New insights into effective targeted therapy for the treatment of adult acute lymphoblastic leukemia

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“Emerging therapies offer the promise of improved response rates and opportunities for long-term survival, with the potential for less toxicity than chemotherapy.”

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Despite high rates of initial response to frontline therapy, the prognosis of adult patients with acute lymphoblastic leukemia (ALL) remains unsatisfactory. The incorporation of intensive, pediatric-inspired chemotherapy regimens has led to survival improvements as compared with historical controls at least in patients aged up to 55 years [1]. The results of allogeneic hematopoietic stem cell transplantation (HSCT) for adults with ALL have also improved significantly over time [2] but it is unlikely that the sole intensification of regimens can continue to improve prognosis substantially. Treatment of relapsed ALL also remains a substantial challenge. Here only allogeneic HSCT can potentially lead to cure, but substantial numbers of patients cannot successfully be bridged to transplantation [3,4]. The modern era of genomics, including gene expression profiling, single polymorphism array analysis, array-comparative genomic hybridization and next-generation sequencing, has allowed the definition of new ALL sub-entities. Philadelphia-like ALL is characterized by genetic lesions similar to Philadelphia chromosome-positive ALL (but lacking the *BCR-ABL1* fusion gene), associated with *IKZF1* deletion, *CLRF2* overexpression and tyrosine kinase activating rearrangements involving *ABL1*, *JAK2*, *PDGFRB* and several other genes [5]. Other recently individualized clinical and biological ALL subtypes include *MEF2D*-rearranged ALL that are exquisitely sensitive to histone deacetylase inhibition, *ZNF384*-rearranged leukemia either diagnosed as B-cell lineage ALL with aberrant myeloid antigen expression or as mixed phenotype acute leukemia and *DUX4/ERG* ALL [6]. Early T-precursor ALL is characterized by lack of CD1a and CD8, weak CD5 expression, at least one myeloid/stem cell marker, a specific transcriptional profile and the possible involvement of several critical genes [7]. Several potential molecular targets in B-lineage ALL have therefore been identified [8]. However, except for the substantial therapeutic improvements with imatinib mesylate (NCT00327678) [9] or next-generation tyrosine kinase inhibitors in Philadelphia chromosome-positive ALL (NCT01424982) [10], target drugs are still unavailable for most of the newly defined molecular ALL subtypes. The most promising treatment advances for adults with ALL throughout the past years have resulted from immunotherapeutic approaches. In this setting, immunotherapy targeting cell surface molecules using monoclonal antibodies-derived

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therapeutics has extended the armamentarium. Likewise, harnessing the T-cell repertoire continues to yield another promising approach, represented by the activation of patient T cells directed against their own leukemic cells either through a bispecific antibody, or chimeric antigen receptor (CAR) modified T cells.

The use of monoclonal antibodies in adult ALL has rapidly grown over the recent years. Various categories of monoclonal antibodies are currently available, including naked antibodies killing the leukemic cells through antibody-dependent cell-mediated cytotoxicity, antibody-drug conjugates and antibody-immunotoxin conjugates which exert their cytotoxic effect via chemotherapeutic agents or natural toxins, and bi-specific T-cell engager (BiTE) antibodies that engage cytotoxic CD3⁺ T cells and drive them in contact to a cell surface antigen on leukemic cells [11]. Monoclonal antibodies targeting leukemic blast surface antigens offer the promise of unique mechanisms of action, distinct from traditional cytotoxics and a reduction in off-target adverse effects. Among these new treatments, three agents have been shown to be particularly promising: the anti-CD20 rituximab, the anti-CD22 calicheamicin-conjugated antibody inotuzumab ozogamicin and the anti-CD19/CD3 BiTE antibody blinatumomab that consist of variable binding domains of the respective parental antibodies connected by short linker peptides. Unconjugated monoclonal antibody rituximab targeting CD20 has had impressive effects when combined with cytotoxic regimens in CD20⁺ ALL (NCT00327678) [12]. A 2-year overall survival (OS) of 71 versus 52% and a 2-year event-free survival of 65 versus 52% were reported confirming previous retrospective reports from the MD Anderson Cancer Center [13] and from the German group [14]. Adding rituximab to standard chemotherapy was well tolerated with no significant differences in incidence of deaths between the randomized regimens. It should therefore be standard of care for these patients, although optimal dose schedule remains to be determined. In the pivotal INOVATE randomized Phase III study comparing inotuzumab ozogamicin, which links an anti-CD22 antibody to the chemotherapeutic agent calicheamicin, to standard chemotherapy, complete response (CR) was obtained in 81 versus 29%, respectively and minimal residual disease (MRD)-negativity in remitters was observed in 78% of cases versus 28% (NCT01564784) [15].

In older patients, the overall response rate was 97% with MRD-negativity in 100% of cases and the 2-year OS was 64% with the mini-Hyper-CVD (cyclophosphamide-vincristine-dexamethasone)-inotuzumab ± rituximab regimen (NCT01371630) [16]. Blinatumomab contains the variable domains of a CD19 antibody and CD3 antibody, which are joined by a nonimmunogenic linker. On binding to CD19, cytotoxic T cells become activated and induce cell death via the pore-forming perforin system. Blinatumomab, as compared with standard-of-care chemotherapy, improved the remission rate and OS in the Phase III TOWER study of adults with relapsed/refractory Philadelphia-negative B-cell precursor ALL (NCT02013167) [17]. The CR rate with blinatumomab was 39 versus 19% with standard-of-care chemotherapy. Median OS was 7.7 versus 4 months. This confirmed a previous report in adult patients with refractory/relapsed ALL showing a response rate of 43% and MRD response rate in remitters of 82% (NCT01466179) [18]. In MRD-positive ALL, blinatumomab allowed a switch to MRD-negativity in 80% of cases [19]. Most frequently reported adverse events were grade 3/4 reversible neurological events (17–19%) [19,20]. Monoclonal antibody development could, however, be challenged by the novel approach based on CART cells. This approach which uses T cells genetically engineered to express CAR targeting a cell surface molecule has shown increasing promise. The basic steps involved in the manufacture of CAR T cells comprise an apheresis procedure to collect autologous T cells followed by the genetical modification of T cells with the CAR construct and their *ex vivo* expansion. Before infusion, patients typically receive chemotherapy in an effort to induce tumor reduction and lymphodepletion to enhance CAR T-cell expansion and persistence *in vivo*. CD19 represents the target antigen for which most clinical data for CAR therapy exist. CD19-targeting CAR T cells displayed significant activity in patients with relapsed/refractory ALL [21–24]. Initial CR rates range from 70 to 90%. A high level of efficacy with CTL019 therapy was recently confirmed with a safety profile similar to that initially observed (NCT01626495) [25]. An overall remission rate of 83% was achieved with a significant rate of MRD-negativity and no cases of refractory cytokine release syndrome (CRS). CD22 is also highly expressed on ALL cells and anti-CD22-based immunotoxins are able to

induce anti-leukemic effects. The first-in-human anti-CD22 CAR T-cell therapy was shown safe, feasible and clinically active in ALL patients who have undergone prior CAR therapy [26]. MRD negative CRs were seen in patients who were both CAR-naïve or had previously been treated with anti-CD19 CAR and were CD19 negative. Adoptive transfer of mature allogeneic natural killer (NK) cells in the nontransplant or transplant setting has also been shown to be safe and feasible and its efficacy in human is under investigation [27]. Two approaches are currently in development: the adoptive transfer of NK cells from an alloreactive donor with a moderate lympho-depleting chemotherapy to induce homeostatic lymphocyte proliferation with transient expansion of the transferred allogeneic NK cells without the establishment of a permanent donor hematopoiesis [28]; and the transplant of hematopoietic stem cells after myeloablative therapy with the permanent engraftment of donor NK cells, establishing the donor killer immunoglobulin-like receptors phenotype in the patient [29].

Genetic alteration discoveries have recently transformed the way to consider the leukemogenic process and the approach to molecular diagnosis, risk stratification and targeting ALL. Now, the introduction of newer cytotoxic monoclonal antibodies and CAR modified T cells is opening a new age in the management of B-cell precursor adult ALL, yielding effective therapy in patients with particularly unfavorable characteristics and achievement of complete MRD clearance in most patients with high leukemic burden. MRD has been shown as the most important prognostic factor in ALL and thus a major component of a personalized treatment algorithm. Despite achievement of major molecular responses with these new treatments, the median duration of response is far from optimal. The mechanisms of resistance to monoclonal antibodies and CAR-modified T cells are not fully elucidated. After CD19-targeted therapy, two modes of disease recurrence have been observed: CD19 positive and CD19 negative. Extramedullary relapse in immunological sanctuaries has been observed after treatment with blinatumomab or CAR T cells. Blinatumomab might neither affect the leukemic stem cell compartment nor prevent the emergence of genetically unstable subclones [30]. Research is still active in order to improve the concept of bispecific antibodies. BiTE offers

favorable features related to its small size yielding target and effector cells in close proximity [31]. Other bispecific antibody formats are currently in development [32]. In this setting, the DART format has demonstrated more efficacy than the BiTE format with a higher association rate for CD3 and a lower dissociation rate for CD19 [33]. These new bispecific antibodies should become the next wave of antibody-based therapies.

With the development of specific monoclonal antibodies and CAR T cells, one of the main questions to be addressed is the role of allogeneic HSCT in the immunotherapy era. HSCT was proposed to all eligible patients, as immunotherapy was seen primarily as a bridge to transplant. However, no difference was noted in terms of survival when censoring for HSCT in blinatumomab and inotuzumab trials [15,20]. Similarly, it remains to be determined whether anti-leukemic effects induce by CAR therapy can translate into long-term cure and potentially even preclude the need for additional consolidation therapy such as allogeneic HSCT. Optimizing CAR-based therapy could abrogate the need for allogeneic HSCT in the treatment of ALL. If the acute inflammatory toxicity associated with CD19-CAR therapy can be prevented by treating patients with lower disease burdens, one could consider incorporating this therapy earlier in the course of the disease. CRS, resulting from the high magnitude of immune activation by CAR therapy, is the most significant treatment-related toxicity. The clinical signs of CRS with blinatumomab are similar to those observed with CAR T cells. Dose-limiting toxicity was represented by CNS adverse events. Risk factors for CRS include disease burden and initial starting dose. However, side effects were generally transient and reversible.

Emerging therapies offer the promise of improved response rates and opportunities for long-term survival, with the potential for less toxicity than chemotherapy. Based on initial highly promising activity, plans to incorporate inotuzumab or blinatumomab into frontline and salvage ALL treatment are underway. Inotuzumab combined with chemotherapy was shown useful in the treatment of older ALL patients [11]. The patients who received blinatumomab in the TOWER study were reported to have quality-of-life benefits that should be taken into consideration when evaluating monoclonal antibody therapy [17]. Based on recent results, blinatumomab was approved by US FDA and EMA for

the treatment of patients with relapsed/refractory ALL. Similarly, studies are underway to continue to optimize the incorporation of rituximab into ALL treatment regimens. Progress has also continued in the use of CAR T-cell therapy and recently published data highlight the pivotal role of NK cells in the development of novel immunologic approaches to the clinical management of acute leukemia [27]. Further improvements include the development of combinatorial approaches testing multiple monoclonal antibodies incorporated into the same regimen and/or combined with standard chemotherapy. It also remains to be determined whether they should be administered simultaneously or sequentially, and what should be the optimal sequence. Furthermore, if these new therapeutic strategies induce higher rates of

MRD-negativity, we can wonder whether this will reduce the need for intensive and prolonged chemotherapy schedules [11]. The expanded integration of the full spectrum of immunotherapeutic agents in the treatment of adult ALL is ongoing.

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References

- Huguet F, Leguay T, Thomas X *et al.* The upper age limit for a pediatric-inspired therapy in younger adults with Ph-negative acute lymphoblastic leukemia (ALL)? Analysis of the Graall-2005 study. *Blood* 128, 762 (2016).
- Giebel S, Labopin M, Socié G *et al.* Improving results of allogeneic hematopoietic cell transplantation for adults with acute lymphoblastic leukemia in first complete remission: an analysis from the Acute Leukemia Working Party of the European Society for blood and marrow transplantation. *Haematologica* 102, 139–149 (2017).
- Tavernier E, Boiron JM, Huguet F *et al.* Outcome of treatment after first relapse in adults with acute lymphoblastic leukemia initially treated by the LALA-94 trial. *Leukemia* 21, 1907–1914 (2007).
- Gökbuget N, Stanze D, Beck J *et al.* Outcome of relapsed adult lymphoblastic leukemia depends on response to salvage chemotherapy, prognostic factors, and performance of stem cell transplantation. *Blood* 120, 2032–2041 (2012).
- Herold T, Schneider S, Metzeler KH *et al.* Adults with Philadelphia chromosome-like acute lymphoblastic leukemia frequently have IGH-CRLF2 and JAK2 mutations, persistence of minimal residual disease and poor prognosis. *Haematologica* 121, 130–138 (2017).
- Mullighan CG. The evolving genomic landscape of acute lymphoblastic leukemia: subtypes, genes and targets. *Ann. Hematol.* 96(Suppl. 1), S32 (2017).
- Coustan-Smith E, Mullighan CG, Onclu M *et al.* Early T-cell precursor leukaemia: a subtype of very high-risk acute lymphoblastic leukaemia. *Lancet Oncol.* 10, 147–156 (2009).
- Yang JJ, Cheng C, Davidas M *et al.* Ancestry and pharmacogenomics of relapse in acute lymphoblastic leukemia. *Nat. Genet.* 43, 237–241 (2011).
- Chalandon Y, Thomas X, Hayette S *et al.* Randomized study of reduced-intensity chemotherapy combined with imatinib in adults with Ph-positive acute lymphoblastic leukemia. *Blood* 125, 3711–3719 (2015).
- Sasaki K, Jabbour EJ, Ravandi F *et al.* Phase II study of the frontline Hyper-CVAD in combination with ponatinib for patients with Philadelphia chromosome positive acute lymphoblastic leukemia. *Blood* 128(22), 757 (2016).
- Jabbour E, O'Brien S, Ravandi F, Kantarjian H. Monoclonal antibodies in acute lymphoblastic leukemia. *Blood* 125, 4010–4016 (2015).
- Maury S, Chevret S, Thomas X *et al.* Rituximab in B-lineage adult acute lymphoblastic leukemia. *N. Engl. J. Med.* 375, 1044–1053 (2016).
- Thomas DA, O'Brien S, Faderl S *et al.* Long-term outcome after hyper-CVAD and imatinib (IM) for de novo or minimally treated Philadelphia chromosome-positive acute lymphoblastic leukemia (Ph-ALL). *J. Clin. Oncol.* 28, 6506 (2010).
- Hoelzer D, Huettmann A, Kaul F *et al.* Immunotherapy with rituximab improves molecular CR rate and outcome in CD20+ B-lineage standard and high risk patients; results of 263 CD20+ patients studied prospectively in GMALL study 07/2003. *Blood* 116(21), 170 (2010).
- Kantarjian HM, DeAngelo DJ, Stelljes M *et al.* Inotuzumab ozogamicin versus standard therapy for acute lymphoblastic leukemia. *N. Engl. J. Med.* 375, 740–753 (2016).
- Jabbour E, O'Brien S, Sasaki K *et al.* Frontline inotuzumab ozogamicin in combination with low-intensity chemotherapy (mini-hyper-CVD) for older patients with acute lymphoblastic leukemia (ALL). *Blood* 126(23), 83 (2015).
- Topp MS, Stein A, Gökbuget N *et al.* Blinatumomab improved overall survival in patients with relapsed or refractory Philadelphia negative B-cell precursor acute lymphoblastic leukemia in a randomized, open-label Phase III study (TOWER). *Haematologica* 101(S1), 24–25 (2016).
- Topp MS, Gökbuget N, Stein AS *et al.* Safety and activity of blinatumomab for adult patients with relapsed or refractory B-precursor acute lymphoblastic leukaemia: a multicenter, single-arm, Phase II study. *Lancet Oncol.* 16, 57–66 (2015).
- Topp MS, Kufer P, Gökbuget N *et al.* Targeted therapy with the T-cell-engaging antibody blinatumomab of chemotherapy-refractory minimal residual disease in B-lineage acute lymphoblastic leukemia patients results in high response rate and prolonged leukemia-free survival. *J. Clin. Oncol.* 29, 2493–2498 (2011).
- Topp MS, Gökbuget N, Zugmaier G *et al.* Phase II trial of the anti-CD19 bispecific T cell-engager blinatumomab shows hematologic and molecular remissions in patients with relapsed or refractory

- B-precursor acute lymphoblastic leukemia. *J. Clin. Oncol.* 32, 4134–4140 (2014).
- 21 Lee DW, Shah NN, Sterler-Stevenson M *et al.* T cells expressing CD19 chimeric antigen receptors for acute lymphoblastic leukaemia in children and young adults: a Phase I dose-escalation trial. *Lancet* 385, 517–528 (2014).
- 22 Davila M, Riviere I, Wang X *et al.* Efficacy and toxicity management of 19–28z CAR T cell therapy in B cell acute lymphoblastic leukemia. *Sci. Transl. Med.* 6, 224ra25 (2014).
- 23 Turtle CJ, Hanafi LA, Berger C *et al.* CD19 CAR-T cells of defined CD4+:CD8+ composition in adult B cell ALL patients. *J. Clin. Invest.* 126, 2123–2138 (2016).
- 24 Maude SL, Frey N, Shaw PA *et al.* Chimeric antigen receptor T cells for sustained remissions in leukemia. *N. Engl. J. Med.* 371, 1507–1517 (2014).
- 25 Grupp SA, Laetsch TW, Buechner J *et al.* Analysis of a global registration trial of the efficacy and safety of CTL019 in pediatric and young adults with relapsed/refractory acute lymphoblastic leukemia (ALL). *Blood* 128(22), Abstract 221 (2016).
- 26 Shah NN, Sterler-Stevenson M, Yuan CM *et al.* Minimal residual disease complete remissions following anti-CD22 chimeric antigen receptor (CAR) in children and young adults with relapsed/refractory acute lymphoblastic leukemia (ALL). *Blood* 128(22), Abstract 650 (2016).
- 27 Handgretinger R, Lang P, André MC. Exploration of natural killer cells for the treatment of acute leukemia. *Blood* 127, 3341–3349 (2016).
- 28 Miller JS, Soignier Y, Panoskaltsis-Mortari A *et al.* Successful adoptive transfer and in vivo expansion of human haploidentical NK cells in patients with cancer. *Blood* 105, 3051–3057 (2005).
- 29 Pical-Izard C, Crocchiolo R, Granjeaud S *et al.* Reconstitution of natural killer cells in HLA-matched HSCT after reduced-intensity conditioning: impact on clinical outcome. *Biol. Blood Marrow Transplant.* 21, 429–439 (2015).
- 30 Thomas X. Blinatumomab: a new era of treatment for adult ALL? *Lancet Oncol.* 16, 6–7 (2015).
- 31 Rader C. DARTs take aim at BiTEs. *Blood* 117, 4403–4404 (2011).
- 32 Kontermann RE, Brinkmann U. Bispecific antibodies. *Drug Discov. Today* 20, 836–847 (2015).
- 33 Moore PA, Zhang W, Rainey GJ *et al.* Application of dual-affinity re-targeting molecules to achieve optimal redirected T-cell killing of B-cell lymphoma. *Blood* 117, 4542–4551 (2011).