

Chimeric antigen receptor-modified T-cells in acute lymphoblastic leukemia

See Commentary on page 254

Recent studies have documented the ability of chimeric antigen receptor-modified autologous T cells targeting the CD19 antigen on B cells to induce rapid and deep remissions in adult and pediatric patients with relapsed or refractory acute lymphoblastic leukemia (ALL).^{1,2}

Study in 5 adults with relapsed B-cell ALL

Brentjens and colleagues found that molecular remission was rapidly induced in patients with relapsed B-cell ALL using autologous T cells modified to express a CD19-specific CD28/CD3- ζ dual-signaling chimeric antigen receptor (CAR; 19-18z CAR-modified T cells).¹ Five adult patients (age range, 23-66 years) who had not previously received allogeneic hematopoietic stem cell transplantation (HSCT) received the adoptive T-cell therapy after conditioning therapy with cyclophosphamide. Treatment consisted of an infusion of $1.5\text{-}3.0 \times 10^6$ autologous 19-18z CAR-modified T cells/kg. Eligible patients subsequently underwent allogeneic HSCT.

Of the 5 patients, 2 had persistent chemotherapy-refractory disease after salvage therapy (63% and 70% blasts in bone marrow). Two others had achieved morphologic complete remission (CR) during salvage therapy with evidence of minimal residual disease (MRD) on deep sequencing polymerase chain reaction (PCR) and fluorescence-activated cell sorting (FACS), and 1 patient was MRD negative after salvage therapy.

All of the patients were MRD negative on PCR after adoptive T-cell therapy. Of the 2 patients with persistent refractory disease after salvage therapy, 1 achieved morphologic CR by day 11 after T-cell infusion and MRD-negative status by day 59, and the other achieved both morphologic CR and MRD-negative status by day 8. Of the 2 other MRD-positive patients, 1 was MRD negative by day 28 and the other was MRD negative at day 30 and remained MRD negative up to the time of allogeneic HSCT at 122 days. Four patients underwent allogeneic HSCT at 1 to 4 months after T-cell therapy. One patient, who was ineligible for allogeneic HSCT (due to multiple pre-existing comorbidities) and additional T-cell therapy, relapsed at

What's new, what's important

Clinical development of chimeric antigen receptors is a fascinating novel approach in cancer treatment. One of the major barriers of T-cell therapy is tolerance to target tumor-associated antigens. Scientists have been able to overcome this tolerance by using the chimeric antigen receptor (CAR) to redirect T-cell specificity to tumor-associated antigens expressed on the cell surface. CARs are made by collecting T cells from the patient and genetically modifying them to recognize the antigen-binding site on the cancer cells. Then the cells are re-infused into the patient. Once the CAR cells have been infused back into the patient, they can potentially expand in number and last for a long time. These cells are capable of killing any cells that have the target antigen.

As per investigators, the full therapeutic potential of CAR depends on 3 factors:

- Persistence - the cells need to survive in the host environment and expand. This could be achieved by preparing the host with chemotherapy or radiation, though radiation has its own drawbacks.
- Homing - the infused CARs need to track the tumor cells and grow in the sites of malignancy, but the capability of migration could be compromised during genetic modification.
- Overcoming resistance - CARs' ability to achieve full tumor kill depend on their ability to overcome "adverse regulatory effects within the tumor microenvironment" (*Blood*. 2010;116:1035-1044).

First generation of CARs have shown moderate clinical activity, but the clinical efficacy of second- and third-generation with up to 3 chimeric moieties are much more robust. Initial results from small clinical trials in hematological malignancy are very promising. Continued evolution of this technology and better understanding of the host and tumor immune mechanism will make this approach another exciting targeted treatment for patients with refractory cancer.

— Jame Abraham, MD

How I treat ALL

The treatment of acute lymphoblastic leukemia (ALL) in adults remains challenging. It is often intense and prolonged. Initial chemotherapy is focused on obtaining a remission as well as identifying patients who need to consider allogeneic stem cell transplantation early (in first remission). Consolidation of remission occurs afterward. Patients treated with chemotherapy also receive maintenance therapy. Given the ability of ALL to enter central nervous system, prophylaxis and treatment into the CNS is necessary to prevent relapse.

For adults younger than 30 years, there is some evidence that treatment using pediatric protocols improved patient outcome. For other adults, I often use an induction and consolidation strategy of hyperCVAD (cyclophosphamide, Adriamycin, vincristine and dexamethasone) alternating with high-dose methotrexate and cytarabine. This therapy incorporates CNS treatment. Rituximab is added to the treatment for those who express CD20.

The most common cytogenetic abnormality in patients with ALL is the presence of t(9;22), the Phila-

delphia chromosome. Those patients also benefit from the addition of a tyrosine kinase inhibitor to the induction and consolidation therapy. It is important to identify patients who are unlikely to be cured with chemotherapy early in their course, so they may be referred promptly for consideration of allogeneic stem cell transplant. This would include patients with high-risk cytogenetic such as the t(9;22) or chromosome 11q23 abnormality, or those with persistent evidence of disease (minimal residual disease) positivity. Intensive supportive care with antimicrobials and transfusion support is necessary for patients with ALL. For those who relapse, re-induction therapy to enter second remission is a bridge to allogeneic transplantation if a donor is available. For patients with multiply relapsed or refractory ALL, a new agent – liposomal vincristine – was approved by the Food and Drug Administration earlier this year. However, it is not yet widely available.

— Michael Craig, MD

90 days after T-cell therapy after receiving high-dose steroid therapy for cytokine-induced toxicities.

Treatment with CD19-targeted CAR-modified T cells has been associated with high fever, hypotension, and elevated pro-inflammatory cytokine levels in patients with chronic lymphocytic leukemia and B-cell non-Hodgkin lymphoma. Monitoring of serum cytokine levels in the current study showed that the 2 patients with the highest tumor burden at the start of T-cell therapy exhibited the greatest cytokine elevations, which began at 5 days after T-cell infusion. These patients also had the highest numbers of detectable CAR-modified T cells after treatment. Cytokine elevations were much smaller or undetectable in the other patients, suggesting that cytokine elevation was associated with bulk of residual disease at the time of T-cell therapy. Three patients had transient fevers after T-cell infusion, with the 2 with the greatest disease burden at the time of treatment exhibiting increased fever severity and persistence, hypotension, and mental status changes. Both were treated with high-dose lymphotoxic steroid therapy starting on day 6 and tapered slowly thereafter, with constitutional symptoms rapidly improving and cytokine levels becoming normalized.

The ability to measure persistence of the CAR-modified T cells was limited by the performance of allogeneic HSCT relatively soon after treatment in 4 patients and the need for high-dose steroid therapy in 2. The T cells were detectable by PCR or FACS in the blood and

bone marrow 3 to 8 weeks after infusion. Expansion of the T cells was correlated with tumor burden at the time of T cell therapy. Recovery of normal B cell clones was observed in all patients, consistent with the waning persistence of the CAR-modified T cells and recovery of normal B cell lymphopoiesis.

Of the 4 patients undergoing allogeneic HSCT, 1 died of suspected pulmonary embolism at 2 months after transplantation while in CR with no evidence of disease, and the other 3 patients remained in CR at 6 weeks to 18 months after HSCT at the time of the report. In the patient who relapsed at 90 days after T-cell therapy and after high-dose steroid therapy for cytokine-mediated toxicities, the relapsed tumor cells exhibited the same malignant IgH rearrangement as the initial malignant clone, expressed target CD19 antigen at the same levels as before T-cell treatment, and remained sensitive to lysis by the CD19-targeted T cells. Relapse in this patient after achievement of MRD-negative status thus appeared to be due in part to the limited persistence of T cells resulting from the high-dose steroid therapy, rather than to escape from the effects of the CD19-targeted T cells.

Study in 2 children with relapsed/refractory pre-B-cell ALL

In a study reported by Grupp and colleagues,² 2 children (aged 7 and 10 years) with relapsed and refractory pre-B-cell ALL received infusions of T cells transduced with

anti-CD19 antibody and a T-cell signaling molecule (CTL019 CAR T cells) at a dose of 1.4×10^6 to 1.2×10^7 CTL019 cells/kg. CTL019 T cells expanded in both patients to a level that was more than 1,000 times higher than the initial engraftment level, with the T cells being identified in bone marrow and in cerebrospinal fluid, where they persisted at high levels for at least 6 months. Eight grade 3 or 4 adverse events were observed, consisting of febrile neutropenia, hypotension, acute vascular leak syndrome, and acute respiratory distress syndrome in 1 patient and febrile neutropenia, encephalopathy, elevated AST, and elevated ALT in the other patient. Cytokine-release syndrome and B cell aplasia developed in both patients. The cytokine-release syndrome was severe in 1 patient; cytokine blockade with etanercept and tocilizumab reversed the syndrome and did not interfere with expansion of the CAR T cells or reduce antileukemic efficacy.

Complete remission was observed in both patients and was ongoing in 1 child at 11 months after treatment at the time of the report. The other patient had a relapse with blast cells that no longer expressed CD19 at about 2 months after treatment. The investigators noted that although CAR-modified T cells are capable of killing aggressive, treatment-refractory acute leukemia cells, the emergence of tumor cells that no longer express the target antigen indicates a need to target other molecules in addition to CD19 in some patients with ALL.

References

1. Brentjens RJ, Davila ML, Riviere I, et al. CD19-targeted T cells rapidly induce molecular remissions in adults with chemotherapy-refractory acute lymphoblastic leukemia. *Sci Transl Med*. 2013;5:177ra38. doi: 10.1126/scitranslmed.3005930.
2. Grupp SA, Kalos M, Barrett D, et al. Chimeric antigen receptor-modified T cells for acute lymphoid leukemia. *N Engl J Med*. 2013;368:1509-1518.