Anticuerpos Biespecíficos Monoclonales

Bispecific Antibody Therapy for Hematologic Conditions: A Review of Current Therapies and Emerging Treatments.

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Inmunoterapia en Oncohematología: conquistas y desafíos

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Introduction

Cancer immunotherapy utilizes the specificity, diversity, and memory of the human immune system to recognize and remove malignant cells. There are now many different types of immune therapies available or under development including monoclonal antibodies, PD-1/PD-L1 inhibitors, chimeric antigen T cells and TCRs, and bispecific antibodies. Each of these therapies takes a different approach towards redirecting the immune system to target malignant cells or other disease processes.

Antibodies are one of the primary means of recognizing and recruiting the immune system to attack and remove foreign substances from the body. It has been recognized for many years that if the two antigen-binding domains on an antibody could be altered to recognize separate antigens that that this could be used for therapeutic benefit. While this concept has been around for decades, it has taken time to develop, synthesize, and produce therapeutic quantities of antibodies for clinical development and use.[1]

Prior to the development of bispecific antibodies, monoclonal antibodies (mAb) were the first therapeutic antibodies developed with Rituxan approved in 1997 for the treatment of non-Hodgkin's lymphoma. Other monoclonal antibodies have since been developed for both solid and hematologic malignancies; however, they have had overall limited activity in treating cancer. Antibody drug conjugates (ADC) contain a monoclonal antibody connected by a linker sequence to a chemotherapy molecule and use agents that are too toxic to give systemically but have potent activity when administered in a targeted fashion. The first approved ADC was gemtuzumab ozogamicin (GO). It is a humanized anti-CD33 mAb connected to a calicheamicin derivative and

was granted accelerated approval for treatment of AML, but the drug was withdrawn from the market in 2010 after the Phase III trial SO106 showed increased mortality and no improvement in response rates or survival when added to standard induction chemotherapy.[2] Recent studies have shown that certain subsets of patients may benefit from GO such as those with favorable risk cytogenetics, and APL.[3]
Brentuximab vedotin is an anti-CD30/MMAE ADC approved for refractory Hodgkin Lymphoma and systemic anaplastic large cell lymphoma. Limitations of antibody drug conjugates include requirements for internalization of the antibody and drug, need for adequate expression of the target antigen, and linker technology able to release the toxin in a controlled fashion. Nevertheless, this is an exciting new field, with many new drugs in development.[4]
Bispecific Antibodies are synthesized protein structures with two distinct antigen-binding sites that are being developed for various clinical applications including management of malignancy, inflammation, and coagulation. In cancer immunotherapy, these molecules are used to force an interaction between cytotoxic immune cells and the desired tumor cell. There are many different bispecific structures that have been developed, and most of these are proprietary. They all share the common feature of combining two or more different antigen-binding domains onto the same molecule.
There are two main categories of bispecific structures. The first group approximates a full IgG-like antibody structure with an Fc region. Maintaining the Fc region incorporates functions such as antibody-dependent cellular cytotoxicity and complement fixation, and these structures have extended serum half-lives. Another type of compound consists of much smaller structures that contains minimized antigen binding domains in an antibody like structure that eliminates the Fc-like portion. When the Fc portion is not included, the structures have much shorter half-lives and may have increased toxicity as a result of their small size.[5] Most bispecific antibodies in development do not contain Fc regions and are smaller structured compounds.
Bispecific antibodies are currently in development for many different malignancies including hematologic and solid tumors. The sections below will discuss the recently approved BiTE, blinatumomab, and also discuss a few bispecifics currently in pre-clinical and clinical development for hematologic conditions.

BiTE Therapy and Blinatumomab
Bispecific T Cell Engaging Antibodies, or BiTE Antibodies, are a type of bispecific antibody composed of a single polypeptide chain that contains only the minimal variable binding regions for two different antigens connected by non-immunogenic linker sequences to form a single-chain antibody (scFv).[6]
These molecules are approximately 55kDa and much smaller than a typical antibody. BiTEs bind with one domain to CD3 epsilon, which is a subunit of the T cell receptor complex present on all T cells, and to the cell surface of target cells with the other binding domain.[7] BiTEs bind to CD3 with low affinity and only trigger cytolytic T-cell responses when simultaneously bound to their clinical antigen, and are active in the pg/mL range.[8] They recruit polyclonal T-cell responses for serial rounds of target cell lysis while avoiding toxicity from non-specific activation of T-cells[9]. By interacting independently of MHC and antigen presentation, BiTEs bypass common tumor immune escape mechanisms.[10]
Blinatumomab is a BiTE construct that links CD3 with CD19 and was approved in December 2014 by the US Food and Drug Administration for treatment of Philadelphia chromosome negative relapsed or refractory acute lymphoblastic lymphoma under an accelerated process using data from a Phase II study. In this multi-center study of 189 patients, 81 (43%) achieved a complete remission (CR) or complete remission with partial hematologic recovery (CRh). 64 patients in the study had had a previous allogeneic stem cell transplant, and 29 (45%) of these prior transplant patients had a CR or CRh. 32 patients with CR or CRh (40%) went on to allogeneic stem cell transplant in the study. Median relapse free survival for responders was 5.9 months and median overall survival was 6.1 months in this high risk and heavily treated population.[11] Neurotoxicity, occurring in 58% of patients, and cytopenias were the most common adverse events. Grade III cytokine release syndrome occurred in only 3 patients, but patients received pretreatment with dexamethasone. A phase III confirmatory study in ALL is pending completion. Blinatumomab has been evaluated in Non-Hodgkin's lymphoma, MRD-positive B precursor ALL, pediatric ALL, and a phase II study in Diffuse Large B Cell Lymphoma is ongoing.[12,13]
While Blinatumomab shows promising response rates and overall safety, administration is cumbersome and requires close evaluation and follow-up.

The drug has a short half-life of 2 hours and requires 24-hour continuous infusion for 4 weeks of a 6-week cycle to provide therapeutic levels. Hospitalization is recommended for the first nine days of cycle 1 and also the first 2 days of cycle 2 to monitor for signs of cytokine release syndrome and other toxicity.[14] Home administration requires the patient to wear a continuous infusion pump and for adequate pharmacy and home health support to change drug supplies and manage the infusion. This requires specialized care as well as significant cost.

Neurotoxicity and Cytokine Release Syndrome

Significant toxicities associated with immunotherapy include neurotoxicity and the cytokine release syndrome (CRS). These have been well documented with blinatumomab and will presumably occur with other bispecific antibodies that recruit T cell responses. The neurotoxicity observed with blinatumomab ranges from mild symptoms such as tremor and dizziness to severe events with seizures and encephalopathy.[15] Neurotoxicity may be due to the small size of the BiTEs and ability to cross the blood-brain-barrier or to release of excess cytokines.[16] Neurotoxicity resolves when the drug is withheld. In clinical trials, some patients tolerated lower doses of Blinatumomab when re-challenged after developing neurotoxicity.[17]

Cytokine Release syndrome (CRS) is an emerging condition associated with cancer immunotherapy that arises from extreme immune system activation and elevated levels of cytokines. Cytokines associated with CRS include elevated levels of IL-6, IL-10, INF-γ, and TNFα.[18] Presentations of CRS include high fever, hypotension, and multi-organ dysfunction including ARDs, cardiotoxicity, acute kidney injury requiring hemodialysis, transaminitis, DIC, and neurotoxicity. With Blinatumomab, CRS has been associated with initial infusions and rapid destruction of CD19 positive cells. Dexamethasone premedication, drug dose escalations, and cytoreduction with cyclophosphamide help to attenuate the development and effects of CRS, but may also counteract effects of therapy.[19] Given the pattern of cytokine elevation and clinical presentation, there may be overlap with hemophagocytic lymphohistiocytosis, and macrophage activation may play a role in CRS.[20] IL-6 is thought to be the key mediating cytokine, but serum IL-6 levels are not predictive of CRS. Diagnosis of CRS can be difficult to distinguish from infection in neutropenic patients, and treatment can also be complicated by tumor lysis syndrome. Treatment of CRS is complicated because reversal of cytokine release with immunosuppression such as steroids neutralizes the effects of immunotherapy. Tocilizumab is a humanized IgG anti IL-6R monoclonal antibody that was approved for use in rheumatologic conditions that has been effective at treating CRS, and may not attenuate the effects of the immunotherapy. Tocilizumab does not cross the blood brain barrier and is not as effective for neurotoxicity. Steroids reverse the symptoms of CRS, and do cross the blood brain barrier and may be needed to treat neurotoxicity.[21]

CD3xCD33 BiTE for treatment of AML

AMG 330 is a CD3 xCD33 BiTE currently under development for treatment of acute myeloid leukemia. The antigen CD33 is found on 85-90% of AML blasts and appears to be a suitable target for AML directed immunotherapy.[22] Pre-clinical studies suggest that AMG330 is active at low concentrations as well as low CD33 density on AML cells. Activity is not affected by ABC transporter protein expression, and treatment with AMG330 does not lead to decreased expression of CD33 over time. These results suggest that the bispecific approach of AMG330 may avoid some of the obstacles encountered with lintuzumab, a CD33 monoclonal antibody, and gemtuzumab ozogamicin, a CD33 antibody-drug conjugate.[23] A concern with anti-CD33 therapy is that normal stem cells and hematopoietic progenitor cells express CD33 and may be affected by therapy. Significant cytopenias will be an expected side effect.[24] It is not clear that targeting CD33 will completely eliminate the source of AML clones, but any advance in treatment would be progress for a disease that has seen little change in management in 40 years and still carries a dismal prognosis for most adult patients. Clinical trials with AMG330 have not yet begun.

CD3xCD123 DART for treatment of AML

Dual-affinity re-targeting (DART) bispecifics are another form of bispecific therapy. DARTs consist of two protein chains with the VH of the first variable region linked to the VL of the second variable region, and the VH of the second variable region linked to the VL of the first variable region. Covalent disulfide linkages stabilize the two protein chains.[25] MGD006 is a bispecific CD3xCD123 DART that re-directs T lymphocytes to CD123. CD123 is the interleukin 3 receptor alpha chain and is overexpressed in AML and other hematologic malignancies including hairy cell leukemia and some B cell malignancies.[26] CD123 is not expressed on normal

hematopoietic stem cells and MGD006 will hopefully target primarily malignant cells.[27] Preclinical data for MGD006 has been promising.[28] MGD006 is currently in phase I trials for relapsed/refractory AML (NCT02152956).

CD16AxCD30 and CD19xCD3 TandAb

AFM13 is a bispecific, tetravalent chimeric antibody construct (TandAb) that contains a binding site for the isoform CD16A, which recruits NK Cells, and for CD30 on Hodgkin Lymphoma cells. The TandAb antibody is formed from a single polypeptide that contains 2 binding domains for each antigen that homodimerizes and is held together by non-covalent interactions between the immunoglobulin heavy and light variable chains. Phase I study results published in June 2015 reported that in 28 heavily pretreated patients with relapsed/refractory Hodgkin's lymphoma that 3 of 26 patients had a partial remission and 13 patients had stable disease with overall disease control rate of 61.5%. These results are promising and AFM13 is currently in phase II studies.[29] AFM11 is a CD19xCD3 TandAb currently in Phase 1 trials for patients with relapsed/refractory CD19 positive malignancies.[30]

Anti-Factor IXa/X Forced Association for treatment of Hemophilia

In a departure from the use of bispecific constructs to re-direct cytolytic components of immune system to destroy target cells, ACE910 is a humanized IgG bispecific antibody that binds to Factor IXa and Factor X to mimic the function of Factor VIII in the coagulation cascade and is under development for treatment of hemophilia A.[31] ACE910 has been evaluated in primates and has been shown to have hemostatic activity and to prevent spontaneous bleeding. ACE910 does not generate Factor VIII inhibitors, functions in the presence of inhibitors, and has a half-life of 3-weeks.[32] Current treatment for hemophilia A involves frequent injections of recombinant Factor VIII, which has a short half life, and the development of Factor VIII inhibitors is a common complication. A treatment that provides hemostatic control with fewer injections and without the risk of inhibitor development would be an important advance for the treatment of Hemophilia A. ACE910 is currently in phase1/2 studies.

Limitations

While many of these drugs show clinical promise they do have limitations. The bispecifics that lack an Fc like portion have very short serum half-lives and require continuous infusion and CRS type syndrome and neurotoxicity will likely complicate most immune directed therapies. While the early data for blinatumomab was very promising with high response rates, in the Phase II study, response rates were lower and rather short lived. It is likely that other drugs with high response rates in phase I/II trials may have lower response rates when taken to larger trials. With disease relapsing after treatment with targeted immunotherapy, it is likely that the immunotherapies are not targeting the malignant cells of origin, and it does not appear that these drugs are yet able replace the role of allogeneic stem cell transplant.

In addition to bispecific therapies, chimeric antigen T cells and T cell receptors are also under development. Bispecific antibodies have the benefit of a short and defined half-life, and appear to have less toxicity than CAR-T cells. CAR-T cells have the potential to induce longer lasting responses, but carry a risk of severe cytokine release syndrome and autoimmune toxicity. Unlike with bispecifics, CAR-T cell infusions cannot merely be turned off. TCRs are another interesting class of therapies that have less off target immune effects, but rely on MHC presentation. It is unclear what role each will play in therapy, but given the complexity of hematologic malignancies and patient diversity, there will likely be applications for each of these different approaches.

With immunotherapy, there are now drugs in development that hold the hope of potentially transforming the treatment landscape for hematologic malignancies. Bispecific antibodies as a treatment class are still in the early stages of development but have shown that they can have strong clinical responses with acceptable toxicity profiles and many varied applications. With time and improved technology, some of the limitations such as shortened half-life, administration, and toxicity will improve. It will be interesting to follow the development of this new class of therapeutics.

Declaration of conflict of interest:
I have a small amount of stock and I derive research support from Amgen.

Table 1: Bispecific Antibodies in Development for Hematologic Conditions

Drug Name	Class	Target	Condition	Clinical Status
Blinatumomab (Amgen)	BiTE	CD3xCD19	relapsed/refractory ALL, relapsed/refractory pH+ALL DLBCL	Approved/Phase III Phase II Phase II
AMG330	BiTE	CD3xCD33	AML	n/a
MGD006 (Macrogenics/Servier)	DART	CD3xCD123	AML	Phase I
AFM11 (Affimed)	TandAb	CD3XCD19	Non-Hodgkin's Lymphoma	Phase I
AFM13	TandAb	CD16AxCD30	Hodgkin's Lymphoma	Phase I/II
ACE910		Factor IXa/X	Hemophilia A	Phase I/II

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