverage antibody and bispecific antibody targets being evaluated as part of the ongoing Sanofi immune oncology collaboration.

Shire (Baxalta) – Allogeneic CAR-T

Shire (SHP.L) acquired Baxalta in 2Q16, and with it, a collaboration with Precision Biosciences. Baxalta entered into a collaboration with Precision Biosciences in 1Q16 to develop up to 6 allogeneic CAR-T products, with clinical trials scheduled to begin in late 2017. Precision has presented data showing the use of synthetic nuclease-mediated removal of the TCR and HLA class I from a donor T cell.

Sorrento Therapeutics and NantKwest – CAR-T and NK- cell-based CARs

Sorrento established the TNK subsidiary following the acquisition of two private companies and their pre-clinical and clinical stage CAR-T therapies, which include fully human CAR-T against CD38 and CD123, with INDsU445235 planned for submission in 1H17. Separately, the TNK subsidiary has entered into two cell-therapy collaborations providing an NK platform for expression of Sorrento's CARs. The first collaboration, with NantKwest, is to use the proprietary NK92 NK cell line as a platform to express CARs with HER2 CAR, a PD-L1 CAR and a ROR1 CAR in pre-clinical development. Most recently, Sorrento and South Korean biotech CHA Biotech created a joint venture to develop and commercialize CAR-based products using CHA "activated NK" (AKC) cells. Separately, Sorrento gained commercialization rights to the AKC product, which requires a 3-week manufacturing process to produce sufficient cells for several treatment cycles. CHA notes that the company has produced more than 1,000 batches of AKC for use in ongoing clinical studies in Japan and that use of AKC does not require prior lymphodepletion. In 2016, NantKwest entered into a co-development agreement with Altor Biosciences covering the therapeutic application of Altor's ALT-801 and or ALT-803 targeting p53 and IL-15, respectively.

Takara Bio

Takara Bio is known as a supplier of RetroNectin, an ingredient used for retroviral transduction, with KITE and CLLS as notable licensees. Takara is also developing HF10, an oncolytic virus currently in phase 2 testing; however, in the area of Adoptive Cellular Therapy (ACT), the company has a collaboration with the University of Toronto and recently, that institution listed a phase 1 trial of an NY-ESO-1 targeted eTCR.

Tmunity

Tmunity is a spin out from Carl June at U Penn with a focus on novel TCR engineered cells, regulatory T cells, universal engineered T cell platforms, and CAR-T. Tmunity intends to develop products with best-in-class control over T cell activation and direction *in vivo*. In 1Q16, Tmunity raised $10MM in equity funding.

TxCell

TxCell is developing CAR-T_{REGs} to target autoimmune disease in addition to non-engineered T_{REG} products. The CAR-Treg pipeline includes transplant, dermatology, neurology, and lupus. In 2017, the company intends to produce proof of concept in the setting of transplant with collaborators at the University of British Columbia, and conduct a first in man study in 2018.

Unum/Seattle Genetics – Combining universal IO with external antibodies

Unum's concept for CAR-T therapy is to use a CD16 expressing CAR allowing for use with any antibody. The scientific founder of Unum is Dario Campana, who created one of the first CD19 CAR-T programs while at St Jude. Now at the National University of Singapore, Dr. Campana and colleagues transfect mRNA, allowing for a 1-week window of CAR expression in autologous T cells. These T cells are co-administered with RITUXAN in a first phase 1 trial currently being conducted in Singapore. Unum has recently initiated a similar trial in the United States, but using a durably expressed version of the CAR – ACTR087. Separately, Unum and SGEN have a collaboration covering 3 SGEN antibodies.

Exhibit 51. UNUM and Collaborator CAR ACT Trials

Company	Product	Target	Autologous or allogeneic	Lead Indications
U Singapore	ATTCK20	CD16	Autologous	Inviestigator sponsored trial in CD20+ve refractory hemtatologic malignancy with RITUXAN
Unum	ACTR087	CD16	Autologous	Relpased/ refractory CD20+ve hemtatologic malignancy with RITUXAN

Source: www.clinical trials.gov and Wells Fargo Securities, LLC

Ziopharm

ZIOP is combining the non-viral CAR-T expertise of MD Anderson, with switch and engineering technology from XON. ZIOP continues to support the legacy CD19 CAR-T work at MDACC, which is further optimizing Sleeping Beauty non-viral transfection. In 2017, ZIOP intends to initiate clinical development of an activated NK cell and a CD33 CAR-T in AML. ZIOP and NCI recently entered into a collaboration designed to move a point-of-care neoantigen TCR product into the clinic. Such products will likely incorporate ZIOP's membrane bound IL-15 and veledimex Rheostat technologies; and the company recently extended the Merck KGaA agreement to develop membrane bound IL-15 CAR-T products, with the first scheduled to enter the clinic in 2018.

ACT Clinical Trial Review

We have conducted a review of ACT trials that have been listed on the clinicaltrials.gov website since 2009, noting approximately 250 different trials. In the United States, academic institutions dominate CAR-T trials with University of Pennsylvania/ Children's Hospital, National Cancer Institute (NCI), Baylor College of Medicine, City of Hope, MD Anderson, Memorial Sloan Kettering, Fred Hutchinson/ Seattle Children's ,and recently, the University of North Carolina listing trials. Corporate CAR-T sponsors include BlueBird Bio, BLCM, KITE, JUNO, NVS, and ZIOP, and more recently, Chinese companies. Exhibit 52 summarizes selected CAR-T trials listed by a corporate sponsor, but does not include trials listed by an academic sponsor, even if the commercialization rights lie with a corporation.

Of the nearly 250 ACT trials we selected for review, more than 80 are targeting CD19, with more than 35 different sponsors. With the emergence of resistance to CD19 CAR-T therapy particularly in leukemia, several sponsors are testing alternative pan B cell markers, such as CD22 or CD20, either following CD19 CAR-T failure, or in combination with a CD19 CAR-T, with a view to increasing the barrier for resistance. Several CAR-T products are being evaluated with suicide switches in both liquid and solid tumors, and most are using BLCM's iCAS9.

Only two other targets, CD30 and mesothelin, reach double figures. Targets with 5 or more trials include diasialoangliside (mainly pediatric neuroblastoma), glypican-3 (mainly liver cancer), epithelial cell adhesion molecule (EpCAM), epidermal growth factor receptor, including variant 3, CD22, *HER2,* and B cell maturation antigen for myeloma. Currently, the majority of CAR-T trials are in liquid tumors. Several trials are targeting glioblastoma in particular, with a variant-3 specific EGFR CAR-T and in the pediatric setting of neuroblastoma, GD2. In Asia, liver cancer is a significant problem, and there are several CAR-T trial targeting antigens such as GPC3, and several trials are investigating local delivery in this tumor. While melanoma is a highly immunogenic tumor, most of the ACT trials are TIL or eTCR trials. Mesothelin is being evaluated across a basket of solid tumors.

Several trials are evaluating 3rd and 4th generation product that include dual co-stimulation, usually 41BB with CD20 or OX40 and/or additional features such as BLCM's iCas9 suicide switch or a variety of strategies to modulate the tumor microenvironment. BioLeucid/KCL's T4 uses a chimeric receptor to turn negative Il-4 signaling into a pro-T cell signal, while JUNO's partner has moved two 'armored" CARs into the clinic. A MUC16 CAR-T is endowed with the ability to secrete IL-12, while a CD19 CAR-T secretes 41BBL. Humanized/human scFvs are becoming more commonly incorporated into CARs, as is the use of checkpoint inhibitors and in particular, a PD-1 inhibitor, either an approved monoclonal antibody dosed with the CAR-T, or engineered into the T cell.

Corporate-Sponsored ACT Trial Review

A selected list of corporate ACT trials is presented in Exhibit 52. ADAP has INDs open for 4 TCR-based SPEAR products: NY-ESO1, MAGE-A10, alpha fetoprotein, and MAGE A3/4, all of which use ADAP" affinity optimization of the TCR. ADAP has faced challenges identifying eligible patients given the need for both the correct antigen and the correct HLA. ADAP has recently entered into a collaboration with MDACC, which allows the company to offer patients more than one protocol based on the results tumor analysis for more than one antigen. For example, an NSLC patient can be screened for three, rather than a single protocol. ADAP recently entered into a collaboration with BLCM, which combines BLCM's Go-TCR technology with ADAP's target expertise. BLCM recently initiated clinical development of a native TCR to PRAME, as well as a CAR to PSCA. The PRAME TCR includes BLCM's CaspaCIDe rimiducid activated suicide switch, while the PSCA CAR includes rimiducid activation of costimulatory domain. Later in 2017, BLCM's academic partner intends to initiate clinical testing of a CD19 CAR-T that incorporates CaspaCIDe in 2017; MDACC recently listed a CAR-NK trial that is to target CD19 and includes BLCM's CaspaCIDe. BLUE and partner CELG's bb2121 is the leading CAR-T targeting BCMA for myeloma. BCMA is emerging as the most competitive target for myeloma, with KITE, JUNO, NVS, and PFE also in the clinic or intending to enter the clinic; the largest BCMA CAR-T trial is being conducted by the Chinese biotech, Nanjing Legend, which initiated a 100-patient clinical trial in 3Q15, with the clinicaltrial.gov website stating a primary completion date of December 2017. CELG has long had a cellular therapy interest, which has recently embraced oncology. In this application, CELG's first in-house product is an NK cell produce derived from cord blood, PNK-007. CELG is testing PNK-007 in two clinical trials, one in AML, and one in myeloma. JUNO recently suspended development of JCA015 in adult ALL after concluding that clinical delays had negated a time advantage that JCAR015 held over JCAR017, a product JUNO considers to be more optimal. JCAR is rapidly expanding the ongoing evaluation of JCAR017 in DLBCL with a view to submitting a BLA in 2018. JUNO has several hypothesis generating trials ongoing with founding collaborators including FHCRC, which is evaluating JCAR014 with AZN's durvalumab and an ROR1 CAR-T, and MSKCC, which has recently initiated clinical testing of two "armored" CARs. One targeting the ectodmain of MUC16 contains an IL-12 gene to secrete low doses of IL-12, while a CD19 CAR-T can secrete

41BB ligand, ensuring the presence of a costimulatory signal in the TME. JUNO has interests in developing engineered TCR T cell products and has rights to FHCRC's WT-1 product. Moving forward, JUNO intends to use naturally occurring TCRs. KITE recently completed a rolling BLA for the first CD19 CAR-T to treat DLBCL, whereas rival NVS announced acceptance of the first BLA for a CAR-T by FDA 2 days earlier. DLBCL is the first of several refractory B cell malignancies for which KITE plans to evaluate AxiCel, and the AxiCel development program also includes combination therapy with Roche's TECENTRIQ, as well as a plan to evaluate AxiCel in earlier stage patients. Like JUNO, KITE has a plan to develop TCR-based products using naturally occurring TCRs. KITE's academic partner, NCI has conducted several TCR trials, including HPV+ve cervical cancer. As noted above, NVS is the first company to have a BLA accepted by the FDA for a CAR-T. CTL019 is a CD19 CAR-T and NVS is seeking approval for pediatric ALL based on positive data from the ELIANA trial; later in 2017, NVS plans to seek approval for DLBCL based on the outcome of the JULIET trial. NVS's key collaborator is U Penn, which has tested several CAR and TCR products in the clinic; however, NVS has yet to list a clinical trial for a non-CAR-T product. JUNO, KITE, and NVS have all developed human or humanized CD19 scFvs, which the companies expect to reduce immunogenicity associated with first generation murine-based scFv products. While CLLS has received clearance from the FDA to begin clinical testing of a CD123 CAR-T in the United States, the company's first clinical candidate, UCART19, is in clinical trials in Europe with Servier, and Servier's U.S. partner for UCART19 soon plans to add U.S. sites to the adult ALL trial. While CLLS is the leading company developing allogeneic OTS CAR-T products, Unum Therapeutics has embraced a universal CAR-T approach that is intended to harness cellular therapy with existing monoclonal antibodies. By using a CD16 CAR, Unum can co administer a CAR-T with an antibody such as RITUXAN. A recent newcomer to the clinical field of ACT is the Japanese company Takara Bio, which is well known in the cell production field as a supplier of key ingredients. Takara's first cell therapy being tested in the United States is an NY-ESO1 TCR, which was developed using the company's proprietary technology. LBIO is the leading TIL company, and recently initiated a trial in its second indication, squamous head and neck cancer. LBIO has a key strategic relationship with the Moffitt Cancer Center, which is conducting small proof-of-concept trials combining TIL with checkpoint inhibitors.

FATE, NK-U.S., and ZIOP are the leading proponents of allogeneic NK-based therapies that offer an opportunity for off-the-shelf supply. NK-U.S. is developing products based on an immortalized cell line, NK)92, which is irradiated prior to administration. Once irradiated, the product has an in-vivo life of approximately 3 days. NK-U.S. refers to this product as an activated NK, aNK, and is currently evaluating aNK in Merkel cell carcinoma, in combination with Altor Biosciences' IL-15 product, ALT-803. In engineering aNK with IL-2 and CD16, NK-U.S. has developed haNK and with a CAR tank. FATE has recently received clearance from the FDA to initiate clinical testing of NK-100 with its partner the University of Minnesota (UM). FATE Nk100 is designed to mimic the characteristic of a subset of NK cell described by UM to be associated with improved outcomes in AML. ZIOP also intends to develop an OTS NK product from donors, noting that its key collaborator, MDACC, has considerable experience with NK-cell approaches; the aforementioned MDACC CD19 NK-CAR includes a tethered IL-15, which ZIOP believes may obviate the need for lymphodepleting chemotherapy.

Several Chinese companies are investigating CAR-T therapies, primarily in China, although some have become listed in the United States and intend to expand clinical development into the United States. One such company is CBMG, currently evaluating CD19 CAR-T in both DLBCL and ALL. PersonGenBio has a number of ACT products in the clinic, including CD19 and CD30 CARs for hematology, a MUC1 CAR in solid tumors, and three CAR-NK products, one targeting MUC1, one CD33, and one CD7. Sinobioway Cell Therapy and Shanghai GeneChem also have several different ACT products in the clinic; interestingly, Shanghai GeneChem is evaluating a CD19 CAR-T in the setting of systemic lupus erythematosus.

Exhibit 52. CAR-T trials listed by Corporate Sponsor

Sponsor/ Collaborators	Product	Conditions	Target	Phase	N=	Start Date	End Date	Endpoint	Summary	Recruitment
ADAP	MAGE-A10 SPEAR	Melanoma, urothelial of H&N cancers MAGE A10 +ve	MAGE-A10	1	22	10/16	11/19	Safety	Flu/Cy followed by MAGE-A10 SPEAR - HLA-A*0201/06	Recruiting
ADAP	MAGE A10 SPEAR	NSCLC - MAGE A10 +ve	MAGE-A10	1/2	32	12/15	11/17	DLT	Cy followed by 3 dose cohorts: 0.1x10^9, 1x10^9 and 5x10^9 - HLA-A*0201/06	Recruiting
ADAP	NY-ESO-SPEAR	Multiple myeloma	NY-ESO1	1/2	26	3/11	4/21	Safety	NY-ESO SPEAR(>0.1-1x10^10) post auto-SCT - HLA A*02	Active, not recruiting
ADAP	NY-ESO-SPEAR	NSCLC - NY-ESO1 +ve	NY-ESO1	1/2	10	10/15	11/17	Safety	Cy followed by 5x10^9 NY-ESO SPEAR - HLA A*02	Recruiting
ADAP	NY-ESO-SPEAR	Sarcoma - myxoid/ round cell	NY-ESO1	1/2	15	11/16	5/18	CR rate	Flu/Cy followed by NY-ESO SPEAR 1-8x10^9 - HLA A*02	Recruiting
ADAP	NY-ESO SPEAR	Sarcoma - synovial	NY-ESO1	1/2	65	3/11	10/17	CR/PR	Flu/Cy high NY-ESO1, Flu/Cy low NY-ESO1, Cy high NY-ESO1, RD Flu/Cy high NY-ESO1	Recruiting
Beijing Bio healthcare, China	CART20	Non-Hodgkin lymphoma	CD20	1/2	15	8/16	1/18	All cause mortality		Recruiting
Beijing Doing Biomedical, China	CD19 CART	B cell malignancy	CD19	1	100	1/16	10/18	Safety	Lymphodepletion followed by anti-CD19 CAR-T	Not yet recruiting
Beijing Doing Biomedical, China	allo-CD19 CARγδT	B cell malignancy	CD19	1	48	1/16	1/19	Safety	Lymphodepletion followed by allo-CD19 CARγδT cells	Not yet recruiting
Beijing Sanwater Biological Technology, China	CART-19	Acute lymphocytic leukemia	CD19	1	20	5/16	12/17	Safety	Lymphodepletion followed by 2-5x10^6/Kg to 2.5x10^8/Kg	Recruiting
BLCM	BPX-701	AML - PRAME+ve	PRAME	1	48	3/17	12/19	MTD	Dose escalation de-escalation of BPX-701 with iC9 rimiducid safety switch - HLA-A*02	Recruiting
BLCM	BPX-601	Pancreatic cancer	PSCA	1	30	11/16	12/19	MTD	BPX-501 dosing will be followed by rimiducid to induce CAR-T activation	Recruiting
BLUE	bb2121	Multiple myeloma - BCMA+ve	BCMA	1	50	1/16	12/18	Safety	Flu/Cy followed bu dose escalation bb2121	Recruiting
Carsgen Therapeutics, China	CAR-GPC3	Squamous Cell Lung	Glypican 3	1	20	3/16	10/17	Safety	Flu/Cy followed by 1x10^5 - 2x10^9 CAR-GPC3 T cells	Recruiting
CELG	PNK-007	Acute myeloid leukemia	NA	1	28	8/16	12/19	DLT	Flu/Cy followed by allo-NK (PNK-007) at one of 4 dose levels and IL-2	Recruiting
CELG	PNK-007	Multiple myeloma	NA	1	40	1/17	11/18	DLT	auto-SCT followed d14 by dose escalated allo NK (PNK-007)	Recruiting
Cell Medica	CMD-003	Lymphoma - EBV+ve Post-transplant lymphoproliferative disorder	Epstein Barr virus	2	70	11/17	12/17	ORR	3 cohorts of patients: DLBCL, HL or PTLD receive up to 5 doses of EBV-specific T cells (2x10^7/m^2) as 2nd line treatment	Recruiting
Cell Medica	CMD-003	Lymphoma - EBV	Epstein Barr virus	2	30	9/17	12/17	ORR	Patients with EBV+ve extranodal NK/T-cell lymphoma receive 2x10^7/m^2 days 1 & 15 with 3 additional infusions weeks 8, 12 and 24	Recruiting
Cell Therapy Catapult	WT1 TCR	AML/MDS - WT1+ve	WT-1	1/2	25	9/15	12/18	Safety	Lymphodepletion followed by WY-1 TCR (≤2 x10^7/Kg) followed by IL-2 - HLA-A*02	Recruiting
Cellular Biomedicine Group (CBMG)	C-CAR011	Acute lymphocytic leukemia	CD19	1	20	1/17	8/17	DLT	3 dose levels, 0.5x10^6, 1.5x10^6 or 3x10^6/Kg	Recruiting
Cellular Biomedicine Group (CBMG)	C-CAR011	Diffuse large B cell lymphoma	CD19	1	15	12/16	11/17	DLT	3 dose levels, 0.8x10^6, 2.5x10^6 or 5x10^6/Kg	Recruiting
Celyad	NKR-2 CART	Acute myeloid leukemia	NKG2D	1	12	3/15	3/17	Safety	1x10^6, 3x10^6, 1x10^7 or 3x10^7 cells	Active, not recruiting
Celyad	NKR-2 CART	Solid tumors (CRC, ovarian, bladder, TNBC and pancreatic), AML or MM	NKG2D	1	24	12/16	5/20	Safety	NKR-2 CAR will be dose escalated with each dose given every 2 weeks for a total of 3 infusions	Recruiting
FATE	NK100	Acute myeloid leukemia	NA	1	29	5/17	1/21	MTD	Flu/Cy followed by 1x10^7-1x10^8/kg	
Hebei Senlang Biotechnology, China	CD19 CART	B cell malignancy	CD19	1/2	10	6/16	6/17	ORR	Lymphodepletion followed by 3rd gen CD28/41BB CART	Recruiting
Immatics	IMA101	Solid tumors	ACTolog Ag	1/2	31	10/16	9/18	Safety	HLA A*02 patients receive CD8 IMA101 product following Flu/Cy if their tumor contains a relevant antigen	Not yet recruiting
Innovative Cellular Therapeutics, China	CD19 CART	B cell malignancy	CD19		30	6/15	1/21	Safety	Cy/Flu followed by dose escalated CD19 CART (1x10^5 - 1x10^7/Kg)	Recruiting
JUNO	JCAR015	Acute lymphocytic leukemia	CD19	2	110	8/15	5/18	ORR	Flu/Cy followed by JCAR015	Suspended
JUNO	JCAR017	Non-Hodgkin lymphoma	CD19	1	144	12/15	1/18	Safety	Flu/Cy followed by JACAR017 as a single or double dose	Recruiting
JUNO	JCAR014 Durvalumab	Non-Hodgkin lymphoma	CD19 PD-1	1	42	12/16	12/19	AUC JCAR014	Flu/Cy followed in group 1 by JCAR014 d0 and durvalumab d28 and every 4 weeks for up to 10 doses or in group 2 durvalumab d-28 and d-1 followed JCAR014 d0 and	Recruiting
KITE	AxiCel	Acute lymphocytic leukemia	CD19	1/2	75	11/15	3/17	Safety/ORR	Flu/Cy followed by 2x10^6/Kg	Recruiting
KITE	AxiCel	All - pediatric	CD19	1/2	75	12/15	6/17	Safety/ORR	Flu/Cy followed by 2x10^6/Kg	Recruiting
KITE	AxiCel	Diffuse large B cell lymphoma	CD19	1/2	142	1/15	3/17	Safety/ORR	Flu/Cy followed by 2x10^6/Kg	Recruiting
KITE	AxiCel	Mantle cell lymphoma	CD19	2	70	11/15	9/17	ORR	Flu/Cy followed by 2x10^6/Kg	Recruiting
KITE	AxiCel TECENTRIQ	Diffuse large B cell lymphoma	CD19 PD-L1	1/2	31	9/16	6/18	Safety/CR	Flu/Cy followed by 2x10^6/Kg and TECENTRIQ	Recruiting

Source: www.clinicaltrials.gov

Exhibit 52. CAR-T trials listed by Corporate Sponsor - continued

Sponsor/ Collaborators	Product	Conditions	Target	Phase	N=	Start Date	End Date	Endpoint	Summary	Recruitment
LBIO	LN-145	Head & Neck (Squamous)	NA	2	47	3/17	10/18	Safety	Lymphodepletion followed by TIL and IL-2	Recruiting
LBIO	LN-144	Melanoma	NA	2	40	9/15	1/18	Safety	Lymphodepletion followed by TIL and IL-2	Recruiting
Nanjing Legend Bio, China	LCAR-B38M-02	Multiple myeloma	BCMA	1/2	100	10/15	12/17	Safety	LCAR-B38M dosed at 0.5-5x10^6/Kg split over days 0, 2 & 6	Not recruiting
NantKwest	NK-92	Acute myeloid leukemia	NA	1	18	5/14	12/16	Safety	1x10^9, 3x10^9 or 5x10^9/m2 with a 2nd infusion 24h after the first if well tolerated	Active, not recruiting
NantKwest	haNK	Solid tumors	NA	1	16	4/17	12/19	MTD	aNK (NK-92 with IL-2 and CD16) dosed at 2x10^9 or 4x10^9	Not yet recruiting
NantKwest	NK-92 ALT-803	Merkel cell	NA IL-15	2	24	8/15	12/17	PFS	NK-92 dosed at 2x10^9/m^2 day 0 & 1 every 2 weeks with ALT-803 (IL-15) d0 of every cycle	Recruiting
NVS	CTL019	All - pediatric	CD19	2	67	8/14	8/22	ORR	Flu/Cy followed by 2-5x10^6 up to 2.5x10^8	Recruiting
NVS	CTL019	Diffuse large B cell lymphoma	CD19	2	130	7/15	1/24	ORR	Lymphodepletion followed by CTL019	Recruiting
PersonGen Bio, China	PCAR-019	B cell malignancy	CD19	1/2	10	7/16	11/18	Safety		Recruiting
PersonGen Bio, China	PCAR-019	B cell malignancy	CD19	1/2	10	10/16	9/18	Safety		Recruiting
PersonGen Bio, China	PCAR-119	B cell malignancy	CD19	1	10	9/16	9/18	Safety	Patients receive NK-92 modifed with a CD19 CAR ahead of SCT	Recruiting
PersonGen Bio, China	CAR-pNK	Acute myeloid leukemia	CD33	1	10	10/17	9/17	Safety	Patients receive NK-92 modifed with a CD33 CAR	Recruiting
PersonGen Bio, China	CAR-pNK	B cell malignancy	CD7	1/2	10	3/16	3/17	Safety	Patients receive NK-92 modifed with a CD7 CAR	Recruiting
PersonGen Bio, China	anti-MUC1 CART	Glioma, gastric or CRC - MUC1+ve	MUC1	1/2	20	10/15	10/17	Safety		Recruiting
PersonGen Bio, China	anti-MUC1 CART	HCC, NSCLC, pancreatic or TNBC - MUC1+ve	MUC1	1/2	20	10/15	10/17	Safety		Recruiting
PersonGen Bio, China	CAR-pNK	Solid tumors - MUC1+ve	MUC1	1	10	7/16	7/17	Safety	Patients receive NK-92 modifed with a MUC1 CAR	Recruiting
Servier	UCART19	All - pediatric	CD19	1	10	6/16	7/19	Safety	CAMPATH lymphodepeltion followed by dose escalated UCART19 ahead of a planned allo-SCT	Recruiting
Servier	UCART19	B cell malignancy	CD19	1	12	8/16	6/18	Safety	CAMPATH lymphodepeltion followed by dose escalated UCART19	Recruiting
Shanghai GeneChem, China	CD19 CART	Acute lymphocytic leukemia	CD19	1/2	30	1/16	6/18	Safety	Flu/Cy followed bu 1-5x10^6 CAR-T/Kg	Recruiting
Shanghai GeneChem, China	CD19 CART	Systemic Lupus Erythematosus (SLE)	CD19	1	5	3/17	1/18	Safety	SLE patients receive Cy followed by 1x10^6-1x10^7 CD19 CAR-T	Recruiting
Shanghai GeneChem, China	TAI-GPC3-CART	Hepatocellular Carcinoma	Glypican 3	1/2	30	3/16	7/18	Safety	Cy followed by transarterial infusion of 1-10x10^6 GPC3-CART	Recruiting
Shanghai GeneChem, China	meso-CART	Pancreatic Cancer	Mesothelin	1	30	3/16	2/18	Safety	Cy followed by transarterial infusion of 1-10x10^6 GPC3-CART	Recruiting
Shanghai Unicar-Therapy, China	CD19 CART	ALL - CNS disease	CD19	1	10	8/16	8/17	CR rate		Recruiting
Sinobioway Cell Therapy, China	GD2 CART	Neuroblastoma	Disialogangli oside (GD2)	1/2	22	9/16	9/20	Efficacy	Cytoreductive chemotherapy followed bu GD2-CAR-T days 0, 1, 2, 29 & 30.	Recruiting
Sinobioway Cell Therapy, China	EPCAM-CART	Acute lymphocytic leukemia	EPCAM	1/2	24	11/15	11/19	DCR	CAR-T dosed d1, 2 & 29.	Recruiting
Sinobioway Cell Therapy, China	EPCAM-CART	Diffuse large B cell lymphoma	EPCAM	1/2	24	7/15	7/19	ORR	CAR-T dosed d0, 1, 4, 7, 28, 31 & 34.	Recruiting
Sinobioway Cell Therapy, China	EPCAM-CART	Hepatocellular Carcinoma	EPCAM	1/2	25	11/15	11/19	DCR	CAR-T dosed d0, 1, 2, 28 & 29.	Recruiting
Sinobioway Cell Therapy, China	EPCAM-CART	Stomach	EPCAM	1/2	19	11/15	11/19	DCR	CAR-T dosed d0, 1, 2, 28 & 29.	Recruiting
Takara	TBI-1301	Solid tumors - NY-ESO1 +ve	NY-ESO1	1	12	3/15	3/17	Safety	Cy or Flu/Cy followed by 5x10^8 or 5x10^9 TBI-1301	Recruiting
Takara	TBI-1301	Solid tumors - NY-ESO1 +ve	NY-ESO1	1	15	9/16	3/18	Safety	Cy followed by TBI-1301	Recruiting
Unum	ACTR087 RITUXAN	Lymphoma	CD16 CD20	1	54	8/16	1/20	Safety	ACTR087 dosed with RITUXAN	Recruiting

Source: www.clinicaltrials.gov

Hematology and specifically, CD19+ve B cell malignancy, are the most common application for ACT. Targeting CD19 with a CAR-T removes both malignant and healthy B cells, and with pharmacologic antibody replacement, B cell aplasia can be managed. In fact, the loss of B cell aplasia is often a harbinger of disease relapse in ALL, at least. While most of the knowledge on loss of response to CAR-T therapy comes from ALL, there is a common acceptance that mule-antigen targeting will likely be required both to increase efficacy and to reduce relapse. The latest estimates in ALL predict 30% loss of response in patients achieving remission while maintaining B cell aplasia due to the loss of the target antigenic site on CD19; such a phenomenon has not been described on healthy B cells. In the setting of antigen loss, NCI has shown the ability to rescue patients using an anti-CD22 CAR-T; however, downregulation of CD22 is associated with loss of response and

NCI intends to initiate clinical development of a dual CD19/CD22 CAR-T. The Medical College of Wisconsin recently listed a clinical trial to evaluate a tandem CD19/CD20 CAR-T, NCT03019055, and at least 3 Chinese academic institutions are testing combination of CD19/CD20 or CD19/CD22 CAR-T therapies.

While loss of B cells can be managed, the same cannot be said about either T cells, or myeloid cells where their loss is considered incompatible with life. T cell malignancy is also a particular challenge for ACT as the workhorse for ACT is the T cell and thus, growing T cells to kill T cells is not feasible without engineering. CLLS has developed strategies to develop CARs that target antigens found on T cells by removing those antigens from the T cell before the addition of a CAR. Myeloid malignancies such as AML can be targeted with a CAR-T; but the question for the field is, can it be done without stem cell rescue? Certainly use of a CAR-T as a bridge to transplant would be welcomed as a treatment for AML, but with CAR-T being considered as a potential strategy to obviate the need for stem cell transplant, strategies to develop safe but effective CAR-T for AML are of high interest.CD33 is the index antigen for AML based on its use as a target for Mylotarg and indeed, ZIOP is intending to go into the clinic with a CD33 CAR-T in 2017; several Chinese investigators are already in the clinic with a CD33 CAR-T. CD123 is also an antigen of high interest given that the differential in expression between leukemic and healthy stem cells may be larger than that for CD33. Two of the pioneers in the field of ACT, U Penn and City of Hope, have approached a CD123 CAR-T differently. U Penn is using a biodegradable RNA CAR, while City of Hope is evaluating a standard DNA CAR, and both CARs use the same scFv. CD123 is a pan-myeloid/erythroid cell marker and theoretically, a patient might require continuous stem cell support until the CD123 CAR was eliminated. U Penn's approach to targeting antigens shared by healthy tissue is to evaluate an RNA CAR describing these CARs as biodegradable. In pre-clinical studies, U Penn has shown striking tumor response data for a CD123 CAR, but with considerable on-target/off-tumor toxicity, Gill et al., Blood 123 (15), 2014. Pre-clinical data for the City of Hope CAR appear to suggest that their conventional CD123 CAR-T killed AML blasts, but did not eliminate granulocyte/macrophage of erythroid colony formation *in vitro*, Mardiros *et al.,* Blood, 122 (18) 2013. City of Hope is conducting trials with a DNA CD123 CAR-T that includes the truncated EGF receptor, which allows for ERBITUX-mediated clearance of T cells in the event of toxicity. While head-to-head data for the 41BB U Penn CAR and the CD28 City of Hope CAR are not available, Gill and colleagues claim superior efficacy for the U Penn CAR based on an absence of long-term survivors in the City of Hope AML model, which is in contrast to the presence of long-term survivors in the U Penn AML model; However, Gill comments that the use of pre-clinical models to select the optimal CAR design, may result in the selection of overly potent CARs.

At the 2016 American Association for Cancer Research (AACR) meeting, CLLS presented pre-clinical data for UCART123, suggesting that healthy hematopoietic progenitors were protected, but AML was not. European researchers have reported a similar observation for CD30 CAR-T, Hombach *et al.*, Mol Ther. 2016 Apr 26. As with CD123 targeting, CD30 targeting may also be limited by unwanted targeting of lymphocytes and hematopoietic stem and progenitor cells (HSPC), which express CD30 during activation. Hombach and colleagues believe that HPSCs are protected against CD30 CAR-T mediated killing by expressing a substantially lower level of CD30 than lymphoma cells and a higher level of a granzyme B inactivating enzyme, which increases further during activation; granzyme B is an enzyme used by T cells to induce apoptosis in target cells. An alternative approach to reducing toxicity toward HSPC while maintaining efficacy toward cancer cell targets was described by Cooper and colleagues: PL0S One. 2016 Aug 22;11(8):e0159477. Cooper evaluated mixing and matching heavy and light chain domains from different antibodies targeting CD123 to create novel scFv domains on CARs, noting that 1 pair showed reduced toxicity to HSPC without diminished toxicity toward AML blasts. Recently, KITE described the antigen CLL1, which is present on leukemic and healthy progenitors and not stem cells. KITE intends to develop a CAR-T to CLL1, which incorporates Cell Design Labs switch technology, which could provide KITE with an option to tune the level of CAR-T activity to eliminate AML, while maintaining sufficient normal hematologic activity.

Solid Tumor ACT Trials

Unlike early and profound success in liquid tumors, CAR-T therapy has yet to show a significant impact in solid tumors. In fact, the only published and peer-reviewed profound response to a CAR-T was reported recently by the City of Hope/Mustang Therapeutics group for an IL-13R2a-based CAR in glioblastoma. In contrast, engineered TCR and TIL therapies have shown efficacy in solid tumors, most notably in the settings of NY-ESO1 positive sarcoma, ADAP/NCI and melanoma – NCI/LBO.

One of the biggest differences conceptual challenges to extending the success of CD19 CAR-T into solid tumors is access to antigen. For CD19, both healthy and diseased cells provide the CAR-T with CD19, and the bone marrow is producing new CD19+ve cells daily. Even here, the Seattle Children's Research group's next CD19 CAR-T trial in pediatric ALL is to include an engineered CD19+ve T cell to act as a circulating adjuvant in the setting of low CD19. For solid tumors, engineered T cells are to have to find their target, which is likely located in a site shielded by a hostile tumor microenvironment. Keeping solid tumor targeting T cells alive long enough to locate and access their target will likely require a different strategy that has been successful in liquid tumors.

We use U Penn's experience with mesothelin CAR-T to juxtapose that institution's experience with a solid and a liquid tumor CAR-T. Mesothelin is being evaluated in over 10 ACT trials including U Penn, which has evaluated a biodegradable RNA CAR-T and two DNA CAR-T products, an earlier product with a murine scFv and a more recent product with a human scFv. U Penn initially investigated an electroporated RNA CAR to mesothelin, but persistence was very short and a second trial was initiated using a conventional lentiviral DNA CAR with or without cyclophosphamide lymphodepletion. At the 2015 American Association of Cancer Research and 2016 ASCO meetings, U Penn presented preliminary data from the phase 1 DNA mesothelin CAR-T trial dosed at 1-3 $x10^7$ CAR-T/m^2. U Penn noted that in comparison to their experience with a CD19 CAR, expansion of the mesothelin CAR was approximately 2 logs lower and persistence much shorter. In 6 ovarian cancer patients a best response of stable disease was noted by day 28 in all 6 patients, including one subject who cleared all malignant cells from their pleural effusion. No cases of Cytokine Release Syndrome (CRS) or macrophage activation syndrome were observed. Again persistence was short, with CAR-T cells detected up to 28 days in peripheral blood and 36 days in pleural effusion. Trafficking of the mesothelin CAR-T cells was noted in both on-tumor and off-tumor sites, but without on-target toxicity in the off-tumor sites. In addressing persistence, we note that BLCM as an early mover with its iMC construct, which allows for small molecule induction of survival signals downstream of the iMC following rimiducid dosing.

We have reviewed the challenges to antigen selection in the setting of solid tumors earlier in this report. One promising target is EGFRvIII since it is found only in the setting of cancer and given the high unmet need on glioblastoma, several CAR targeting EGFRvIII have entered the clinic. However, to date, data have been disappointing, perhaps reflecting in part, heterogeneity of EGFRvIII expression. Similarly, the disialoganglioside, GD2 has been an antigen of high interest in pediatric neuroblastoma and Baylor, for example, has conducted an evolving series of trials that currently include a trial of a 3rd generation CAR-T using OX40 and CD28 co-stimulation and KEYTRUDA.

While the United States dominates CAR-T trial listings, 30% of the trials are being conducted in China, and as in the United States, both academic and corporations are sponsoring trials. In China, 4 companies are sponsoring CAR-T trials, including Beijing Doing Biomedical (CD19), PersonGen Biomedicine (Mucin 1), Shanghai GeneChem (CD19), and Sinobioway Cell Therapy (epithelial cell adhesion molecule -EPCAM), Exhibit 53. Recently, Chengdu MedGenCell and Sichuan University listed a PD-1 knockout clinical trial in non-small cell lung cancer (NSCLC) using autologous peripheral blood mononuclear cells (PBMCs) as a source and CRISPR as the gene modification technique, NCT02793856.

One way to limit on-target off-tissue toxicity is local administration, and such an approach has and continues to be evaluated in primary liver cancer and liver metastases, where local therapy is already a standard of care. Glypican 3 and carcinoembryogenic antigen are both being evaluated as local therapy. At the 2017 AACR, Kings College London presented data for their *erbB* targeting localized CAR-T (T4) in 10 advanced head and neck cancer patients. In this trial, KCL evaluated $1x10^7$, $3x10^7$ or $1x10^8$ one of three dose levels of T4 administered locally without prior lymphodepletion. One patient in cohort 2 died prior to receipt of his product and a second patient in cohort 2 died prior to the day 43 evaluation, suggesting that the patients enrolled were very advanced. A best response of stable disease was noted in 5 patients, including all 3 dosed at the $1x10^8$ level.

Another way to limit off-target toxicity is to target antigens that are not found on healthy tissue, or only on tissue in an immune privileged site. Targeting of cancer testis antigens such as MAGE and NY-ESO1 has attracted considerable attention, and both ADAP and KITE/NCI have significant efforts against these antigens. NY-ES01 is arguably the index antigen for a TCR approach; however, as noted by ADAP, not all NY-ESO1 peptides that have been reported to be targets in the literature are, in their experience, actually presented by cancer cells.

Exhibit 53. Selected Solid Tumor ACT Trials

Sponsor/ Collaborators	Product	Indication	Target	Phase	N=	Start Date	End Date	Endpoint	Summary	Recruitment
2nd Military Medical University, China	DC-PNAT	Biliary Tumor	Neo-Ag	1/2	40	9/15	3/17	OS	Gemcitabine (1/wk x 6) vs gemcitabine plus DC (1x10^7 DC days 19/20, 40/41 & 61/62) + personalized neoantigen (1-6x10^9 on days 21, 42 & 63)	Recruiting
Uni. Penn	cMET RNA CART	Breast Cancer	cMet	1	15	4/13	4/17	Safety	Intratumoral CAR-T in cMet+ve tumor	Active, not recruiting
Fuda Cancer Hospital, China	CAR-T	Breast Cancer	HER2	1/2	60	9/15	3/16	Safety		Recruiting
Memorial Sloan KCC	meso-CART	Breast cancer - meso+ve	Meso-thelin	1	24	6/16	6/19	MTD	Cy followed by dose escalated meso-CAR-T incorporating iCas9	Recruiting
2nd Military Medical University, China	PIK-HER2	Gastric/liver - HER2+ve	HER2	1/2	40	9/15	3/17	OS	DC (1x10^7 on days 19/20, 40/41 & 61/62) + precision multiple antigen T cells (1-6x10^9 on days 21, 42 & 63) vs DC (1x10^7 on days 19/20, 40/41 & 61/62) + pluripotent killer T cells expression anti-HER2 (1-6x10^9 on days 21, 42 & 63)	Recruiting
Beijing Sanbo Brain Hospital, China	EGFRvIII CART	Glioblastoma	EGFRvIII	1	20	7/16	7/18	safety	Flu/Cy followed by dose escalated EGFRvIII CAR-T from 5x10^4 - 1x10^7/Kg slit over 3 days	Recruiting
Duke	EGFRvIII CART	Glioblastoma	EGFRvIII	1	48	2/17	2/20	MTD	Newly-diagnosed patients receive EGFRvIII prior to chemoradiation (CRT). In patients with stable disease post-CRT temodar will be started with EGFRvIII CAR-T will be added during cycle 3; dose-escalation cohorts are 4.5x10^6, 1.5x10^7, 4.5x10^7 and 1.5x10^8/Kg	Recruiting
National cancer Institute	EGFRV3 CART	Glioblastoma	EGFRvIII	1/2	107	9/11	12/18	Safety	Flu/Cy followed by EGFRV3 CART and IL-2	Recruiting
Baylor CoM	iCAR	Glioblastoma	HER2	1	14	2/16	2/18	Safet	Intratumoral/intracavitary administration at 3 dose level: 1x10^7, 3x10^7 and 5x10^7 x2 on consecutive days	Recruiting
City of Hope	IL13Ra2 CART	Glioblastoma	IL13Ra2	1/2	100	5/15	12/18	Safety	IL13Ra2 CAR-T administered intratumoral, intracaritary, intraventricular or intratumoral and intraventricual once every 3 weeks	Recruiting
Beijing Sanbo Brain Hospital, China	Anti-PD-L1 CSR	Glioblastoma	PD-L1	1	20	7/16	7/18	Safety	Flu/Cy followed by dose escalated CSR (PD-1/CD28) T cells from 5x10^4 - 1x10^7/Kg slit over 3 days	Recruiting
Uni. Penn	CART-EGFRvIII	Glioblastoma - EGFRvIII+ve	EGFRvIII	1	12	7/14	7/18	Safety	Residual or first recurrent disease	Active, not recruiting
Fuda Cancer Hospital, China	CAR-T	Glioblastoma - EphA2 +ve	Ephrin A2	1/2	60	9/15	9/16	Efficacy		Recruiting
Baylor CoM	HER-CAR CMV	Glioblastoma - HER2+ve	HER2	1	16	10/10	6/14	Safety	Patients receive 1 of 5 dose levels of HER2 CAR- CMV T cells: 1x10^6, 3x10^6, 1x10^7, 3x10^7 or 1x10^8/m^2	Active, not recruiting
PersonGen Bio, China	anti-MUC1 CART	Glioma, gastric or CRC - MUC1+ve	MUC1	1/2	20	10/15	10/17	Safety		Recruiting
PersonGen Bio, China	anti-MUC1 CART	HCC, NSCLC, pancreatic or TNBC - MUC1+ve	MUC1	1/2	20	10/15	10/17	Safety		Recruiting
King's College London	T4	Head & Neck (Squamous)	erbB	1	30	6/15	12/17	DLT	Intratumoral injection of T4 erbB targeted CAR-T	Recruiting
LBIO	LN-145	Head & Neck (Squamous)	NA	2	47	3/17	10/18	Safety	Lymphodepletion followed by TIL and IL-2	Recruiting
Fuda Cancer Hospital, China	CAR-T Mesothelin	Hepatocellular - GPC3+ve	Glypican 3	1/2	60	6/15	6/16	ORR	CAR-T Cell Immunotherapy for HCC Targeting GPC3	Recruiting
Sinobioway Cell Therapy, China	EPCAM-CART	Hepatocellular Carcinoma	EPCAM	1/2	25	11/15	11/19	DCR	CAR-T dosed d0, 1, 2, 28 & 29.	Recruiting
Baylor CoM	GLYCAR	Hepatocellular Carcinoma	Glypican 3	1	14	1/18	2/21	DLT	Flu/Cy followed by 1x10^7, 1x10^8 or 1x10^9 GLYCAR/m^2	Not yet recruiting
RenJi Hospital	GPC3 CAR T	Hepatocellular Carcinoma	Glypican 3	1	20	3/15	12/16	Safety	GPC3 CAR-T dose escalation	Recruiting
Shanghai GeneChem, China	TAI-GPC3-CART	Hepatocellular Carcinoma	Glypican 3	1/2	30	3/16	7/18	Safety	Cy followed by transarterial infusion of 1-10x10^6 GPC3-CART	Recruiting
2nd Military Medical University, China	PIK-PD-1	Hepatocellular Carcinoma	PD-1	1/2	40	9/15	3/17	OS	DC (1x10^7 on days 19/20, 40/41 & 61/62) + precision multiple antigen T cells (1-6x10^9 on days 21, 42 & 63) vs DC (1x10^7 on days 19/20, 40/41 & 61/62) + pluripotent killer T cells expression anti-PD-1 (1-6x10^9 on days 21, 42 & 63)	Recruiting
National cancer Institute	ACT	Liquid/solid tumor	PD-L1	1	40	2/17	11/20	Safety	ACT followed by TECENTRIQ Q3wk for 17 doses over 12 months starting within 6 months of ACT.	Recruiting
Roger Williams Medical Center	CEA CART	Liver Metastases	CEA	1	8	4/15	9/17	Safety	3 CAR-T hepatic artery infusions over 6 weeks with a single SIRT infusion and low dose IL-2	Active, not recruiting
Roger Williams Medical Center	CEA-CART	Liver Metastases	CEA	1	5	2/17	12/17	Safety	Hepatic arterial infusion weekly x3 followed by low dose IL-2	Recruiting
Xijing Hospital	MG7-CART	Liver Metastases	CEA	P1/2	20	6/16	5/17	Safety	Cy followed by intratumoral MG7-CART 1-6x10^8	Recruiting
Southwest Hospital, China	CEA CAR	Lung, colorectal, gastric, breast and pancreatic cancers	CEA	1	75	12/14	12/18	Safety	Dose ranging	Recruiting

Source: www.clinicaltrials.gov

Exhibit 53. Selected Solid Tumor ACT Trials continued

Sponsor/ Collaborators	Product	Indication	Target	Phase	N=	Start Date	End Date	Endpoint	Summary	Recruitment
Jonsson CC	MART1 TCR	Melanoma	MART-1 TCR	2	22	10/9	12/17	ORR	Flu/Cy followed by MART1-TCR, MART126.35 pulsed DC and IL-2	Recruiting
Netherlands Cancer Institute	MART1 TCR	Melanoma	MART-1 TCR	1/2	25	3/12	1/18	Safety	Flu/Cy followed by MART-1 TCR at: 5x1-^7, 1x10^8, 2.5x10^8 and 1x10^9.	Recruiting
Beijing Cancer Hospital, China	NKT	Melanoma	NA	1	20	10/15	10/16	Safety	3 cohorts: 1x10^9 d1, 2x10^9 d3, 4x10^9 d29 & 8x10^9 d31; 5x10^9 d1, 3, 29 & 31; 5x10^9 d1 & 3 every 28d x 8.	Recruiting
Fred Hutch CC	TIL	Melanoma	NA	2	20	7/26	10/17	Effiacy	Flu/Cy followed by TIL and IL-12 q8h 1-5 days	Recruiting
LBIO	LN-144	Melanoma	NA	2	40	9/15	1/18	Safety	Lymphodepletion followed by TIL and IL-2	Recruiting
MD Anderson CC	CD8+ T cell	Melanoma	NA	2	30	1/15	1/19	ORR	Cy followed by CD8+ve T cells (10x10^10/m^2) followed by IL-2 BID for 14 days with YERVOY doses between day 0 and 14 repeated every 3 weeks x 4 total	Recruiting
MD Anderson CC	CXCR2/NGFR TIL	Melanoma	NA	1/2	6	1/15	1/19	Safety	Flu/Cy followed by CXCR2/NGFR TIL and up to 15 doses of IL-2 over 5 days	Active, not recruiting
MD Anderson CC	dnTGFB/NGFR T TIL	Melanoma	NA	1	15	10/14	10/19	Feasibility	Flu/Cy followed by dnTGFBR TIL and equal dose of NGRF TIL up to 1.5x10^11 followed by inpatient high dose IL-2 on days 1-5 & 22-26.	Recruiting
Moffitt	TIL	Melanoma	NA	1	19	10/9	1/14	TIL growth	Flu/Cy & TIL + IL-2	Active, not recruiting
Netherlands Cancer Institute	TIL	Melanoma	NA	3	168	9/14	9/19	PFS	Flu/Cy followed by TIL and IL-2 vs YERVOY	Recruiting
Sheba Med Instit	TIL	Melanoma	NA	2	15	10/13	10/15	Effiacy	TIL plus YERVOY	Recruiting
Moffitt	TIL YERVOY	Melanoma	NA CTLA4	1/2	13	3/12	6/16	Safety	YERVOY followed by TIL acquisition and ~ 4 weeks after the 2nd dose of YERVOY, Flu/Cy followed by TIL and IL-2	Active, not recruiting
Loyola University	13831 TCR	Melanoma	Tyro-sinase	1	18	2/15	9/28	Safety	7.5x10^6, 2.5x10^7 or 7.5x10^7/Kg followed by low dose IL-2	Recruiting
Loyola University	TIL 1383I TCR	Melanoma	Tyro-sinase	1	15	7/12	9/13	Safety	2.5x10^6/Kg, 7.5x10^6/Kg or 2.5x10^7/Kg or 7.5x10^7/Kg followed by low dose IL-2	Recruiting
Moffitt	TIL	Melanoma	PD-1	1	12	3/16	8/19	Safety	Patients 1-6 receive TIL, patients 7-12 receive TIL on the background of OPDIVO; in all 12 patients TIL is manufactured in presence of 41BB activating antibody	Recruiting
Uni. Penn	RNA-CART-cMet	Melanoma or breast cancer	cMet	1	10	12/21	8/18	Safety	Patients receive up to 6 doses of 1x10^8 RNA-CART-cMet	Recruiting
ADAP	MAGE-A10 SPEAR	Melanoma, urothelial of H&N cancers MAGE A10 +ve	MAGE-A10	1	22	10/16	11/19	Safety	Flu/Cy followed by MAGE-A10 SPEAR - HLA-A*0201/06	Recruiting
NantKwest	NK-92 ALT-803	Merkel cell	NA IL-15	2	24	8/15	12/17	PFS	NK-92 dosed at 2x10^9/m^2 day 0 & 1 every 2 weeks with ALT-803 (IL-15) d0 of every cycle	Recruiting
Uni. Zurich	FAP-T cels	Mesothelioma	FAP	1	6	10/14	12/16	Safety	On d0 of 3rd cycle of palliative chemotherapy, 1x10^6 FAP CD8 T cells will be administered into the pleural effusion	Recruiting
Memorial Sloan KCC	iCasp9M28z	Mesothelioma	Meso-thelin	1	24	5/15	4/18	Safety	Dose escalation of iCasp9M28z up to a maximum of 3x10^6/Kg with or with out prior Cy	Recruiting
Fred Hutch CC	WT-1 TCR	Mesothelioma or NSCLC - WT1+ve	WT-1	1/2	20	5/15	6/17	Safety	HLA A*02 patients receive CD8 central memory/naïve WT-1 TCR+ve T cells days 0 & 14 , Cy days 11 &12 and IL-2 days 0-14 in stage 1; Cy followed by WT-1 TCR and IL-2 in stage	Recruiting
National cancer Institute	VEGFR2 CART	Metastatic Cancer	VEGFR	1/2	24	10/10	2/16	ORR	Flu/Cy followed by: 1x10^6, 3x10^6, 1x10^7, 3x10^7,1x10^8, 3x10^8 or 1x10^9 plus HD IL-2, OR 1x10^9, 3x10^9, 1x10^10 or3x10^10 + LD IL-2	Completed
Sichuan University	EPCAM-CART	Nasopharyngeal or breast cancer	EPCAM	1	30	7/16	7/17	Safety	Cy followed by dose escalated 3rd gen (CD28/41BB) EpCAM CAR-T	Recruiting
Seattle Children's	CD171 CART	Neuroblastoma	CD171	1	40	11/14	11/17	Safety	Flu/Cy followed by CD4:CD8 41bb or CD28/41BB dosed at 1x10^6, 5x10^6, 1x10^7, 5x10^7 or 1x10^8/Kg	Recruiting
Cancer Research UK	1RG-CART	Neuroblastoma	Disialogangli oside (GD2)	1	27	2/16	2/23	Feasibility	5 cohorts: 1x10^7/m^2 d0; Cy followed by 1x10^7/m^2, Flu/Cy followed by 1x10^7/m^2, Flu/Cy followed by 1x10^8/m^2, Flu/Cy followed by 5-10x10^8/m^2	Recruiting
Children's Mercy Hospital	GD2 CART	Neuroblastoma	Disialogangli oside (GD2)	1	5	10/11	1/15	Safety	Post allo-SCT multivirus GD2 CAR-T	Completed
Sinobioway Cell Therapy, China	GD2 CART	Neuroblastoma	Disialogangli oside (GD2)	1/2	22	9/16	9/20	Efficacy	Cytoreductive chemotherapy followed bu GD2-CAR-T days 0, 1, 2, 29 & 30.	Recruiting
Zhujiang Hospital	4SCAR-GD2	Neuroblastoma	Disialogangli oside (GD2)	2	30	5/16	5/17	Safety	Flu/Cy followed by 4SCAR-GD2 (CD28, 41BB & iCas9)	Recruiting
Baylor CoM	GD2 CART	Neuroblastoma	Disialogangli oside (GD2)	1	11	8/13	12/15	Safety	GD2 CAR-T (CD28/OX40 co-stim + iC9) fresh vs frozen CAR-T at 1x10^7, 1x10^8 or 2x10^8/M2 or Flu/Cy followed by fresh CAR-T 1.5x10^8 or 2x10^8 with KEYTRUDA dosed day -1 and d21	Active, not recruiting
Baylor CoM	GINAKIT	Neuroblastoma - pediatric	Disialogangli oside (GD2)	1	18	8/16	9/19	DLT	Flu/Cy followed by NKT iCas9 CD28/OX40 GD2 CAR	Not yet recruiting

Source: www.clinicaltrials.gov

Exhibit 53. Selected Solid Tumor ACT Trials continued

Sponsor/ Collaborators	Product	Indication	Target	Phase	N=	Start Date	End Date	Endpoint	Summary	Recruitment
ADAP	MAGE A10 SPEAR	NSCLC - MAGE A10 +ve	MAGE-A10	1/2	32	12/15	11/17	DLT	Cy followed by 3 dose cohorts: 0.1x10^9, 1x10^9 and 5x10^9 - HLA-A*0201/06	Recruiting
ADAP	NY-ESO-SPEAR	NSCLC - NY-ESO1 +ve	NY-ESO1	1/2	10	10/15	11/17	Safety	Cy followed by 5x10^9 NY-ESO SPEAR - HLA A*02	Recruiting
Guangzhou Institute , China	TAEST16001	NSCLC - NY-ESO1+ve	NY-ESO1	1	20	3/17	12/17	Safety	Cy followed by 5x10^9 anti-NY-ESO1 TCR T cells and IL-2 for 14 days - HLA A*0201	Recruiting
Christie NHS Foundation Trust	NY-ESO SPEAR	Oesophageal Cancer	NY-ESO1	2	28	10/14	12/18	ORR	Flu/Cy followed by NY-ESO SPEAR (HLA A*02) and IL-2	Active, not recruiting
Baylor CoM	iC9-GD2-CAR-VZV	Osteosarcoma	Disialogangli oside (GD2)	1	26	4/14	4/18	Safety	3rd generation GD-2 CAR-T with CD28/OX40 costim and iC9 suicide switch dosed a 1x10^ or 1x10^7/m^2 followed by VZV vaccine d42, and VZV vaccine followed within 48h by 1x10^7 or 1x10^8/m^2.	Recruiting
Herlev Hospital, DK	TIL	Ovarian Cancer	NA	1	6	7/15	12/16	Safety	Cy/Flu followed by TIL and IL-2 (decrescendo)	Active, not recruiting
Roswell Park	NY-ESO1 TCR	Ovarian Cancer	NY-ESO1	1	12	2/17	10/19	Safety	Decitabine (increase NY-ESO1 expression) d1-3 followed by Cy d5-6 and NY-ESO1 TCR IV & IP d9 with IL-2 d10-23 for course 1 with decitabine d31-33 and NY-ESO1 IV/IP d37 and IL-2 d38-51.	Not yet recruiting
Shanghai GeneChem, China	meso-CART	Pancreatic Cancer	Meso-thelin	1	30	3/16	2/18	Safety	Cy followed by transarterial infusion of 1-10x10^6 GPC3-CART	Recruiting
Uni. Penn	Meso RNA CAR	Pancreatic Cancer	Meso-thelin	1	16	7/13	12/15	Safety	1x3^8/m^2 3 times weekly for 3 weeks.	Active, not recruiting
Uni. Penn	meso-CART CD19 CART	Pancreatic cancer	Meso-thelin	1	12	5/15	9/17	Safety	Cy followed by a mixed dose of meso & CD19 CART (1-3x10^7 or 1-3 x 10^8/m^2)	Not recruiting
BLCM	BPX-601	Pancreatic cancer	PSCA	1	30	11/16	12/19	MTD	BPX-501 dosing will be followed by rimiducid to induce CAR-T activation	Recruiting
Uni. Penn	CART-meso	Pancreatic, ovarian or mesothelioma	Meso-thelin	1	19	6/14	11/15	Safety	CART-meso +/- Cy at 1-3x10^7 or 1-3 x10^8/m^2	Active, not recruiting
Uni. Penn	CART-PSMA- TGFβRDN	Prostate cancer	PSMA	1	18	3/17	9/20	Safety	Three cohorts of patients will receive a PSMA CAR in a TGFbetaRdn T cell: 1-3x10^7, 1-3x10^8 or Cy followed by MTD of the CART	Recruiting
Memorial Sloan KCC	PSMA CART	Prostate Cancer	PSMA	1	18	6/10	6/18	Safety	Cy followed by PSMA CART 1x10^7/Kg, 3x10^7/Kg or 1x10^8/Kg	Recruiting
Fred Hutch CC	NY-ESO1 CD8+ve	Sarcoma	NY-ESO1	1	6	1/12	9/13	Safety	Cyclophosphamide followed by NY-ESO1+ve CD8 T cells	Safety
ADAP	NY-ESO-SPEAR	Sarcoma - myxoid/ round cell	NY-ESO1	1/2	15	11/16	5/18	CR rate	Flu/Cy followed by NY-ESO SPEAR 1-8x10^9 - HLA A*02	Recruiting
ADAP	NY-ESO SPEAR	Sarcoma - synovial	NY-ESO1	1/2	65	3/11	10/17	CR/PR	Flu/Cy high NY-ESO1, Flu/Cy low NY-ESO1, Cy high NY-ESO1, RD Flu/Cy high NY-ESO1	Recruiting
Immatics	IMA101	Solid tumors	ACTolog Ag	1/2	31	10/16	9/18	Safety	HLA A*02 patients receive CD8 IMA101 product following Flu/Cy if their tumor contains a relevant antigen	Not yet recruiting
MD Anderson CC	TIL	solid tumors	NA	1	40	2/17	2/21	Safety	Cy followed by TIL (20x10^9/m2) and IL-2 d0-14 and KEYTRUDA day 1, weeks 3, 6, 9, 12 & 15.	Not yet recruiting
NantKwest	haNK	Solid tumors	NA	1	16	4/17	12/19	MTD	aNK (NK-92 with IL-2 and CD16) dosed at 2x10^9 or 4x10^9	Not yet recruiting
Chinese PLA General Hospital, China	CART-133	Solid tumors - CD33+ve	CD33	1	20	9/15	10/17	Safety	CART-133 dose escalation - de-escalation	Recruiting
Chinese PLA General Hospital	EGFR CART	Solid tumors - EGFR+ve	EGFR	1/2	60	5/13	12/16	Safety		Recruiting
Ningbo Cancer Hospital, China	HerinCAR-PD1	Solid tumors - EGFR+ve	EGFR PD-L1	1/2	20	8/16	3/18	ORR	Patients receive 3 cycles of HerinCAR-PD1 cells dosed at 1-5x10^7/Kg	Recruiting
First Affiliated Hospital of Chengdu Medical College	EpCAM CART	Solid tumors - EpCAM+ve	EpCAM	1/2	60	1/17	12/18	Safety	Lymphodepletion followed by CAR-T (1-10x10^6/Kg)	Recruiting
Southwest Hospital, China	HER2 CAR_T	Solid tumors - HER2 +ve	HER2	1/2	60	3/16	9/18	Safety		Recruiting
National cancer Institute	MAGE-A3 TCR	Solid tumors - MAGE-A3 +ve	MAGE-A3-DP4	1/2	102	5/14	6/17	Safety	MAGE-A3, HLA A*01 - Flu/Cy followed by TCR and IL-2 for up to 5 days	Recruiting
National cancer Institute	MAGE A3-DP4	Solid tumors - MAGEA3=ve	MAGE-A3-DP4	1/2	107	1/14	12/17	Safety	Flu/Cy followed by MAGE-A3-DP4 TCR for melanoma or a 2nd cohort of renal and other metastatic cancer followed by IL-2	Recruiting
China Meitan General Hospital	meso-CART	Solid tumors - meso+ve	Meso-thelin	1	20	8/16	8/18	Safety	Cy followed by anti-mesothelin CAR-T dose ranged from 5x10^4 - 1x10^7/Kg split over 3 days	Recruiting
Chinese PLA General Hospital	CART-meso	Solid tumors - meso+ve	Meso-thelin	1	20	10/15	11/17	Safety	CART-meso dose escalation - de-escalation	Recruiting
Uni. Penn	huCART-meso	Solid tumors - meso+ve	Meso-thelin	1	30	3/17	3/21	Safety	huCART-meso dose escalated from 1-3x10^7/m^2 with or without prior Cy lymphodepletion	Not yet recruiting
Ningbo Cancer Hospital, China	meso-CART	Solid tumors - meso+ve	Meso-thelin	1/2	40	2/17	12/18	Safety	Patients receive PD-1 antibody expressing meso-CART	Recruiting
National cancer Institute	CAR-T Mesothelin	Solid tumors - meso+ve	Meso-thelin	1/2	136	3/12	12/18	Safety	Flu/Cy followed by mesothelin CAR-T and IL-2	Recruiting
PersonGen Bio, China	CAR-pNK	Solid tumors - MUC1+ve	MUC1	1	10	7/16	7/17	Safety	Patients receive NK-92 modifed with a MUC1 CAR	Recruiting
Memorial Sloan KCC	MUC16ecto	Solid tumors - MUC16ecto+ve	MUC16	1	30	8/15	8/18	MTD	Cy followed by 2 infusions - IV and then IP 1-3 days later - 4 dose levels: 3x10^5, 1x10^6, 3x10^6 & 1x10^7 - T cells also engineered for IL-12	Recruiting

Source: www.clinicaltrials.gov

Exhibit 53. Selected Solid Tumor ACT Trials continued

Sponsor/ Collaborators	Product	Indication	Target	Phase	N=	Start Date	End Date	Endpoint	Summary	Recruitment
Jonsson CC	NY-ESO1 TCR	Solid tumors - NY-ESO1 +ve	NY-ESO1	1	12	7/14	2/19	Safety	Flu/Cy followed by NY-ESO-1 TCR (HLA A*0201) followed d0/1 by YERVOY (dosed q3wk x 4) and NY-ESO1 peptide (157-165) pulised DC day 1, 14 and 30 and low dose IL-2 BID days 1-14.	Recruiting
Jonsson CC	TCR- NY-ESO1	Solid tumors - NY-ESO1 +ve	NY-ESO1	1	12	6/17	6/18	Safety	Flu/Cy followed by NY-ESO1 TCR d 1, 14 & 28 with IL-2 d1-7 and OPDIVO starting d0 or 1 for 2 years	Not yet recruiting
Takara	TBI-1301	Solid tumors - NY-ESO1 +ve	NY-ESO1	1	12	3/15	3/17	Safety	Cy or Flu/Cy followed by 5x10^8 or 5x10^9 TBI-1301	Recruiting
Takara	TBI-1301	Solid tumors - NY-ESO1 +ve	NY-ESO1	1	15	9/16	3/18	Safety	Cy followed by TBI-1301	Recruiting
Albert Einstein CoM	mTCR	Solid tumors - NY-ESO1 +ve	NY-ESO-1		10	8/16	1/18	Safety	Flu/Cy followed by murine-ESO TCR and IL-2 (days0-4) and filgrastim (days 1-4) in HLA A*02	Not yet recruiting
Baylor CoM	GLYCAR	Solid tumors - ped GPC3 +ve	Glypican 3	1	14	3/18	4/21	DLT	Flu/Cy followed by 1x10^7, 1x10^8 or 1x10^9 GLYCAR/m^2	Not yet recruiting
Celyad	NKR-2 CART	Solid tumors (CRC, ovarian, bladder, TNBC and pancreatic), AML or MM	NKG2D	1	24	12/16	5/20	Safety	NKR-2 CAR will be dose escalated with each dose given every 2 weeks for a total of 3 infusions	Recruiting
Shenzhen Geno-Immune, China	4SCAR-GD2	Solid tumors GD2+ve	Disialoganglioside (GD2)	1/2	100	5/16	6/18	Safety	Flu/Cy followed by 4SCAR-GD2 (CD28, 41BB & iCas9)	Recruiting
National cancer Institute	GD2 CART	Solid tumors GD2+ve	Disialoganglioside (GD2)	1	15	2/14	1/17	Safety	Cy followed by 1x10^5, 1x10^6, 3x10^6 or 1x10^7/Kg of CD28/OX40 iC9 CAR-T	Completed
Roswell Park	NY-ESO-1 TCR dnTGFbRII TIL	Solid tumors NY-ES01+ve	NY-ESO1	1/2	24	5/17	10/18	DLT	Cy followed by NY-ESO1 TCR and dnTGFRbRII TIL	Not yet recruiting
Fred Hutch CC	ROR1 CART	Solid/liquid tumors - ROR1+ve	ROR1	1	60	3/16	12/21	MTD	Flu/Cy followed by anti-Receptor tyrosine kinase-like orphan receptor 1 (ROR1) CAR-T with an option fo a 2nd dose with or without additional cytoreductive chemotherapy at a higher dose or at the MTD	Enrolling by invitation
Carsgen Therapeutics, China	CAR-GPC3	Squamous Cell Lung	Glypican 3	1	20	3/16	10/17	Safety	Flu/Cy followed by 1x10^5 - 2x10^9 CAR-GPC3 T cells	Recruiting
Sinobioway Cell Therapy, China	EPCAM-CART	Stomach	EPCAM	1/2	19	11/15	11/19	DCR	CAR-T dosed d0, 1, 2, 28 & 29.	Recruiting
MD Anderson CC	TIL	Uveal melanoma	NA	1	30	4/17	4/21	MTD	Following an initial cohort of CD8+ve TIL (starting at 3.3x10^9 and escalated to MTD), patients will receive Cy followed by CD8+ve TIL at MTD and IL-2 for 14 days and YERVOY starting 1 day after TIL and dosed every 3 weeks for 4 doses.	Not yet recruiting

Source: www.clinicaltrials.gov

Neoantigens offer a potential to both solve the issue of antigen expression by the tumor and the issue of on-target off-tumor toxicity as *apriori,* a neoantigen is unique to the tumor, and selection of a neoantigen for targeting would depend on its actual, or in the case of a multiple neoantigen approach, predicted expression by the patient's tumor. The challenge, however, has been the ability to develop a product feasibly and in a relevant time frame. The latter obviates the use of viruses to create a neoantigen ACT product and clearly was a driver in NCI collaborating with ZIOP to use Sleeping Beauty as the means to rapidly create a neoantigen approach. ZIOP intends to be ready by YE17 to move a neoantigen approach into the clinic.

Paying for Adoptive Cellular Therapies (ACT)

With no ACT product yet approved, the logistics of insurance coverage remains to be elucidated. Arguably, stem cell transplant (SCT) provides a model for reimbursement, although we note that stem cell transplant is not an FDA-approved procedure. For certain indications, CMS covers allogeneic and autologous transplantation, including stem cell mobilization, stem cell harvesting, high-dose chemotherapy/radiotherapy, and transplant of stem cells. Private payers also cover the costs associated with stem cell transplant and each insurer has contracts with top-tier transplant centers to provide transplant services. Payments for transplant use both global case rates and Medicare diagnostic-related group (DRG) codes.

In a presentation from the National Marrow Donor Program (NMDP), the Director of Payer Policy noted that stem cell transplant has limited coverage and reimbursement by government payers, that the commercial market is becoming more splintered and is decreasing with pressure to decrease cost, and there is an increase in measurement of value, with a focus on quality of life and similar patient-centered outcomes. Health exchange networks (HIX) further reduce access to transplant, with HIX plans estimated to have 42% fewer oncologists than commercial plans, and no requirement to provide access to transplant centers. Limited access to transplant centers also reflects some transplant centers choosing not to contract with HIX plans due to low reimbursement, and some payers seeking single case agreements, rather than contracting.

Majhail and colleagues at the NDMP have published costs over the first 100 days following an autologous or an allogeneic transplant using claims from the Thomson Reuters MarketScan database, Bone Marrow Transplant. 2013 Feb;48(2):294-300. Majhail and colleagues analyzed data on 3365 patients, the vast majority of whom had a diagnosis of cancer, and approximately 60% of subjects received an autologous transplant and 40%, an allogeneic transplant. Following transplant, patients receiving an autologous transplant spent 19 days in hospital, compared to those receiving an allogeneic transplant that spent 31 days in hospital. Over the first 100 days following transplant, the costs associated with an allogeneic transplant were approximately 2-fold higher than those for an autologous transplant, and this difference was exacerbated by the 50% premium in costs for pediatric/adolescent versus adult recipients of an allogeneic transplant. As noted in the bottom of Exhibit 54, cost of the first 100 days of care for 20% of pediatric/adolescent allogeneic recipients exceeded $500,000.

Exhibit 54. Costs of Autologous and Allogeneic Transplant over 100 Days Post-Transplant

	Autologous	Allogeneic
Median hospitalization		
Number	1	1
Days	19	31
Costs	$ 99,899	$ 203,026
>20yr	$ 102,458	$ 191,142
<20yr	$ 117,674	$ 302,822

Cost $	Autologous <20yr	Autologous >20yr	Allogeneic <20yr	Allogeneic >20yr
<100K	13%	53%	3%	13%
100-200K	52%	39%	22%	41%
200-300	17%	5%	24%	23%
300-400	10%	1%	20%	12%
400-500	3%	1%	12%	5%
>500	6%	1%	20%	6%

Source for both tables: Mijhail and Wells Fargo Securities, LLC

In this experience, less than 1% of patients required hospitalization for longer than 100 days (n=27) where a median duration for hospitalization was reported as 112 days. The actuarial firm Milliman has also published transplant-related cost estimates reporting charges 30 days prior to and 180 days following a transplant for the year 2014. Milliman estimates that in 2014, 8,709 allogeneic and 12,460 autologous transplants were performed in the United States for estimated costs of $930.600 and $378,000, respectively, including duration of hospitalization ranging from 20 days for autologous transplant to 33 for an allogenic transplant. In Exhibit 55, Milliman used the Center for International Blood and Marrow Transplant registry for procurement costs, with other costs estimated from proprietary billing data. Milliman also presents the cost of transplant as a cost per member, per month of $2.22 and $1.11 for the younger than 65 population for an allogeneic and an autologous transplant, respectively, based on 1 million people. For context, the PMPM costs for a liver or kidney transplant is $1.11 and $1.29, respectively.

Exhibit 55. Milliman Analysis for Costs of Autologous and Allogeneic Transplant

	Allogeneic	Autolgous
Total	$ 930,600	$ 378,000
30-days prior to	$ 57,600	$ 56,300
Procurement	$ 55,700	$ 10,700
Transplant - hospital admission	$ 479,600	$ 212,300
Physician costs	$ 23,400	$ 10,800
180-day post	$ 290,300	$ 81,800
Drug costs	$ 24,000	$ 6,100

Source: Milliman and Wells Fargo Securities, LLC

The transplant-related costs reflect the use of myeloablative conditioning and associated in-patient costs. In 2014, researchers from the Fred Hutchinson Cancer Research Center (FHCRC) and Dana Faber/Brigham and Women's (DFBW) published a cost analysis for the use of reduced intensity conditioning prior to an allogeneic transplant performed between 2008 and 2010, The Oncologist 2014;19:639–644 . For the first 100 days post-transplant estimated costs in 2010 ranged from $96,000 at DFBW to $129,000 at FHCRC. The median number of in-patient days was 9-11, although in-patient costs were higher at DFBW, due to the requirement for in-patient conditioning vs. outpatient at FHCRC. DFBW also reported a median $39,000 cost between day 100 and 2 years, reflecting costs for chronic GvHD and death. Note that as data from additional patients enrolled into KITE's AxiCel ZUMA-1 trial have been reported, serious CRS and NT rates have fallen, with more aggressive use of Actemra and other strategies to blunt the development of these AEs to a level requiring admission to the ICU, which would incur additional costs.

Medicare reimbursement is inadequate for transplant

The Medicare in-patient payment system for 2016 related to allogeneic or autologous transplants lists:

- MS-DRG 014: allogeneic - $62,245 inclusive of donor search and acquisition cost
- MS-DRG 016: autologous with major complications or comorbidities - $33,153
- MS-DRG 017: autologous without major complications or comorbidities - $23,475

And for outpatient services, Medicare provides $3045.31 for either auto or allo transplant.

In a NMBP survey of 61 transplant centers, more than 30% of respondents stated that steerage of patients through narrow networks of benefit tiers was their most concerning financial issue for the transplant program. Additional top financial concerns were administrative issues with payer, pre-authorization, and contracting issues, cited by 28%, growth in patients with government payers, cited by 23%, and increasing pressure to reduce cost, cited by 13%. The greatest barriers to HCT included high deductible and or co-pay/co-insurance cited by 26%, inadequate donor search/ cell acquisition benefit/coverage, cited by 19%, and inadequate/no coverage for certain indications, cited by 14%.

ACT- Approval is the Gateway to Reimbursement

With formal FDA approval, ACT-based therapies can seek Current Procedural Terminology (CPT) codes through the American Medical Association. A new code takes approximately 2 years to achieve, after which transplant centers and payers can contract easily and correctly. The category III CPT code is specific for emerging technology.

Following FDA approval, commercial insurance companies determine coverage. This is a 2-step process that involves a medical review, followed by the actuary review to establish the per-patient per-month cost to the company. In working with experts on reimbursement, we understand that insurance companies pay particular attention to costs in excess of $0.1 PMPM. Even for an approved product, as recently evidenced for Sarepta's (SRPT) EXONDYS 51, the insurer Anthem determined the product to be experimental and refused coverage.

Commercial insurers can decide if ACT is covered as a drug or is part of a capitated payment covering a treatment episode. As a drug, the reimbursement for the therapy is simplified as hospitals can be reimbursed for the drug; but in this case, they also need to seek reimbursement for the treatment costs before and after the ACT product. For usual care such as treating infections, toxicity associated with treatment, and disease relapse, we assume this would be straight forward, but there are several aspects of ACT that may not be as easily considered standard of care:

- Apheresis is standard of care for a stem cell transplant, but not outside of SCT.
- Lymphodepleting chemotherapy is not standard of care since it is not designed to target the tumor.
- ACTEMRA is approved as a drug for rheumatoid arthritis –writing a script to cover use in treating CRS would not be standard care.

The alternative would be for insurers to capitate payment for a treatment episode covering a certain period of time before to a certain period of time following ACT. This payment would cover the time line between preparation for apheresis to say, 30 days after ACT. In SCT transplant the post-transplant period of coverage is usually 30 or 100 days.

Medicare reimbursement presents another challenge as Medicare reimbursement for SCT loses the hospital money. Hospitals put up with this on the assumption that losses are more than made up for by margin from commercial payers for SCT.

Disclaimer: The external website links included in this publication are not maintained, controlled, or operated by Wells Fargo Securities. Wells Fargo Securities does not provide the products and services on these websites and the views expressed on these websites do not necessarily represent those of Wells Fargo Securities. Please review the applicable privacy and security policies and terms and conditions for the website you are visiting.

Required Disclosures

This is a compendium report, to view current important disclosures and other certain content related to the securities recommended in this publication, please go to https://www.wellsfargoresearch.com/Disclosures or send an email to: equityresearch1@wellsfargo.com or a written request to Wells Fargo Securities Research Publications, 7 St. Paul Street, Baltimore, MD 21202.

Additional Information Available Upon Request

I certify that:
1) All views expressed in this research report accurately reflect my personal views about any and all of the subject securities or issuers discussed; and
2) No part of my compensation was, is, or will be, directly or indirectly, related to the specific recommendations or views expressed by me in this research report.

Wells Fargo Securities, LLC does not compensate its research analysts based on specific investment banking transactions. Wells Fargo Securities, LLC's research analysts receive compensation that is based upon and impacted by the overall profitability and revenue of the firm, which includes, but is not limited to investment banking revenue.

STOCK RATING
1=Outperform: The stock appears attractively valued, and we believe the stock's total return will exceed that of the market over the next 12 months. BUY
2=Market Perform: The stock appears appropriately valued, and we believe the stock's total return will be in line with the market over the next 12 months. HOLD
3=Underperform: The stock appears overvalued, and we believe the stock's total return will be below the market over the next 12 months. SELL

SECTOR RATING
O=Overweight: Industry expected to outperform the relevant broad market benchmark over the next 12 months.
M=Market Weight: Industry expected to perform in-line with the relevant broad market benchmark over the next 12 months.
U=Underweight: Industry expected to underperform the relevant broad market benchmark over the next 12 months.

VOLATILITY RATING
V = A stock is defined as volatile if the stock price has fluctuated by +/-20% or greater in at least 8 of the past 24 months or if the analyst expects significant volatility. All IPO stocks are automatically rated volatile within the first 24 months of trading.

As of: April 19, 2017

42% of companies covered by Wells Fargo Securities, LLC Equity Research are rated Outperform.	Wells Fargo Securities, LLC has provided investment banking services for 44% of its Equity Research Outperform-rated companies.
56% of companies covered by Wells Fargo Securities, LLC Equity Research are rated Market Perform.	Wells Fargo Securities, LLC has provided investment banking services for 31% of its Equity Research Market Perform-rated companies.
2% of companies covered by Wells Fargo Securities, LLC Equity Research are rated Underperform.	Wells Fargo Securities, LLC has provided investment banking services for 26% of its Equity Research Underperform-rated companies.

Important Disclosure for International Clients

EEA – The securities and related financial instruments described herein may not be eligible for sale in all jurisdictions or to certain categories of investors. For recipients in the EEA, this report is distributed by Wells Fargo Securities International Limited ("WFSIL"). WFSIL is a U.K. incorporated investment firm authorized and regulated by the Financial Conduct Authority. For the purposes of Section 21 of the UK Financial Services and Markets Act 2000 ("the Act"), the content of this report has been approved by WFSIL a regulated person under the Act. WFSIL does not deal with retail clients as defined in the Markets in Financial Instruments Directive 2007. The FCA rules made under the Financial Services and Markets Act 2000 for the protection of retail clients will therefore not apply, nor will the Financial Services Compensation Scheme be available. This report is not intended for, and should not be relied upon by, retail clients.

Australia – Wells Fargo Securities, LLC is exempt from the requirements to hold an Australian financial services license in respect of the financial services it provides to wholesale clients in Australia. Wells Fargo Securities, LLC is regulated under U.S. laws which differ from Australian laws. Any offer or documentation provided to Australian recipients by Wells Fargo Securities, LLC in the course of providing the financial services will be prepared in accordance with the laws of the United States and not Australian laws.

Canada – This report is distributed in Canada by Wells Fargo Securities Canada, Ltd., a registered investment dealer in Canada and member of the Investment Industry Regulatory Organization of Canada (IIROC) and Canadian Investor Protection Fund (CIPF). Wells Fargo Securities, LLC's research analysts may participate in company events such as site visits but are generally prohibited from accepting payment or reimbursement by the subject companies for associated expenses unless pre-authorized by members of Research Management.

Hong Kong – This report is issued and distributed in Hong Kong by Wells Fargo Securities Asia Limited ("WFSAL"), a Hong Kong incorporated investment firm licensed and regulated by the Securities and Futures Commission of Hong Kong ("the SFC") to carry on types 1, 4, 6 and 9 regulated activities (as defined in the Securities and Futures Ordinance (Cap. 571 of The Laws of Hong Kong), "the SFO"). This report is not intended for, and should not be relied on by, any person other than professional investors (as defined in the SFO). Any securities and related financial instruments described herein are not intended for sale, nor will be sold, to any person other than professional investors (as defined in the SFO). The author or authors of this report is or are not licensed by the SFC. Professional investors who receive this report should direct any queries regarding its contents to Mark Jones at WFSAL (email: wfsalresearch@wellsfargo.com).

Japan – This report is distributed in Japan by Wells Fargo Securities (Japan) Co., Ltd, registered with the Kanto Local Finance Bureau to conduct broking and dealing of type 1 and type 2 financial instruments and agency or intermediary service for entry into investment advisory or discretionary investment contracts. This report is intended for distribution only to professional investors (Tokutei Toushika) and is not intended for, and should not be relied upon by, ordinary customers (Ippan Toushika).

The ratings stated on the document are not provided by rating agencies registered with the Financial Services Agency of Japan (JFSA) but by group companies of JFSA-registered rating agencies. These group companies may include Moody's Investors Services Inc., Standard & Poor's Rating Services and/or Fitch Ratings. Any decisions to invest in securities or transactions should be made after reviewing policies and methodologies used for assigning credit ratings and assumptions, significance and limitations of the credit ratings stated on the respective rating agencies' websites.

About Wells Fargo Securities

Wells Fargo Securities is the trade name for the capital markets and investment banking services of Wells Fargo & Company and its subsidiaries, including but not limited to Wells Fargo Securities, LLC, a U.S. broker-dealer registered with the U.S. Securities and Exchange Commission and a member of NYSE, FINRA, NFA and SIPC, Wells Fargo Prime Services, LLC, a member of FINRA, NFA and SIPC, Wells Fargo Securities Canada, Ltd., a member of IIROC and CIPF, Wells Fargo Bank, N.A. and Wells Fargo Securities International Limited, authorized and regulated by the Financial Conduct Authority.

This report is for your information only and is not an offer to sell, or a solicitation of an offer to buy, the securities or instruments named or described in this report. Interested parties are advised to contact the entity with which they deal, or the entity that provided this report to them, if they desire further information. The information in this report has been obtained or derived from sources believed by Wells Fargo Securities, LLC, to be reliable, but Wells Fargo Securities, LLC does not represent that this information is accurate or complete. Any opinions or estimates contained in this report represent the judgment of Wells Fargo Securities, LLC, at this time, and are subject to change without notice. All Wells Fargo Securities research reports published by its Global Research Department ("WFS Research") are disseminated and available to all clients simultaneously through electronic publication to our internal client websites. Additional distribution may be done by sales personnel via email, fax or regular mail. Clients may also receive our research via third party vendors. Not all research content is redistributed to our clients or available to third-party aggregators, nor is WFS Research responsible for the redistribution of our research by third party aggregators. For research or other data available on a particular security, please contact your sales representative or go to http://www.wellsfargoresearch.com. For the purposes of the U.K. Financial Conduct Authority's rules, this report constitutes impartial investment research. Each of Wells Fargo Securities, LLC and Wells Fargo Securities International Limited is a separate legal entity and distinct from affiliated banks. Copyright © 2017 Wells Fargo Securities, LLC

SECURITIES: NOT FDIC-INSURED/NOT BANK-GUARANTEED/MAY LOSE VALUE