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Review Article

Toxicities Associated with CAR-T Cell Therapies

Ugo Testa[1], Germana Castelli[1], Elvira Pelosi[1], Eugenio Galli[2] and Patrizia Chiusolo[2].

[1] Department of Oncology, Istituto Superiore di Sanità, ROME, Italy.
[2] Hematology Department, Fondazione Policlinico Universitario Agostino Gemelli, Rome, Italy.

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Abstract. Chimeric antigen receptor (CAR) T-cell therapy has improved the outcomes of patients with relapsed/refractory B-cell lymphomas, B-cell acute lymphoblastic leukemia, and multiple myeloma. However, CAR-T cell therapy is also associated with distinct toxicities that contribute to morbidity and mortality. A large number of studies now define the different toxicities associated with CAR-T cell therapy and have, in part, clarified their mechanisms. In particular, cytokine release syndrome (CRS) and immune effector cell-associated neurotoxicity syndrome (ICANS) are the two main acute toxicity events that occur after CAR-T cell infusion. Other CAR-T-related toxicities occur later after CAR-T cell infusion and include B-cell aplasia, hypogammaglobulinemia, infections, and cytopenias. Infections represent the main cause of non-relapse death observed in patients undergoing CAR-T cell therapy. Second primary malignancies are rare and are mainly represented by myeloid malignancies.

Keywords: Heparin-binding protein; Systematic review, Meta-analysis, Diagnosis, Prognosis, Infectious disease, Sepsis, Organ failure, Mortality.

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Correspondence to: Ugo Testa. Department of Oncology, Istituto Superiore di Sanità, Rome, Italy. E-mail: ugo.testa@iss.it

Introduction. Chimeric antigen receptor T-cells (CAR-T) are T-cells engineered with fusion proteins to target specific antigens present on tumor cells and generate an antitumor immune response. CAR-T cells now represent an established treatment for patients with relapsed/refractory large B-cell lymphomas (LBCL), B-cell acute lymphoblastic leukemia (B-ALL), and multiple myeloma (MM).

Five CD19-directed, namely Axicabtagene autoleucel (Axi-Cel), Brexucabtagene autoleucel (Brexu-Cel), Tisagenlecleucel (Tisa-Cel) Lisocabtagene maraleucel (Liso-Cel), and Obecabtagene autoleucel (Obe-cel), and two BCMA-directed CAR-T, namely Idecabtagene vicleucel (Ide-Cel) and Ciltacabtagene autoleucel (Cilta-Cel), have been approved against B-NHL, B-ALL, and MM so far.

The therapeutic efficacy of CAR-T reaches a plateau of progression-free survival ranging from 25-45%, mainly differing according to CAR-T product and type of disease. However, CAR-T is associated with unique early (within the first month) and late toxicities that require careful monitoring and prompt interventions. (**Figure 1**)

Non-relapse mortality (NRM) after CAR-T cell therapy. A recent systematic meta-analysis extended to 7,604 patients enrolled in 18 clinical trials, and 28 real-world studies analyzed the NRM observed after CAR-T in LBCL and MM patients.[1] NRM significantly differed for various diseases, varying across mantle cell lymphoma (10.6%), multiple myeloma (8%), LBCL (6.1%), and indolent lymphoma (5.7%).[1] CAR-T products impacted NRM in a disease-specific manner: for LBCL, Axi-Cel was associated with higher NRM

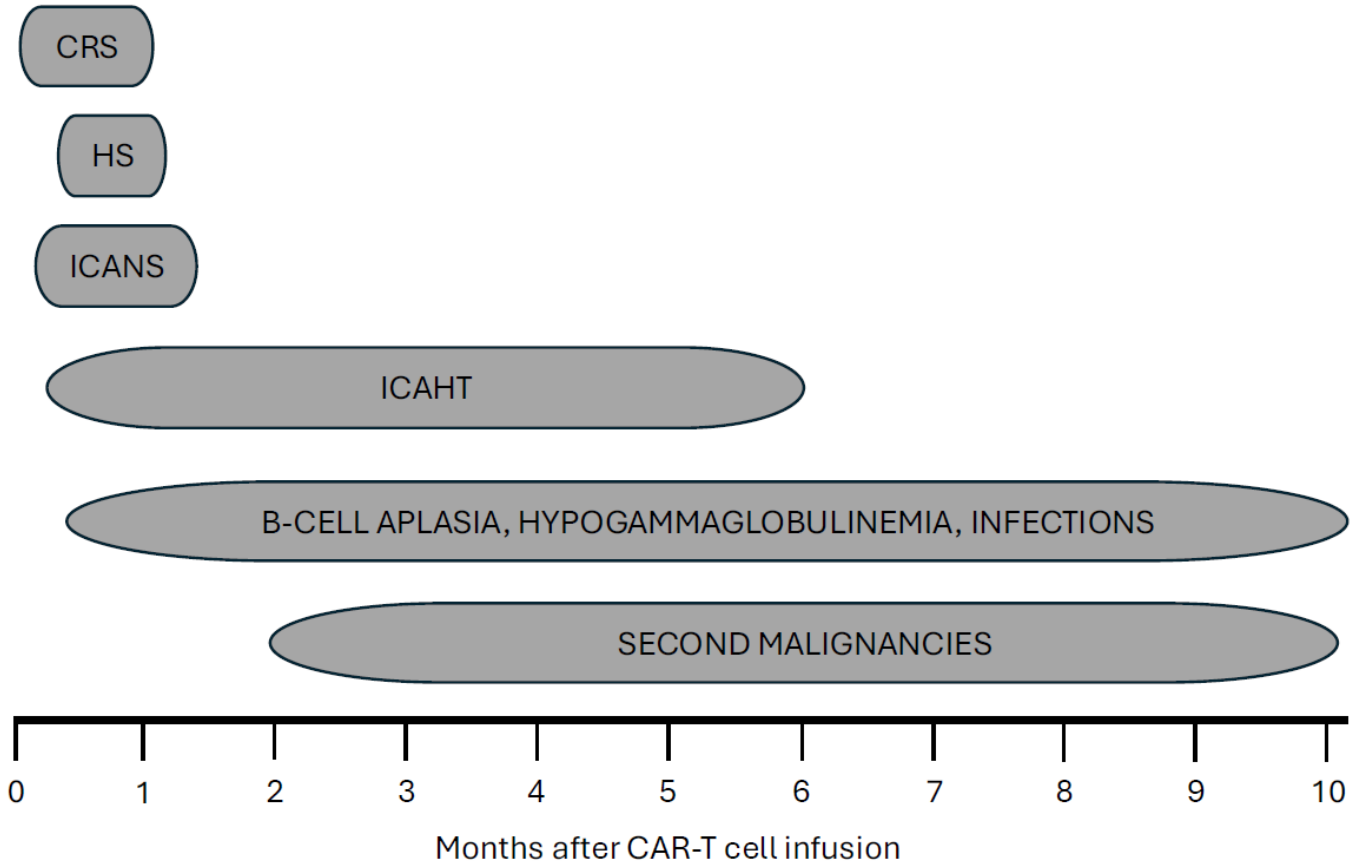

Figure 1. Timing of the main CAR-T cell-related adverse events. Approximate timeline of occurrence of main toxicities related to CAR-T cell infusion; for each of these toxicities, the typical time of onset is underlined by the start of each bar, the time to resolution by the end of the bar, and the duration by the length of the bar.

compared with Liso-Cel and Tisa-Cel (7.4% vs 3.8% vs 4.1%, p=0.004); for MM, Cilta-cel was associated with significantly higher NRM compared with Ide-Cel (15.2% vs. 6.3%, p<0.001).[1] 50.9% of NR deaths were attributed to infections, 7.8% to the development of a second malignancy, 7.3% to cardiovascular or respiratory events, 4.7% to cytokine release syndrome (CRS), 5.2% to immune effector cell-associated neurotoxicity syndrome (ICANS), and 1.5% to hemophagocytic lymphohistiocytosis.[1]

Cytokine Release Syndrome (CRS). The most prominent toxicity of CAR-T cells is CRS, a clinical syndrome affecting multiple organs that is characterized by a multitude of systemic symptoms. Usually, initial symptoms of CRS consist of fever and tachycardia. In more severe cases, CRS is associated with hypotension, hypoxia, capillary leak syndrome, multiple organ failures, and disseminated intravascular coagulation. CRS can onset in the very first days after CAR-T infusion, with a duration of some days in most cases. According to a recent consensus paper, CRS can be graded as follows:[2]

- Grade 1: Fever ≥38°C without hypotension or hypoxia.
- Grade 2: Fever ≥38°C with hypotension responsive to fluids or requiring low-dose vasopressors and/or hypoxia requiring low-flow supplemental oxygen (up to 6 L/min).
- Grade 3: Fever ≥38°C with hypotension requiring high-dose vasopressors and/or hypoxia requiring high-flow oxygen (>6 L/min) or non-invasive ventilation.
- Grade 4: Fever ≥38°C with hypotension requiring multiple vasopressors or inotropic support and/or hypoxia requiring invasive mechanical ventilation.
- Grade 5: Death related to CRS.

Morris and coworkers have described five key phases in the development of CRS.[3] Phase 1 involves CAR-T cell infusion, tumor site trafficking, and antigen recognition. Phase 2 includes CAR-T activation, proliferation, cytokine release, and tumor killing, defining the onset of CRS. Phase 3 is characterized by a systemic inflammatory response, corresponding to the peak of cytokines, endothelial cell injury with consequent vascular leakage, and organ dysfunction. Phase 4 involves damage to the blood-brain barrier (BBB), associated with the diffusion of cytokines and transmigration of CAR-T cells in the cerebrospinal fluid (CSF), leading to neurotoxicity. Phase 5 concludes with CAR-T apoptosis, a decrease of serum cytokine levels, and resolution of systemic inflammation, with long-term

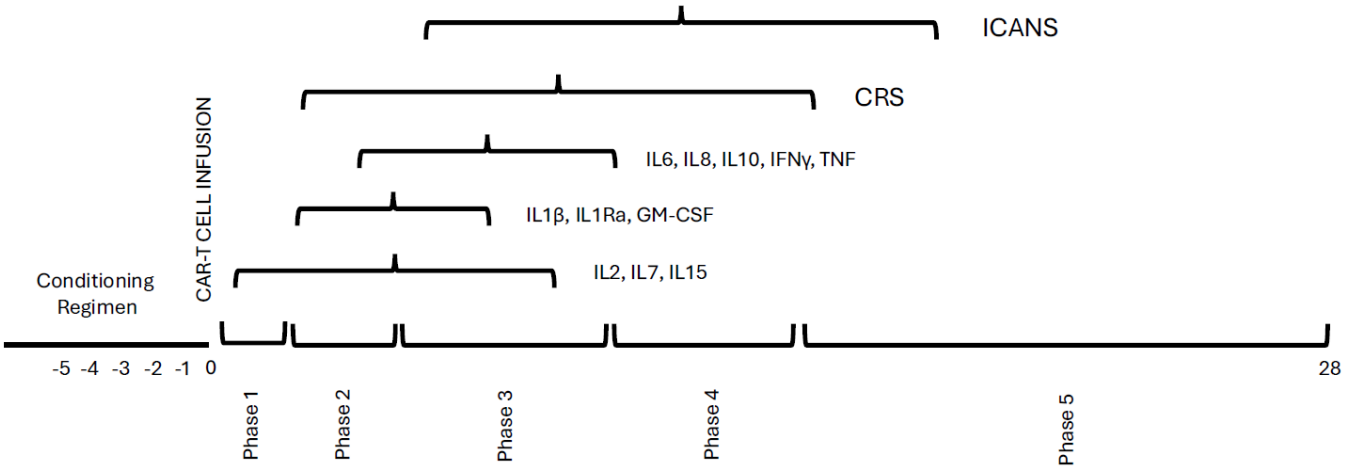

Figure 2. Acute toxicities (CRS and ICANS) occurring after CAR-T cell therapy. The period of first months after CAR-T cell infusion is subdivided into 5 phases, according to Morris et al.[2] The timing of CRS and ICANS development is outlined, indicating the most frequent times of CRS and ICANS peaks. Also, the kinetics of the production of groups of cytokines at different times after CRA-T cell infusion are shown.

memory CAR-T persistence in responders (**Figure 2**).[3]

The pathogenesis of CRS presupposes a complex interaction between activated T cells, tumor antigens, and tumor microenvironment involving IL-6, IL-10, IL-8, IFN-γ, and TNF-α.[3] Monocytes and macrophages have been identified as the main source of IL-6 and IL-1, which represent major drivers for the induction and progression of CRS.[4-5] IFN-γ released by activated CAR-T cells is a potent activator of monocytes/macrophages and was also associated with severe CRS.[6] In xenograft models, the inhibition or genetic deletion of IFN-γ induced decreased macrophage activation and inflammatory cytokines production, with reduced immune checkpoint expression,[7] also observed with pharmacological inhibition of IFN-γ by Emapalumab.[8] Interestingly, IFN-γ blockade did not affect CAR-T cells against $CD19^+$ hematologic malignancies but possibly reduced immune-related adverse effects.[6,7,9]

CD4+ T-cells play a major role in mediating the CRS.[10] Circulating $CD4^+$ CAR-T cells, but not CD8+, could be major drivers of CRS manifestations, together with a high tumor burden.[10]

Activated CAR-T cells also release proteolytic enzymes to trigger the induction of target cell death. CAR-Ts rapidly activate caspase-3 through the release of granzyme B, which induces the cleavage of gastrin E (GSMDE), a pore-forming protein highly expressed in B-leukemic cells, resulting in the induction of an extensive pyroptotic process.[11] Differently from apoptosis, pyroptosis is an inflammatory cell death. The activation of pyroptosis determines caspase-1-mediated cleavage of GSDMD in macrophages, with consequent release of cytokines and induction of CRS.[11] Knocking out GSMDE, depleting macrophages, or inhibiting caspase 1 abrogates CRS occurrence in murine models. High expression of GSMDE in tumor cells determines preferential pyroptosis and associates with CRS, despite apoptosis and pyroptosis are two cell death processes mediated by caspases.[11,12]

Another player in the pathophysiology of CRS is the activation of vascular endothelium. Endothelial cell activation is characterized by decreased Angioipoietin-1 (Ang-1) and increased levels of Ang-2 and von Willebrand Factor (VWF), sE-selectin, soluble intercellular adhesion molecule (sICAM-1) and soluble vascular cell adhesion molecule (sVCAM-1).[13] Coagulopathy can also be observed, with elevated prothrombin time, activated partial thromboplastin time values, D-dimer levels, and decreased fibrinogen levels and platelet counts in patients developing CRS, despite kinetics of fibrinogen is not clear in reason of its role as an acute phase protein. Particularly, the alteration of endothelial cell activation biomarkers, such as increased VWF levels and Ang-2/Ang-1 ratio, correlates with the development and degree of CRS.[14]

The modified version of the Endothelial Activation and Stress Index (mEASIX) is a transplant-derived score that associates with endothelial stress considering platelets, C-reactive protein, and lactate dehydrogenase. In patients with lymphoproliferative diseases treated with CAR-T, prothrombin time and activated thromboplastin time, fibrinogen, D-dimer, factor VIII, VWF level increased during CRS graded 2 or more, while platelet count and antithrombin levels decreased. A higher mEASIX score correlated with altered coagulopathy markers.[15] Baseline EASIX score combined with ferritin stratified patients in three groups with low, intermediate, and high risk of developing CRS.[16]

Interleukin 6 (IL-6) is one of the most significantly elevated cytokines during CRS occurring after CAR-T cell therapy. IL-6 is secreted by monocytic cells in response to CAR-T cell activation in a contact-independent mechanism following T-cell engagement of target cells.[17] IL-6 produced during CRS does not impact CAR-T cell cytotoxicity or transcriptional profiles, and

inhibition of IL-6 signaling in patients undergoing CAR-T cell treatments is a procedure not affecting CAR-T cell efficacy.[17]

Nowadays, current guidelines recommend administering Tocilizumab for grade >=2 CRS, with steroids required for refractory or ICANS-associated forms. Hypotension and hypoxia require specific treatment. Further treatments for severe and unresponsive forms involve anakinra, ruxolitinib, or high-dose steroids.[18]

IL-6 signaling contributes to many of the symptoms characterizing CRS, such as vascular endothelium activation and leakage, activation of complement, and coagulation cascade inducing disseminated intravascular coagulation. Blocking IL-6 Signaling using the monoclonal antibody Tocilizumab (targeting IL-6R) has dramatically improved the treatment of patients developing grade 2-4 CRS after CAR-T cell therapy, despite showing two disadvantages: the first is related to the cost and the second is that binding IL-6R, may result in the peak of serum IL-6, which could cross BBB and contribute to neurotoxicity. In order to obtain a more efficacious strategy for blocking IL-6 signaling, CD19- or BCMA-CAR-T cells were engineered to secrete an anti-IL-6 scFv and IL-1 receptor antagonist.[19] Next-generation CAR-T-secreting cytokine antagonists have been conceived, aiming at neutralization of IL-6 storm and block of IL-1 signaling or silencing the IL-6 gene (ssCAR-19) with a small hairpin RNA (shRNA).[20] Standard CD19 CAR-T and modified CAR-T (SSCAR-19) obtained similar complete remission rate (91.5% CR with median OS not reached with SSCAR-19, compared to 85% CR and median OS of 32.9 months in the standard CAR-T). Investigators observed grade 1-2 CRS in 53%, grade 3-4 CRS in 14.9%, and grade 1 ICANS in 4.2% of SSCAR-19 patients, compared to grade 1-2 CRS in 47.5% and grade 3-4 CRS in 37.5% and grade 1-2 ICANS in 10% of patients treated with stCAR-19.[20] Importantly, peak levels of serum IL-6, IL-2, and TNF-α levels were significantly lower in SSCAR-19 than in stCAR-19 patients.[20]

An alternative anti-IL-6 approach has been proposed. This approach exploits temperature-sensitive hydrogel conjugated with antibodies for IL-6, subcutaneously injected before the infusion of CAR-T cells. This hydrogel is efficient in reducing IL-6 levels and CRS .[21]

An optimal debulk of the disease may mitigate CRS; in this direction, Sakamura et al. have proposed treating patients with B-ALL with the anti-CD19 mAb Tafositamab before administering CD19-CAR-T cells.[22] In preclinical *in vivo* models, this two-step treatment reduced the incidence and severity of CRS and improved antitumor effects with better OS compared to CD9-CAR-T cells alone.[22]

CAR-T related neurologic events: ICANS. Neurotoxicity (ICANS) is the second most common adverse event associated with CAR-T cell therapies, defined as a disorder "resulting in the activation or engagement of endogenous or infused T-cells and/or other immune effector cells".[23]

The occurrence of ICANS stands around 30%, according to different reports and studies.

Usually, ICANS in CAR-T cell-treated patients is a mild entity associated with common symptoms, such as dysgraphia, word-finding difficulties, tremors, confusion, and somnolence. ICANS normally occurs at least 4 to 7 days after CAR-T infusion, with a duration limited to a few days in most patients; however, in a minority and variable proportion of patients, these symptoms progress in a few hours up to fatal outcomes due to fulminant cerebral edema.

ICANS can occur concomitantly with CRS or shortly after CRS subsides; being ICANS and CRS are etiologically related, there is often a correlation between their occurrence (**Figure 3**). This was also confirmed in a wider analysis of CD19, BCMA, and CD22-directed CAR-T cells.[24] The incidence of overall and grade ≥3 ICANS was more frequent in studies involving CAR-T containing CD28 costimulatory domains compared to those with 4-1BB costimulatory domains (**Figure 3**).

Neuroimaging of the CNS in patients with ICANS shows a symmetric distribution of abnormalities, particularly involving the thalamus (indicative of interstitial or vasogenic edema) and deep grey matter. These findings support the view of ICANS as promoted by a systemic inflammatory process.[25]

Several studies measuring cytokine levels in serum/plasma and cerebrospinal fluid (CSF) have supported their possible role in the pathophysiology of ICANS. Increased serum/plasma levels of IFN-γ and IL-15, as well as IL-6, IL-10, GM-CSF, IL-2, IL-1RA, and CXCL10, were associated with ICANS.[24]

Endothelial activation and disruption of the BBB are major mechanisms in the development of ICANS.

In 133 patients treated with CD19-CAR-T, high $CD19^{+}$ cells in bone marrow, high CAR-T cell dose, CRS, and preexisting neurologic comorbidities were associated with an increased risk of neurologic events.[26] Patients with severe ICANS exhibited evidence of endothelial cell activation, including disseminated intravascular coagulation, capillary leak syndrome, and increased BBB permeability.[26] The endothelial activation is also supported by higher Ang2 levels and lower Ang1/Ang2 ratio in patients with high-grade ì ICANS.[27] These observations are paralleled by the observation of increased serum concentrations of von Willebrand factor released by activated endothelium in patients with severe ICANS.[26]

Severe ICANS was associated with BBB disruption. High levels of proinflammatory cytokines such as IL-6, IL-8, MCP1, and IP10 were observed in the CSF of

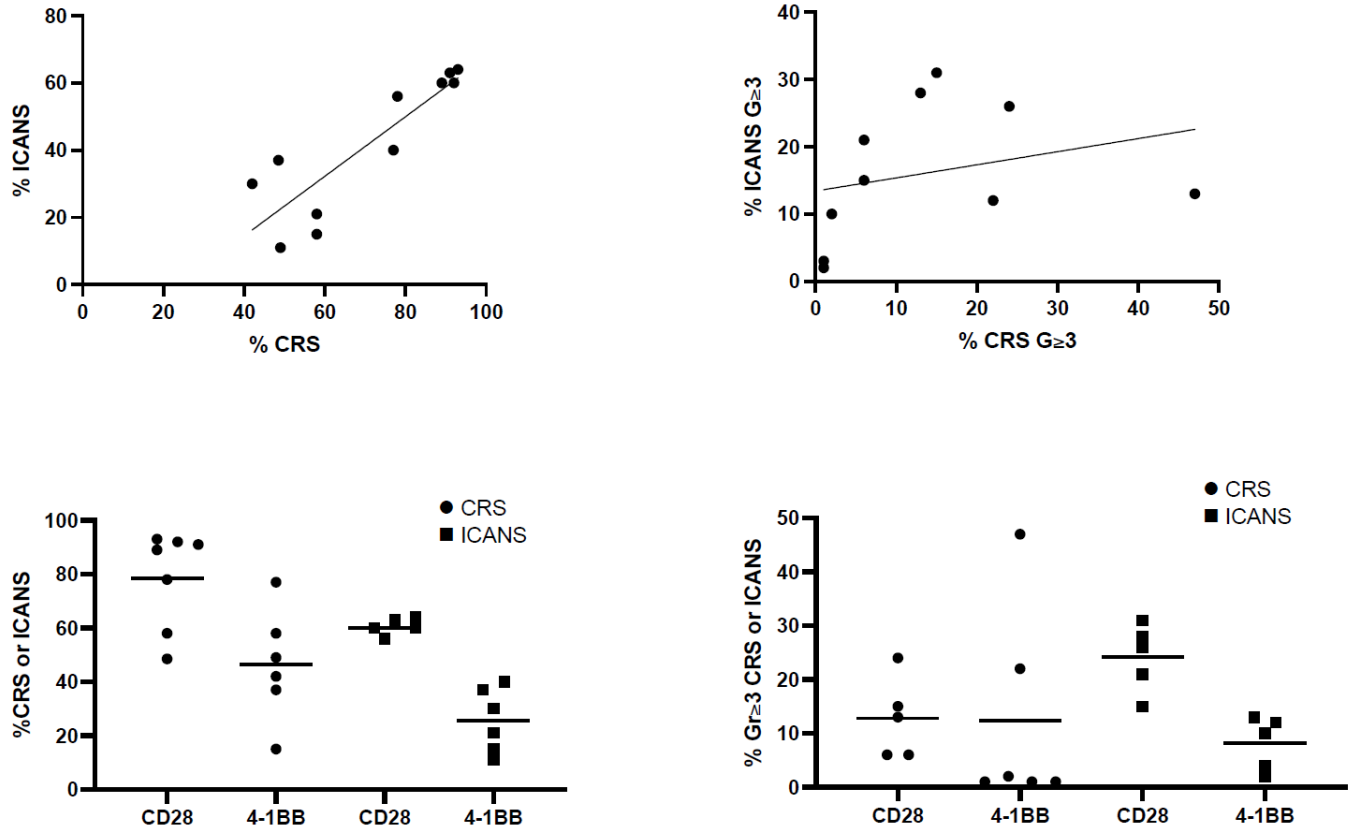

Figure 3. Correlation between the frequency of the occurrence of ICANS and CRS observed in the major clinical trials carried out with commercial CD19-CAR-T cells: left up panel: all ICANS and CRS events; right up panel: grade ≥3 ICANS and grade ≥3 CRS. Incidence of CRS and ICANS in clinical trials involving CD19-CAR-T cells containing CD28 or 4-1BB costimulatory domains: left down panel: all CRS and all ICANS; right down panel: grade ≥3 CRS and ICANS events.

patients with severe ICANS, suggesting a CNS production of these cytokines or passage through the BBB.[27]

Recent studies suggest that microglia activation also plays a relevant role in the development of ICANS. Studies by translocator protein positron-emission-tomography (PET) and imaging mass cytometry showed a shift from resting to activated microglia in patients with ICANS.[28] Accordingly, an animal model of ICANS showed that the TGFβ-activated kinase-1 (TAK1)-NFkB-p38 MAPK pathway is activated in microglia after CD19 T cell transfer, while its pharmacological inhibition improved neurocognitive activity in ICANS mice.[28]

CAR-T cells can release extracellular vesicles (EVs) carrying CAR constructs; in vivo, CART EVs are detectable as early as 1hr after CAR-T cell infusion, and their level in the plasma preceded the instauration of ICANS.[29]

A meta-analysis of 6 different clinical trials, as well as a real-world comparison, showed that axi-cel had a higher incidence of both CRS and ICANS compared to tisa-Cel, with ORs of 3.84 and 4.4, respectively.[30,31]

CAR-T related neurologic events: movement disorders. One case of progressive movement disorder resembling parkinsonism was reported in a patient with MM from the CARTITUDE-1 study approximately three months after BCMA-CAR T infusion.[32] In this patient, CAR-T cells persisted in circulation and in cerebrospinal fluid; basal ganglia showed lymphocyte infiltration.[32] Immunohistological studies documented BCMA expression on neurons and astrocytes of the patient's basal ganglia. Analysis of transcriptomic data evidenced BCMA mRNA expression in the caudate region of the normal human brain, thus suggesting that it could represent an on-target off-tumor effect of therapy.[32] Twelve out of 97 patients from the CARTIDUDE-1 trial reported non-ICANS neurotoxicity, with five patients' having movement and neurocognitive adverse events grade ≥3 parkinsonism in three of them.[33]

Overall, movement and neurocognitive disorders occurred in 5% of patients from CARTITUDE-1.[34] Potential factors associated with movement disorder were high baseline tumor burden, high baseline IL-6 levels, grade ≥2 CRS, the incidence of ICANS, high CAR-T cell expansion and persistence, and high absolute lymphocyte counts after CAR-T cell infusion.[34] Of these 5 patients, one recovered, one maintained persisting symptoms, and three died.[34] Importantly, the onset of movement disorders was delayed with respect to the onset of ICANS (27 days vs 5-9 days, respectively).[34] Additional neurologic toxicities observed in the CARTITUDE-1 study included also altered mental

status, concentration impairment, facial paralysis, diplopia, nystagmus, cranial nerve palsy, ataxia, sensory loss and peripheral motor and sensory neuropathies, occurring after resolution of CRS and ICANS.[34]

In another report from Karschnia et al., cells; 41% of patients with MM treated with BCMA-directed CAR-T developed ICANS, with two (2.6%) patients developing extra-pyramidal symptoms after the resolution of acute ICANS. In detail, one patient exhibited confusion and bradykinesia with increased muscle tone on day 22 post-CAR-T cell therapy with FDG-PET, showing bilateral hypometabolism localized at the level of the basal ganglia.[35] The second patient developed confusion, postural instability, mild tremor, bradykinesia, and hypophonia on day 19 after CAR-T cell infusion, with frontal region hypometabolism at FDG-PET.[35]

One more MM cilta-cel treated patient was reported by Gudera and coworkers, with Parkinson-like symptoms (hypomimia, oppositional paratonia, right-predominant rigidity, decreased arm swing, and dysgraphia), at day 24 after CAR-T infusion [36]. In this case, MRI displayed new bilateral hyperintensities located at the caudate head, putamen, and globus pallidus with concomitant lymphocytosis in the CSF and diffuse hypometabolism at the level of cerebral cortex in FDG-PET.[36]

The treatment of patients developing movement disorders after CAR-T cell treatment is challenging, with one experience proposing a role for cyclophosphamide.[37]

It is important to note that a more recent study, carried out using both immunohistochemistry and *in situ* RNA hybridization, questioned the expression of BCMA in normal human brain.[38] The mechanism of BCMA-CAR-T cell therapy-related movement disorders remains unclear.[38]

Hemophagocytic Lymphocytosis. Hemophagocytosis lymphocytosis (HLH) or macrophage activation syndrome (MAS) is a severe syndrome deriving from pathologic immune activation (characterized by fever, hyperferritinemia, hepatosplenomegaly, multi-organ failure, coagulopathy, neurologic toxicities, cytopenias and/or hypertriglyceridemia.[39] Secondary HLH may be observed both after CAR-T cell therapy and hematopoietic stem cell transplantation in 3%- 4% of cases.[40] Immune-effector-associated HLH, named IEC-HS, is typically described with no direct correlation with CRS, typically occurring 10-20 days after CAR-T cell infusion.[41]

Hyperferritinemia (normally above >10,000 ng/ml), cytopenia, hypertriglyceridemia, hypofibrinogenemia, coagulopathy, hepatic transaminitis, hyperbilirubinemia, severe neutropenia, elevated lactate dehydrogenase, and in some patients, hemophagocytosis are typical features of the syndrome. However, in many instances, the distinction between severe CRS and IEC-HS remains difficult.

Hemophagocytosis may be increased in patients treated with CD22-CAR-T, with an incidence of up to 40% in some reports.[42-44] A higher ratio of T cell/ NK cells in bone marrow and elevated inflammatory cytokine levels, including IL18m, may also be associated with the syndrome.[42] Increased IL-18 levels could contribute to the deregulation of the inflammasome observed in an HLH.[43]

Serum ferritin can be considered a biomarker related to hematologic disease. It is higher than 1,000 ng/ml during the early phases of the disease and increases to more than 10,000 ng/ml in correspondence with fully symptomatic HLH, with concurrent disseminated intravascular coagulation.[45]

In some instances, HLH is difficult to distinguish from severe CRS.

The pathophysiological mechanisms responsible for HLH remain largely undefined. Some observations suggest that IL-6 plays a less central role in HLH than in CRS: in fact, peak IL-6 levels did not differ substantially between patients who did not develop HLH. The majority of patients developing CAR-HLH had received prior Tocilizumab and developed HLH despite clinical amelioration of CRS.[42] Alternative cytokine pathways were responsible for the development of HLH. Other observations suggest that IL-1 could play a relevant role in HLH. IL-1β serum levels were markedly more elevated in patients developing HLH compared to those with CRS alone.[42] Anakinra and ruxolitinib are now proposed for the treatment of patients with IEC-HS.[42,46]

Second primary malignancies after CAR-T cell therapy. CAR-T patients share a high risk of developing treatment-related adverse events, including secondary primary malignancies. In 2023, the FDA released a statement concerning some reports on the occurrence of lymphomas and leukemias of the T-cell lineage observed in patients treated with CAR-T cells, either anti-CD19 or anti-BCMA in the context of clinical trials or post-marketing adverse events reporting.[47] According to these reports, the FDA extended the risk of the development of T-cell malignancies to all commercial CAR-T cell products.[47] In detail, these reports concerned 22 cases of T-cell malignancies within 2 years after CAR-T treatment, variously presenting as T-cell lymphomas, T-cell large granular lymphocytosis, peripheral T-cell lymphoma, and cutaneous T-cell lymphoma.[48] Importantly, in three cases in which DNA sequencing was performed, the CAR transgene was detected in the malignant clone.[48]

An exact evaluation of the number and type of adverse events occurring after CAR-T cell therapy remains difficult due to underreporting. Out of the more than 34,000 patients who have been treated, only 8,000 cases have been officially reported in the FDA-adverse

Events Reporting System (FAERS) database.[49]

The FAERS database allows to evaluate the type of SPMs occurring after CAR-T cell therapy. A wide registry analysis by Elsallab and coworkers evaluated 12394 CAR-T adverse event reports included in the FEARS database and observed that 536 (4.3% of SPM events.[50] The most frequent SPMs concerned leukemias, including myelodysplastic syndromes (38.8% of SPMs), acute myeloid leukemia (19.8% of SPMs), and T-cell large granular lymphocytic leukemia (0.4% of SPMs); hematological neoplasms excluding lymphomas and leukemias were less frequent (4.9% of SPMs); T-cell non-Hodgkin lymphomas were identified in 3.2% of all SPMs. Other tumors involved skin, mostly nonmelanomas (10.1%) CNS, respiratory tract, and gastrointestinal tract (3.9%, 3.7%, and 3.5% of the whole SPMs, respectively).[50,51] Analysis of SPM disproportionality in the CAR reports showed significantly higher reporting adding ratio (ROR) for myelodysplastic syndromes in patients treated with CAR-T cells.[50]

In order to explore if there was a higher risk of SPM after CAR-T cell treatment compared to chemotherapy alone, a subgroup analysis compared within FAERS the incidence of SPM between patients who received CAR-T therapy and those who received chemotherapy alone: CAR-T recipients seemed to experience higher incidences of some SPMs, such as T-cell lymphomas (ROR 5.0), squamous cell carcinoma of the skin (ROR 3.08) and MDS (ROR 1.50),[51] despite these data needs to be carefully contextualized. In this sense, a subgroup analysis of four clinical trials, which randomized CAR-T cell therapy versus standard of care, showed a similar risk of SPM with either treatment strategy.[45] In conclusion, these observations, although they raise awareness of SPM as a clinically relevant long-term adverse event in patients undergoing CAR-T cell therapy, do not support the view that CAR-T cell therapy increases the frequency of SPM compared to standard-of-care treatments.[52] It is worth noticing that solid tumors occurred in post-CAR-T cell therapy as SPM with a delayed manifestation compared to hematological malignancies.[51]

In a paper from Ghilardi and coworkers, SPM occurred in 3.6% out of 448 patients treated with commercial CAR-T cells for hematological malignancies at a median follow-up of 10.3 months. Only one patient developed a T-cell lymphoma.[53] Similar incidences were reported in other reports.[54]

It is important to underline that patients with non-Hodgkin lymphoma have an approximately five-fold higher incidence of second primary T-cell lymphoma over time compared to the general population;[55] and that chemotherapy itself determine an increased risk of SPM, particularly of MDS and AML;[56,57] the same is true for patients with MM treated with immunomodulatory agents.[58,59]

A recent paper identified the role of preexisting clonal hematopoiesis, age, and peripheral blood count impairment as possible risk factors for subsequent development of secondary myeloid neoplasia.[60]

B-cell aplasia, hypogammaglobulinemia and infections. B-cell aplasia is frequently observed in patients receiving CD19-directed CAR-T cell therapy and is the result of a direct on-target off-tumor toxicity, which can vary according to CAR-T product and underlying disease ranging between 60% and 80%.[61] B-cell recovery after Tisa-Cel tend to be slower may constitute a predictor of disease relapse. DoR was shorter in patients with recovery at <6 months after CAR-T cell infusion compared with those with B-cell recovering at 6-12 months or >12 months.[55,62]

Similar observations were not concordant when considering lymphomas.[63]

Hypogammaglobulinemia is frequently observed in patients after both CD19-directed and BCMA-directed CAR-T cell therapy.

Sutherland et al. reported that 60% of patients had hypogammaglobulinemia before CAR-T infusion, and 37% of patients developed a serious infection after CAR-T.[64] Preexisting hypogammaglobulinemia worsened after CAR-T therapy; the presence of hypogammaglobulinemia after CAR-T was associated with an increased risk of severe infections post-CAR-T cell therapy.[64] Risk factors for mortality included also hypogammaglobulinemia <400 mg /dL, together infections and related hospitalizations.[64]

Continuous long-term B-cell aplasia after CAR-infusion may be the surrogate of persistent CAR-T cell activity against normal B cells; in this sense, recovery from B-cell aplasia (BCA) could predict the loss of functional CAR-T cells against target antigen. However, B-cell aplasia could also derive from poor hematopoietic reserve.[65] In 57 LBCL patients undergoing CD19-CAR-T cell therapy, lower baseline CD4+, neutrophil counts and worse CAR-HEMATOTOX score (see later) predicted the development of long-term BCA, which was observed in 51% of patients.[66]

Infectious complications represent the main cause of non-relapse mortality after CAR-T, with risk factors varying according to the time elapsed since CAR-T infusion.

Early events occur from day 0 to day 30, are mainly represented by bacterial infections, and may be mostly related to the severe neutropenia typical of those timeframes.[67] These early events may be also exacerbated by the immunosuppressive treatments used to mitigate CRS and ICANS. Late infectious events occur after day 30 and are related to the prolonged depletion of B and T cells, and hypogammaglobulinemia. CD19 and BCMA CAR-T cells induce different immune

deficits due to the expression of their respective targets on normal B cells at different stages of maturation, being extended in the case of BCMA also to mature plasma cells.[67] Late infectious events may be typically related to viruses, bacteria, or fungi.[67]

Hematologic toxicities. Hematologic toxicities are frequently observed after CAR-T cell therapy of hematologic patients and include a spectrum of adverse events, such as cytopenias, B-cell aplasia and coagulopathies. The European Hematology Association (EHA) and the European Society for Blood and Bone Marrow Transplantation (EBMT) classified cytopenias occurring after CAR-T cell therapy as Immune Effector Cell-Associated hematological toxicity (ICAHT).[68] Prolonged ICAHT can be subdivided according to their timing of occurrence into early (0-30 days) or late (after 90 days). Despite ICAHT only takes into account absolute neutrophil counts, a deficiency of hemoglobin and platelets count can frequently be associated.

Cytopenias usually last less than 6 months. Some risk factors have been identified for predicting the development of cytopenias.

CAR-T induced inflammation, prior lines of chemotherapy, baseline hematopoietic reserve, and CRS is are the most-known key determinant in the development of hematologic toxicities.[69]

The CAR-HEMATOTOX score, introduced by Rejeski et al., represents a well-validated prognostic tool for predicting the risk of enduring cytopenias and is based on the evaluation of some inflammatory markers, such as ferritin and C-protein reactive levels, together with blood cell counts.[70] The CAR-HEMATOTOX index predicts long-term hematotoxicity and severe infections in patients receiving CAR-T cell therapy for the majority of current indications.

ICAHT occurrence and severity was documented in LBCL, MCL and MM patients undergoing CAR-T cell therapy, despite could be partially missignificant for patienst with B-ALL, where involvement of bone marrow could have some more relevant role.[71] Thus, the ALL-HEMATOTOX was developed, replacing ferritin with medullary disease burden. The ALL-HEMATOTOX score associated with severe prolonged neutropenia and appropriately identified 47% of patients as high-risk patients.[71]

Strati and coworkers have explored a large group of LBCL patients who have undergone treatment with Axi-Cel; 53% of these patients developed CAR-T -related prolonged cytopenia;[72] patients with prolonged cytopenia experienced more frequently CRS and ICANS.[72] Single cell analysis of bone marrow cells suggested that a subset of IFN-γ-expressing $CD8^+$ T cells may be responsible for prolonged cytopenia, together with an associated enrichment of IFN-γ signaling within bone marrow HSCs.[72]

Treatment of cytopenia may safely include granulocyte colony-stimulating factors (G-CSF), erythropoietin, or thrombopoietin receptor agonists, together with transfusions of irradiated blood products or, more episodely, with stem cells-boosts.[73-75]

Generation of CAR-T cells with a lower toxicity profile. The reduction of toxicities associated with CAR-T cell therapies could be achieved by modifying some constituents of CAR-T cells.

Several strategies have been explored in this sense. Obecabtagene autocel (Obe-Cel) is a CD19-directed CAR-T cell product based on a single chain variable fragment (scFv) with a low antigen affinity owing to rapid dissociation.[76] Obe-cel was evaluated in R/R B-ALL patients, showing a good safety profile with no patient experiencing ≥grade 3 CRS and 15% displaying grade 3 ICANS, rapidly regressing to ≤grade 1 after glucocorticoid treatment.[76] Complete responses were achieved in 78% of cases after Obe-cel.[77]

Another study was based on the development of a CD19-directed CAR construct containing a CD28 costimulatory domain and CD8α hinge and transmembrane domain instead of CD28 hinge, transmembrane, and costimulatory domains, as observed in Axi-Cel.[78] Those CAR-T cells (named Hu19-CD8282) were infused in patients with LBCL, resulting in lower serum cytokine levels and markedly reduced incidence of ICANS compared to fully CD28 co-stimulated CAR-T (5% vs 50%, respectively).[78]

An alternative approach consisted in the interposition of an intracellular domain from Toll-like receptor 2 (TLR2) between CD28 and CD3ζ; preclinical studies have shown that a CD19-CAR T cell generated with this modified structure resulted in reduced production of cytokines such as GM-CSF and IFN-γ, with maintained production of IL-7.[79] The third generation of CAR-T cells developed with this CAR construct was evaluated in 21 LBCL patients (ENABLE clinical trial), resulting in a good safety profile with grade 1-2 CRS in 62% of cases and no grade 3 CRS and no ICANS of any grade.[79]

Since redundancy of CD28 and CD3zeta signaling accelerates T cell differentiation and promotes t cell exhaustion, a further strategy (1XXCARs) consisted of inactivating point mutations in the two distal immunoreceptor tyrosine-based activation motifs (ITAMs) and retaining one functional proximal ITAM; 1XXCARs could exert a significant antitumor activity at lower doses, with reduced incidence of G3-4 CRS and ICANS (3% and 4%).[80]

Another strategy involves designing CARs that incorporate into their structure molecules inhibiting inflammatory cytokines, such as introducing in the CAR-T construct an inductor of an anti-IL6 scFv, an IL1 receptor antagonist (IL-1RA), or an IFNγ antagonist.[81-82]

Yoshikawa and coworkers developed a CAR (G6/7R)

composed of extracellular IL-6 receptors and intracellular IL-7 receptors with a constitutive mutation.[83] G6/7R-expressing CAR-T cells significantly reduce IL-6 levels *in vitro* and demonstrate durable antitumor efficacy in multiple tumor models.[83]

Mice treated with CAR-T cells secreting Tocilizumab-derived single-chain variable fragments were described as having reduced CRS-related toxicity and enhanced antitumor efficacy.[84]

Another approach was based on selecting T cell populations for the generation of CAR-T cells. In this context, Aldoss et al. reported the evaluation of the safety and efficacy of memory-enriched CD19-CAR-T cells in R/R B-ALL.[81] The results observed in 48 R/R B-ALL patients showed a CR rate of 87%, with a good safety profile (7% grade r CRS and 17% of grade ≥3 ICANS).[85]

Conclusions. CAR-T cell therapies are effective for many hematological malignancies, but toxicities may still limit outcomes. Thus, considerable improvements have been made in the treatment of these toxicities, such as CRS and iCANS.

Inflammation is still the major determinant of many toxicities. CRS and ICANS are the most frequent CAR-T-related acute toxicities, depending on CAR-T lytic action, cytokines, and endothelial activation. Treatment of CRS has considerably improved, particularly through the use of the IL-6 receptor antagonist Tocilizumab and glucocorticoids.

Movement disorders are observed in a minority of patients treated with BCMA-CAR-T cells and represents a severe subtype of ICANS, with difficult so far.

B-cell aplasia and hypogammaglobulinemia are related to an on-target toxicity mechanism and require adequate therapeutic management.

Infectious events represent the most frequent cause of nonrelapse mortality in patients treated with CAR-T, resulting from immunosuppression consequent to cytopenias and BCA.

Late-onset hematological toxicities are due to the exposition of bone marrow to logistic flares and preexisting conditions, resulting in impairment of bone marrow function.

Secondary malignancies can be observed after CAR-t cell and can involve myeloid therapy related malignancies. Patients undergoing CAR-T cell therapies should be monitored for secondary malignancies indefinitely after CAR-T cell treatment.

The development of new constructs and products could reduce the inflammatory-related toxicities of CAR-T cells, resulting in a more favorable toxicity profile.

References:

1. Cordas dos Santos, D.; Tix, T.; Shouval, R.; Gafter-Gvili, A.; Alberge, J.B.; Scheffer Cliff, E.; Theurich, S.; Bargwelt-Baildon, M.; Ghobrial, I.M.; Subklewe, M.; et al. A systematic review and meta-analysis of nonrelapse mortality after CAR T cell therapy. Nat Med 2024; 30: 2667-2678. https://doi.org/10.1038/s41591-024-03084-6 PMid:38977912 PMCid:PMC11765209
2. Lee DW, Santomasso BD, Locke FL, Ghobadi A, Turtle CJ, Brudno JN, Maus MV, Park JH, Mead E, Pavletic S, Go WY, Eldjerou L, Gardner RA, Frey N, Curran KJ, Peggs K, Pasquini M, DiPersio JF, van den Brink MRM, Komanduri KV, Grupp SA, Neelapu SS. ASTCT Consensus Grading for Cytokine Release Syndrome and Neurologic Toxicity Associated with Immune Effector Cells. Biol Blood Marrow Transplant. 2019 Apr;25(4):625-638. https://doi.org/10.1016/j.bbmt.2018.12.758 PMid:30592986
3. Morris EC, Neelapu SS, Giavridis T, Sadelain M. Cytokine release syndrome and associated neurotoxicity in cancer immunotherapy. Nat Rev Immunol 2022; 22: 85-96. https://doi.org/10.1038/s41577-021-00547-6 PMid:34002066 PMCid:PMC8127450
4. Giavridis T, van der Stegen SJG, Eyquem J, Hamieh M, Piersigilli A, Sadelain M.CAR T cell-induced cytokine release syndrome is mediated by macrophages and ablated by IL-1 blockade. Nat Med 2018; 24: 731-738. https://doi.org/10.1038/s41591-018-0041-7 PMid:29808005 PMCid:PMC6410714
5. Norelli M, Camisa B, Barbiera G, Falcone l, Purevdorj A, Genua M, Sanvito F, Ponzoni M, Doglioni C, Cristofori P. Monocyte derived IL-1 and IL-6 are differentially required for cytokine release syndrome and neurotoxicity due to CAR-T cells. Nat Med 2018; 24: 739-748. https://doi.org/10.1038/s41591-018-0036-4 PMid:29808007
6. Teachey DT, Lacey SF, Shaw PA, Malenhorst JJ, Maude SL, Frey N, Pequinot E, Gonzalez VE, Chen F, Finklestein J, et al. Identification of predictive biomarkers for cytokine release syndrome after chimeric antigen receptor T-cell therapy for acute lymphoblastic leukemia. Cancer Discov 2016; 6: 664-679. https://doi.org/10.1158/2159-8290.CD-16-0040 PMid:27076371 PMCid:PMC5448406
7. Bailey SR, Vatsa S, Larson RC, Bouffard AA, Scarfò I, Kann MC. Blockade or deletion of IFN reduces macrophage activation without compromising CAR T-cell function in hematologic malignancies. Blood Cancer Discovery 2022; 3: 136-153. https://doi.org/10.1158/2643-3230.BCD-21-0181 PMid:35015685 PMCid:PMC9414118
8. Manni S, Del Bufalo F, Merli P, Silvestris DA, Guercio M, Caruso S. Neutralizing IFNg improves safety without compromising efficacy of CAR-T cell therapy in B-malignancies. Nat Commun 2023; 14: 423. https://doi.org/10.1038/s41467-023-38723-y PMid:37296093 PMCid:PMC10256701
9. McNerney KO, DiNafia AM, Teachey DT, Grupp SA, Maude SL. Potential role of IFN- inhibition in refractory cytokine release syndrome with CART cell therapy. Blood Cancer Discover 2022; 3: 90-94. https://doi.org/10.1158/2643-3230.BCD-21-0203 PMid:35015687 PMCid:PMC9245357
10. Boulch M, Cazaux M, Cuffel A, Ruggiu M, Allain V, Corre B. A major role for CD4+ T cells in driving cytokine release syndrome during CAR T cell therapy. Cell Rep Med 2023; 4: 101161. https://doi.org/10.1016/j.xcrm.2023.101161 PMid:37595589 PMCid:PMC10518592
11. Liu Y, Fang Y, Chen X, Wang Z, Liang X, Zhang T. Gasdermin E-mediated target cell pyroptosis by CAR T cells triggers cytokine release syndrome. Sci Immunol 2020; 5: eaax7969. https://doi.org/10.1126/sciimmunol.aax7969 PMid:31953257
12. Jiang M, Qi L, Li L, Li Y. The caspase-3/GSMDE signal pathway as a switch beteen apoptosis and pyroptosis in cancer. Cancer Death Discov 2020; 6: 112. https://doi.org/10.1038/s41420-020-00349-0 PMid:33133646 PMCid:PMC7595122
13. Hong F, Shi M, Cao J, Wang Y, Gong Y, Gao H, Li Z, Zheng J, Zang L.,He A, et al. Predictive role of endothelial activation in cytokine release syndrome after chimeric antigen receptor T cell therapy for acute lymphoblastic leukemia. J Cell Mol Med 2021; 25: 11063-11074. https://doi.org/10.1111/jcmm.17029

PMid:34734474 PMCid:PMC8650023
14. Hay KA, Hanafi LA, Li D, Gust J, Llies C, Wufel M, Lopez JA, Chen J, Chung D, Harju-Baker S, et al. Kinetics and biomarkers of severe cytokine release syndrome after CD19 chimeric antigen receptor-modified T-cell therapy. Blood 2017; 130: 2295-2306. https://doi.org/10.1182/blood-2017-06-793141 PMid:28924019 PMCid:PMC5701525
15. Galli E, Sorà F, Hohaus S, Fresa A, Pansini I, Autore F. Endothelial activation predicts disseminated intravascular coagulopathy cytokine release syndrome and prognosis in patients treated with anti-CD19 CAR-T cells. Br J Haematol 2023; 201: 86-94. https://doi.org/10.1111/bjh.18596 PMid:36503182
16. Greenbaum U, Strati P, Saliba RM, Torres J, Rondon G, Nieto Y, Hosing C, Srour SA, Westin J, Fayad LE, et al. CRP and ferritin in addition to EASIX predict CAR-T related toxicity. Blood Adv 2021; 5: 2799-2806. https://doi.org/10.1182/bloodadvances.2021004575 PMid:34264268 PMCid:PMC8341350
17. Singh N, Hofmann TJ, Gershenson Z, Lebine BL, Grupp S, Teachey DT, Barrett DM. Monocyte lineage-derived IL-6 does not affect chimeric antigen receptor T-cell function. Cytotherapy 2017; 19: 867-880. https://doi.org/10.1016/j.jcyt.2017.04.001 PMid:28506444 PMCid:PMC6676485
18. Hayden PJ, Roddie C, Bader P, Basak GW, Bonig H, Bonini C, Chabannon C, Ciceri F, Corbacioglu S, Ellard R, Sanchez-Guijo F, Jäger U, Hildebrandt M, Hudecek M, Kersten MJ, Köhl U, Kuball J, Mielke S, Mohty M, Murray J, Nagler A, Rees J, Rioufol C, Saccardi R, Snowden JA, Styczynski J, Subklewe M, Thieblemont C, Topp M, Ispizua ÁU, Chen D, Vrhovac R, Gribben JG, Kröger N, Einsele H, Yakoub-Agha I. Management of adults and children receiving CAR T-cell therapy: 2021 best practice recommendations of the European Society for Blood and Marrow Transplantation (EBMT) and the Joint Accreditation Committee of ISCT and EBMT (JACIE) and the European Haematology Association (EHA). Ann Oncol. 2022 Mar;33(3):259-275. https://doi.org/10.1016/j.annonc.2021.12.003 PMid:34923107
19. Xue L, Xu Q, Wang L, Yang X, Zhang Y, Hua X, Chai X, Yang Y, Chen Y, Tao, G.; et al. Chimeric antigen receptor T cells self-neutralizing IL6 storm in patients with hematologic malignancy. Cell Discov 2021; 7:84. https://doi.org/10.1038/s41421-021-00299-6 PMid:34518515 PMCid:PMC8437938
20. Kang L, Ma J, Lou X, Tang X, Xue S, Chen S, Qiu H, Miao M, Han Y, Wang Y, et al. Efficacy and safety of interleukin-6 -knockdown CD19-targeted CART cells (ssCART-19) for relapsed/refractory B-ALL. Blood 2023; 142 (suppl.1): 704. https://doi.org/10.1182/blood-2023-172615
21. Li X, Gong N, Tian F, Zhang S, Zhang Y, Wang Y, Qing G, Wang Y, Li F, Xu Y, et al. Suppression of cytokine release syndrome during CAR-T cell therapy via a subcutaneously injected interleukin-6-adsorbing hydrogel. Nat Biomed Engin 2023; 7: 1129-1141. https://doi.org/10.1038/s41551-023-01084-4 PMid:37696984
22. Sakamura RL, Roman CM, Harvei P, Siegler EL, Girsch JH, Sirpilla OL, Stewart CM, Yun K, Can I, Ogbodo OJ, et al. CD19 occupancy with Tafasitamab increases therapeutic index of CART19 cell therapy and diminishes severity of CRS. Blood 2024; 143: 258-271. https://doi.org/10.1182/blood.2022018905 PMid:37879074 PMCid:PMC10808250
23. Santomasso BD, Nastoupil LJ, Adkins S, Lacchetti C, Schneider BJ, Anadkat M, Atkins MB, Brassil KJ, Caterino JM, Chau I, Davies MJ, Ernstoff MS, Fecher L, Funchain P, Jaiyesimi I, Mammen JS, Naidoo J, Naing A, Phillips T, Porter LD, Reichner CA, Seigel C, Song JM, Spira A, Suarez-Almazor M, Swami U, Thompson JA, Vikas P, Wang Y, Weber JS, Bollin K, Ghosh M. Management of Immune-Related Adverse Events in Patients Treated With Chimeric Antigen Receptor T-Cell Therapy: ASCO Guideline. J Clin Oncol. 2021 Dec 10;39(35):3978-3992. https://doi.org/10.1200/JCO.22.00279
24. Gust J, Ponce R, Liles C, garden GA, Turtle CJ, Cytokines in CART cell-associated neurotoxicity. Front Immunol 2020; 11: 577027. https://doi.org/10.3389/fimmu.2020.577027 PMid:33391257 PMCid:PMC7772425
25. Ferrari CJ, Bhutani M. Mechanisms and management of CART toxicity. Front Oncol 2024; 14: 1396490. https://doi.org/10.3389/fonc.2024.1396490 PMid:38835382 PMCid:PMC11148294
26. Gust J, Hay KA, Hanafi LA, Li D, Myerson D, Gonzalez-Cuyar LF, Yeung C, Liles WC, Wurfel M, Lopez JA, Chen J, et al. Endothelial activation and blood-brain barrier disruption in neurotoxicity after adoptive immunotherapy with CD19 CAR-T cells. Cancer Discob 2017; 7: 1404-1419. https://doi.org/10.1158/2159-8290.CD-17-0698 PMid:29025771 PMCid:PMC5718945
27. Santomasso BD, Park JH, Salloum D, Riviere I, Flynn J, Mead E, Helton E, Wang X, Senechal B, Purdon T, et al. Clinical and biological correlates of neurotoxicity associated with CAR T-cell therapy in patients with B-cell acute lymphoblastic leukemia. Cancer Discov 2018; 8: 958-971. https://doi.org/10.1158/2159-8290.CD-17-1319 PMid:29880584 PMCid:PMC6385599
28. Vinnekota JM, Biavasco F, Schwabenland M, Chhatbar C, Adams RC, Erny D, Duquesne S, El Khawansky NE, Schmidt D, et al. Targeting TFG□-activated kinase-1 activation in microglia reduces CAR T immune effector cell-associated neurotoxicity syndrome. Nat Cancer 2024; 5: 1227-1249. https://doi.org/10.1038/s43018-024-00764-7 PMid:38741011
29. Storci G, De Felice F, Ricci F, Santi S, Masselodi D, Bertuccio SN, Laprovitera N, Dicataldo M, Rossini L, De Matteis S, et al. CAR+ extracellular vesicles predict ICANS in patients with B cell lymphomas treated with CD19-directed CAR-T cells. J Clin Invet 2024; 134: e173096. https://doi.org/10.1172/JCI173096 PMid:38833312 PMCid:PMC11245152
30. Bachy E, LeGouill S, DiBlasi R, Sesques P, Manson G, Cartron G, Baeuvois D, Roulin L, Gros FX, Rubio MT, et al. A eral-world comparison of tisageleuclecel and axicabtagene ciloleucel CART cells in relapsed or refractory diffuse large B cell lymphoma. Nat Med 2022; 28: 2145-2154. https://doi.org/10.1038/s41591-022-01969-y PMid:36138152 PMCid:PMC9556323
31. Liao CT, Zeng L, Lu S, Zheng S, Guo B, Ke Q, Wang M, Sun J, Rang C, He S, et al. Comparison of the efficacy and safety of axi-cel and tisa-cel based on meta-analysis. J Cancer 2024; 15: 5729-5741. https://doi.org/10.7150/jca.99427 PMid:39308670 PMCid:PMC11414620
32. Van Oekelen O, Aleman A, Upadhyaya B, Schnakenburg S, Madduri D, Gavane S, Teruya-Feldstein J, Crary JF, Fowkes ME, Stacy CB, et al. Neurocognitive and hypokinetic movement disorder with features of parkinsonism following BCMA-targeting CAR-T cell therapy. Nat Med 2021; 27: 2099-2103. https://doi.org/10.1038/s41591-021-01564-7 PMid:34893771 PMCid:PMC8678323
33. Bardeja JG, Madduri D, Usmani SZ, Jakubowiak AJ, Agha M, Cohen AD. Ciltacabtagene autoleucel, a B-cell maturation antigen-directed chimeric antigen receptor T-cell therapy in patients with relapsed or refractory multiple myeloma (CARTITUDE-1): a phase 1b/2 open-label study. Lancet 2021; 398: 314-324. https://doi.org/10.1016/S0140-6736(21)00933-8 PMid:34175021
34. Cohen AD, Parekh S, Santomasso B, Perez-Larraya JG, van de Donk N, Arnulf B, Mateos MV, Lendval N, Jackson CC, De Bragnasca K, et al. Incidence and management of CAR-T neurotoxicity in patients with multiple myeloma treated with ciltacabtagene autoleucel in CARTITUDE studies. Blood Cancer J 2022; 12: 32. https://doi.org/10.1038/s41408-022-00629-1 PMid:35210399 PMCid:PMC8873238
35. Karschnia P, Miller KC, Yee aj, Rejeski K, Connor Johnson C, Raje N, Frigault MJ, Dietrich J. Neurologic toxicities following adoptive immunotherapy with BCMA-directed CAR T cells. Blood 2023; 142: 1243-1247. https://doi.org/10.1182/blood.2023020571 PMid:37471607
36. Gudera JA, Baehring JM, Karschnia P. Parkinsonism following chimeric antigen receptor T-cell therapy. JAMA Neurol doi:10.1001/jamaneurol.2024.2506. https://doi.org/10.1001/jamaneurol.2024.2506 PMid:39133506
37. Graham CE, Lee WH, Wiggin HR, Supper VM, Leick MB, Birocchi F, Yee AJ, Petrichenko A, Everett J, Bushman FD, et al. Chemotherapy-induced reversal of ciltacabtagene autoleucel-associated movement and neurocognitive toxicity. Blood 2023; 142: 1248-1252. https://doi.org/10.1182/blood.2023021429 PMid:37467494 PMCid:PMC10579042
38. Marella M, Yao X, Carreira V, Bustamante M,Clark B, Jackson C, Zudaire E, Schecter J, Glover T, Shenton J, et al. Comprehensive BCMA expression profiling in adult normal human brain suggests a low risk of on-target neurotoxicity in BCMA-taregting multiple myeloma therapy. J Histochem Cytochem 2022 70. 273-287.

https://doi.org/10.1369/00221554221079579
PMid:35193424 PMCid:PMC8971684
39. Hines MR, Knight TE, McNerney KO, Leick MB, Jain T, Ahmed S, Frigault MJ, Hill JA, Jain MD, Johnson WT, et al. Immune effector cell-associated hemphagocytic lymphohitiocytosis-like syndrome. Transplant Cell Ther 2023; 29: 438.e1-438.e.16.
https://doi.org/10.1016/j.jtct.2023.03.006
PMid:36906275 PMCid:PMC10330221
40. Sandler RD, Tattersall RS, Schoemans H, Greco R, Badoglio M, Labopin M, Alexander T, Kirgirou K, Rovira M, Saif M, et al. Diagnosis and management of secondary HLH/MAS following HSCT and CAR-T cell therapy in adults; a review of the literature and a survey of practice within EMBT centres on behalf of the autoimmune diseases working party (TCWP). Front Immunol 2020; 11: 524.
https://doi.org/10.3389/fimmu.2020.00524
PMid:32296434 PMCid:PMC7137396
41. Hines MR, Knight TE, McNerney KO, Leick MB, Jain T, Ahmed S, Frigault MJ, Hill JA, Jain MD, Johnson WT, Lin Y, Mahadeo KM, Maron GM, Marsh RA, Neelapu SS, Nikiforow S, Ombrello AK, Shah NN, Talleur AC, Turicek D, Vatsayan A, Wong SW, Maus MV, Komanduri KV, Berliner N, Henter JI, Perales MA, Frey NV, Teachey DT, Frank MJ, Shah NN. Immune Effector Cell-Associated Hemophagocytic Lymphohistiocytosis-Like Syndrome. Transplant Cell Ther. 2023 Jul;29(7):438.e1-438.e16.
https://doi.org/10.1016/j.jtct.2023.03.006
PMid:36906275 PMCid:PMC10330221
42. Lichtenstein DA, Schischilk F, SAteinber SM, Yates B, Wang HW, Inglefield J, Dulan-Florea A, Ceppi F, Hermida LC, et al. Characterization of HLH-like manifestations of a variant in patients receiving CD22 CAR-T cells. Blood 2021; 138: 2469-2484.
https://doi.org/10.1182/blood.2021011898
PMid:34525183 PMCid:PMC8832442
43. Rocco JM, Inglefield J, Yates B, Lichtenstein DA, Wang Y, Goffin L; Filipovic D, Schiffrin EJ, Shah NN. Free interleukin-18 is elevated in CD22 CAR T-cell-associated hemophagocytic-like toxicities. Blood Adv 2023; 7: 6134-6139.
https://doi.org/10.1182/bloodadvances.2023010708
PMid:37389815 PMCid:PMC10582359
44. Schultz LM, Jeyakumar N, Kramer AM, Sahaf B, Sianagesh H, Shiraz P, Agarwal N, Hamilton M, Erickson C, Jacobs A, et al. CD22 CART cells demonstrate high response rates and safety in pediatric and adult B-ALL: phase 1b results. Leukemia 2024; 38: 963-968.
https://doi.org/10.1038/s41375-024-02220-y
PMid:38491306
45. Fauter M, Mainbourg S, Jammal TE, Guerber A, Zaepfel S, Henry T, Gerfaud-Valentin M, Sève P, Jamilloux Y. Extreme hyperferritinemia: causes and prognosis. J Clin Med 2022; 11, 5438.
https://doi.org/10.3390/jcm11185438
PMid:36143085 PMCid:PMC9505036
46. Bami S, Vagrecha A, Soberman D, Badawi M, Cannone D, Lipton JM, Cross R, Levy CF. The use of anakinra in the treatment of secondary hemophagocytic lymphohistiocytosis. Pediatr Blood Cancer 2020; 67: e28581.
https://doi.org/10.1002/pbc.28581
PMid:32725881
47. FDA Investigation serious risk of T-cell malignancy following BCMA-directed autologous chimeric antigen receptor (CAR) T cell immunotherapies. US Food & Drug Administration. Available online: https://www.fda.gov/vaccines-bloof-biologics/safety-availability-biologics/fda-investigating-serios-risk-t-cell-malignancy-following-bcma-directed-orr-cd19-directed-autologous
48. Verdun N, Marks P. Secondary cancers after chimeric antigen receptor T-cell therapy. N Engl J Med 2024; 390: 584-586.
https://doi.org/10.1056/NEJMp2400209
PMid:38265704
49. Levine BL, Pasquini MC, Connolly JE, Ported DL, Gustafson MP, Boelens EM, Grupp SA, Maus MV, Locke FL, et al. unanswered questions following reports of secondary malignancies after CAR-T cell therapy. Nat Med 2024; 30: 338-341.
https://doi.org/10.1038/s41591-023-02767-w
PMid:38195751 PMCid:PMC11688691
50. Elsallab M, Ellithi M, Lunning MA, D'Angelo C, Ma J, Perales MA, Frigault M, Maus MV. Second primary malignancies afetr commercial CAR T cell tehrapy: analysis of FDA adverse events reporting system (FAERS). Blood 2024; 143: 2099-2105.
https://doi.org/10.1182/blood.2024024166
PMid:38483155
51. Shen J; Hu R, Lin A, Jiang A, Tang B, Liu Z, Cheng Q, Miao K, Zhang J, Luo P. Characterization of second primary malignancies post CAR T-cell therapy: rael-world insights from the two global pharmacovigilance databases of FAERS and Vigibase. Lancet 2024; 73: 102684.
https://doi.org/10.1016/j.eclinm.2024.102684
PMid:39007060 PMCid:PMC11245995
52. Tix T, Alhoumoud M, Shouval R, Scheffer Cliff ER, Perales MA, Dordas dos Santos D, Rejeski K. Second primary nalignancies after CAR T-cell therapy: a systematic review and meta-analysis of 5,517 lymphomas and myeloma patients. Clin Cancer Res 2024; 30: 4690-4700.
https://doi.org/10.1158/1078-0432.CCR-24-1798
PMid:39256908 PMCid:PMC11546643
53. Ghilardi G, Fraietta JA, Gerson JN, Van Deerlin VM, Morrissette J, Caponetti JC, Paruzzo L, Harris JC, Chong EA, Susanibar Adaniya SP, et al. et al. T cell lymphoma and secondary primary malignancy risk after commercial CAR T cell therapy. Nat Med 2024; 30: 984-989.
https://doi.org/10.1038/s41591-024-02826-w
PMid:38266761
54. Hamilton MP, Sugio T, Noordenbos T, Shi S, Bulterys P, Liu CL, Kang X, Olsen MN, Good Z, Dahiya S, et al. Risk of second malignancies and T-cell lymphoma after chimeric antigen receptor T-cell therapy. N Egnl J Med 2024; 390: 2047-2060.
https://doi.org/10.1056/NEJMoa2401361
PMid:38865660 PMCid:PMC11338600
55. Chihara D, Dore GM, Flowers CR, Morton LM. The bidirectional increased risk of B-cell lymphoma and T-cell lymphoma. Blood 2021; 139: 785-789.
https://doi.org/10.1182/blood.2020010497
PMid:33822002 PMCid:PMC8414260
56. Goel H, Rahul E, Gupta I, Chopra A, Ranjan A, Gupta AK, Prasad Meene J, Viswanathan GK, Bakhshi S, Misra A, et al. Molecular and genomic landscapes in secondary & therapy related acute myeloid leukemia. Am J Blood Res 2021; 11: 472-497.
57. Demoor-Goldschmidt C, de Vathaire F. Review of risk factors of secondary cancers among cancer survivors. Br J Radiol 2019; 92: 20180390.
https://doi.org/10.1259/bjr.20180390
PMid:30102558 PMCid:PMC6435077
58. Palumbo A, Bringhen S, Kumar SK, Lupparelli G, Usmani S, Waage S, Larocca A, van der Holt B, Musto P, Offidani M, et al. Second primary malignancies with lenalidomide therapy malignancies with lenalidomide therapy for newly diagnosed myeloma: a meta-analysis of individual patient data. Lancet Oncol 2014; 15: 333-342.
https://doi.org/10.1016/S1470-2045(13)70609-0
PMid:24525202
59. Musto P, Anderson KC, Attal M, Richardon PG, Badros A, Hou J, Comenzo R, Du J, Durie BGM, San Miguerl J, et al. Second primary malignancies in multiple myeloma: an overview and IMWG consensus. Ann Oncol 2017; 28: 228-245.
https://doi.org/10.1093/annonc/mdw606
PMid:27864218
60. Galli E, Rossi M, Pansini I, Viscovo M, Malara T, Colangelo M, Alma E, Valentini CG, Teofili L, Hohaus S, Sica S, Sorà F, Chiusolo P. Predicting therapy-related myeloid neoplasms after CAR-T with the Clonal Haematopoiesis Risk Score (CHRS). Br J Haematol. 2025 Jan;206(1):372-374. doi: 10.1111/bjh.19905. Epub 2024 Nov 13. PMID: 39539012.
https://doi.org/10.1111/bjh.19905
PMid:39539012
61. Maude SL, Laetsch TW, Buechner J, Rives S, Boyer M, Battencourt H, Bader P, Verneris MR, Stefanski HE, Myers GD, et al. Tisagenlecleucel in children and young adults with B-cell lymphoblastic leukemia. N Engl J Med 2018; 378: 439-448.
https://doi.org/10.1056/NEJMoa1709866
PMid:29385370 PMCid:PMC5996391
62. Laetsch TW, Maude SL, Rives S, Hiramatsu H, Bittencourt H, Bader P, Baruchel A, Boyer M, De Moerloose B, Qayed M, et al. Three year update of Tisagenlecleucel in pediatric and young adult patients with relapsed/refractory acute lymphoblastic leukemia in the ELIANA trial. J Clin Oncol 2022; 41: 1664-1669.
https://doi.org/10.1200/JCO.22.00642
PMid:36399695 PMCid:PMC10022844
63. Baird JH, Epstein DJ, Tamaresis JS, Ehlinger Z, Spiegel JY, Craig J. Immune reconstitution and infection complications following axicabtagene ciloleucel tehrapy for large B-cell lymphoma. Blood Adv 2021; 5: 143-155.
https://doi.org/10.1182/bloodadvances.2020002732
PMid:33570626 PMCid:PMC7805341
64. Sutherland NM, Zhou B, Zhang L. On M.S. Hong J.S, Pak A, Liu K.J,

Frigault M.J, Maus M.V, Hill J.A, et al. Association of CD19+-targeted chimeric antigen receptor (CAR) T-cell therapy with hypogammaglobulinemia, infection and mortality. J Alelrg Cel Immunol 2024; in press.
https://doi.org/10.1016/j.jaci.2024.10.021
PMid:39505278

65. Malenhorst JJ, Chen GM, Wang M, Porter DL, Chen C, Collins MA, Gao P, Bandyopadhyay S, Sun H, Zhao Z, Lundh S, et al. Decade-long leukemia remissions with persistence of CD4+ CAR T cells. Nature 2022; 602: 503-509.
https://doi.org/10.1038/s41586-021-04390-6
PMid:35110735 PMCid:PMC9166916
66. Johnson G, Patel K, Arciola B, Perez AP, Wood A, Ionescu F, Logue JM, Albanyan O, Yasin H, Bachmeier CA, et al. Poor hematopoiesis reserve at the time of CD19 CAR T-cell infusion is associated with long-term B-cell aplasia. Blood 2023; 142 (suppl.1): 3512.
https://doi.org/10.1182/blood-2023-185097
67. Kampouri E, Little JS, Rejeski K, Manuel O, Hammond SP, Hill JA. Infections after chimeric antigen receptor (CAR) T-cell therapy for hematologic malignancies. Transpl Infect Dis 2024; 25(suppl.1): e14157.
https://doi.org/10.1111/tid.14157
PMid:37787373
68. Rejeski K, Subklewe M, Aljurf M, Bachy E, Balduzzi A, Barbal P. Immune effector cell-associated hematotoxicity EHA/EBMT consensus grading and best practice recommendations. Blood 2023; 142: 865-877.
https://doi.org/10.1182/blood.2023020578
PMid:37300386
69. Yang Y, Peng H, Wang J, Li F. New insights into CART-cell hematological toxicities: manifestations, mechanisms, and effective management strategies. Exp Hematol Oncol 2024; 13: 110.
https://doi.org/10.1186/s40164-024-00573-9
PMid:39521987 PMCid:PMC11549815
70. Rejesky K, Perez A, Sesques P, Hoster E, Bergi C, Jentzch L. CAR-HEMATOTOX: a model for CAR T-cell-related hematologic toxicity in relapsed/refractory large B-cell lymphoma. Blood 2021; 138: 2499-2513.
https://doi.org/10.1182/blood.2020010543
PMid:34166502 PMCid:PMC8893508
71. Nair MS, Silbert SK, Rejeski K, Wilson KA, Lamble AJ, Valtis YK, Yates B, Arana AM, Shouval R, Curran KJ, et al. Development of ALL-Hematotox (ALL-HT): predicting post CAR T-cell hematotoxicity in B-cell acute lymphoblastic leukemia. Blood 2024; in press.
https://doi.org/10.1182/blood.2024025910
PMid:39561284 PMCid:PMC11923441
72. Strati P, Li X, Deng Q, Marques-Piubelli M, Henderson J, Watson G, Deaton L, Cain T, Yang H, Ravanmehr V, Fayad L, et al. Prolonged cytopenia following CD19 CAR T cell therapy is linked with bone marrow infiltration of clonally expanded IFNγ-expressing CD8 T cells. Cell Rep Med 2023; 4, 101158.
https://doi.org/10.1016/j.xcrm.2023.101158
PMid:37586321 PMCid:PMC10439270
73. Drillet G, Lhomme F, De Guibert S, Manson G, Houot R. Prolonged thrombocytopenia after CAR T-cell therapy: the role of thrombopoietin receptor agonniststst. Blood Adv 2022; 7: 537-540.
https://doi.org/10.1182/bloodadvances.2022008066
PMid:35930692 PMCid:PMC9979763
74. Liévin R, Di Blasi R, Morin F, Galli E, Allain V, De Jorna R, Vercellino L, Parquet N, Mebarki M, Larghero J, de Kerviler E, Madelaine I, Caillat-Zucman S, Chevret S, Thieblemont C. Effect of early granulocyte-colony-stimulating factor administration in the prevention of febrile neutropenia and impact on toxicity and efficacy of anti-CD19 CAR-T in patients with relapsed/refractory B-cell lymphoma. Bone Marrow Transplant. 2022 Mar;57(3):431-439. doi: 10.1038/s41409-021-01526-0. Epub 2022 Jan 30. PMID: 35094012; PMCID: PMC8907072.
https://doi.org/10.1038/s41409-021-01526-0
PMid:35094012 PMCid:PMC8907072
75. Rejeski K, Subklewe M, Aljurf M, Bachy E, Balduzzi A, Barba P, Bruno B, Benjamin R, Carrabba MG, Chabannon C, Ciceri F, Corradini P, Delgado J, Di Blasi R, Greco R, Houot R, Iacoboni G, Jäger U, Kersten MJ, Mielke S, Nagler A, Onida F, Peric Z, Roddie C, Ruggeri A, Sánchez-Guijo F, Sánchez-Ortega I, Schneidawind D, Schubert ML, Snowden JA, Thieblemont C, Topp M, Zinzani PL, Gribben JG, Bonini C, Sureda A, Yakoub-Agha I. Immune effector cell-associated hematotoxicity: EHA/EBMT consensus grading and best practice recommendations. Blood. 2023 Sep 7;142(10):865-877. doi: 10.1182/blood.2023020578. PMID: 37300386.
https://doi.org/10.1182/blood.2023020578
PMid:37300386
76. Roddie C, Dias J, O'Reilly MA, Abassian M, Cadinanos-Garai A, Vispute K, Bosshard-Carter L, Mitsikakou M, Mehra V, Roddy H, et al. Durable responses and low toxicity after off-rate CD19 chimeric antigen receptor-T therapy in adults with relapsed or refractory B-cell acute lymphoblastic leukemia. J Clin Oncol 2021; 39: 3352-3363.
https://doi.org/10.1200/JCO.21.00917
PMid:34464155 PMCid:PMC8791810
77. Jabbour E, Tholouli E, Sandhu K, Shaughnessy P, Barba P, Guerriero M, Yallop D, Logan A, Abedi M, Sirieix PL, et al. Obecabtagene autoleucel (oce-cel, AUTO1) in adults with relapsed/refractory B-cell acute lymphoblastic leukemia (R/R B-ALL): overall survival (OS), event-free survival (EFS) and the potential impact of chimeric antigen receptor (CAR)-T cell persistency and consolidative stem cell transplantation (SCT) in the open-label, single-arm FELIX phase Ib/II study. J Clin Oncol 2024; 42(suppl.16): 6504.
https://doi.org/10.1200/JCO.2024.42.16_suppl.6504
78. Brudno JN, Lam N, Vanasse D, Shen Y, Rose JJ, Rossi J, Xue A, Bot A, Scholler N, Mikkilineni L, et al. Safety and feasibility of anti-CD19 CART cells with fully human binding domains in patients with B-cell lymphoma. Nat Med 2020; 26: 270-280.
https://doi.org/10.1038/s41591-019-0737-3
PMid:31959992 PMCid:PMC7781235
79. Weinkove R, George P, Fyfe R, Dasyam N, Nouri Y, Ostapowicz T, Mullins S, Mester B, Giunti G, Bollard CM, et al. A phase I dose escalation trial at third-generation CD19-directed CAR T-cells incorporating CD28 and Toll-like receptor 2 (TLR2) intracellular domains for relapsed or refractory B-cell non-Hoggkin lymphomas (ENABLE). Blood 2023; 142(suppl.1): 890.
https://doi.org/10.1182/blood-2023-178872
80. Park JH, Palomba ML, Devlin SM, Valtis YK, Sikder D, Senechal B, Wang X, Cathchart E, Liotta K, Yu A, et al. A phase I study of CD19-targeted 19(T2)28z1xx CAR T cells in adult patients with relapsed or refractory large B-cell lymphoma. Blood 2023; 142(suppl.1): 892-893.
https://doi.org/10.1182/blood-2023-187623
81. Xue L, Yi Y, Xu Q, Wang L, Yang X, Zhang Y, Hua X, Chai X, Yang J, Chen Y, et al. Chimeric antigen receptor T cells self-neutralizing IL6 storm in patients with hematologic malignancy. Cell Discov 2021; 7: 84.
https://doi.org/10.1038/s41421-021-00299-6
PMid:34518515 PMCid:PMC8437938
82. Hu B, Yi Y, Wang X, Xue L, Tao G. A potential best-in-class BCMA-CD19 bispecific CART with advanced safety by self-inhibiting IFNG signaling during CRS. J Clin Oncol 2014; 42 (suppl.16): 2502.
https://doi.org/10.1200/JCO.2024.42.16_suppl.2502
83. Yoshikawa T, Ito Y, Wu Z, Kasuya H, Nakashima T, Okamoto S, Amaishi Y, Zhang H, Li Y, Matsukawa T, et al. Development of a chimeric cytokione receptor that captures IL-6 and enhances the antitumoir response of CAR-T cells. Cell Rep Med 2024; 5: 101526.
https://doi.org/10.1016/j.xcrm.2024.101526
PMid:38670095 PMCid:PMC11148643
84. Lin MY, Nam E, Shih RM, Shafer A, Bouren A, Ceja MA, Harris C, Chericha M, Vo KH, Kim M, et al. Self-regulating CAR-T cells modulate cytokine release syndrome in adoptive T-cell therapy. J Exp Med 2024; 221: in press.
https://doi.org/10.1084/jem.20221988
PMid:38607370 PMCid:PMC11010356
85. Aldoss I, Khaled SK, Wang X, Palmer J, Wang Y, Wagner JR, Clark MC, Simpson J, Paul J, Vyas V, Chien SH, et al. Favorable activity and safety profile of memory-enriched CD19-targeted chimeric antigen receptor T-cell therapy in adults with high-risk relapse/refractory ALL. Clin Cancer Res 2023; 29: 742-753.
https://doi.org/10.1158/1078-0432.CCR-22-2038
PMid:36255386 PMCid:PMC10544259