Thursday, June 05 19:00-21:00 (Marquee Parco Ciani)

Genentech BioOncology

On the trail to efficient cell death in b-cell malignancies

Chair: D.G. Maloney (Seattle, USA)

Anti-cd20 antibodies with increased antigen binding affinity and enhanced effector function

H. Lowman (South San Francisco, USA)

Phenotypic and functional characterization of B-cell specific antigens: does targeting them modulate disease?

D.G. Maloney (Seattle, USA)

Targeting trail death receptors in lymphoma

A. Younes (Houston, USA)

Immunotherapy of lymphoma

R. Levy (Palo Alto, USA)

Angiogenic mediators support growth and metastasis in myeloma and lymphoma

J.P. Leonard (New York, USA)

ANTI-CD20 ANTIBODIES WITH INCREASED ANTIGEN BINDING AFFINITY AND ENHANCED EFFECTOR FUNCTION

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We have humanized an antibody binding to an overlapping but distinct epitope on CD20 as compared with rituximab to generate a second generation antibody, PRO70769. This antibody binds with similar affinity as rituximab to human CD20, but shows enhanced ADCC and decreased CDC activity. Alanine-scanning mutagenesis of the variable domain of this antibody resulted in variants with enhanced CD20 binding affinity and CDC activity. Further modifications of the Fc region led to further increases in CDC activity as well as FcgRIII binding affinity and NK-cell dependent ADCC. Third generation anti-CD20 antibodies such as PRO131921 and PRO145097 provide an opportunity for studying the roles of these mechanisms of B-cell depletion in vivo. We used hCD20/ hCD16 transgenic mice to compare normal B-cell depletion in the context of low-affinity human CD16, also known as FcgRIII(F158). The results of these studies show improved potency for B-cell depletion in certain compartments, such as the peritoneal cavity, with engineered anti-CD20s.

PHENOTYPIC AND FUNCTIONAL CHARACTERIZATION OF B-CELL SPECIFIC ANTIGENS: DOES TARGETING THEM MODULATE DISEASE?

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Targeting the B-cell-specific cell-surface protein CD20 with the chimeric monoclonal antibody rituximab has radically changed the treatment for most patients with CD20-positive non-Hodgkin's lymphoma (NHL) or chronic lymphocytic leukemia (CLL). This has led to a renewed interest in the development of next generation anti-CD20 antibodies and for the development of new antibodies targeting a variety of cell-surface proteins. Characteristics of the target antigen are critical in determining the success of antibody-based treatments. The CD20 antigen is B-cell restricted, appears to be present on all of the tumor cells, does not rapidly modulate, and is not secreted or shed from the cell surface at a rapid rate. Thus, the antibody is able to bind to the tumor cell surface and is likely to remain at the cell surface for prolonged periods, allowing for immune-mediated cell killing through antibody-dependent cellular cytotoxicity (ADCC) and the complement pathways. Additional antibody effects can occur through direct signaling through the target antigen upon antibody binding. Ideally, a target antigen would have a critical biologic function that could be modulated by antibody binding. Most anti-CD20 antibodies induce the accumulation of antigen into lipid rafts, while other anti-CD20 antibodies induce homotypic aggregation of cells and a greater degree of growth arrest/apoptosis. Lastly, to limit the emergence of antigen-negative tumor cells, the antigen should represent a stable target, with low rates of mutation and sequence variation and uniform expression. This may be more likely when the antigen has a critical biologic function. Despite the excellent results obtained by targeting CD20, antigen loss has been observed, sequence mutations have been identified, and cells have innate or can develop resistance to rituximab-binding. The use of radiolabeled anti-CD20 antibodies may be one way to overcome antigen heterogeneity and to decrease antigen-negative escape.

A large number of ''next generation'' anti-CD20 antibodies are now in clinical trials, promising to demonstrate greater clinical activity than rituximab. Most of these antibodies are humanized rather than chimeric, which may decrease the development of immune responses to the antibody. In addition, most have augmented interaction with ADCC effector cells through binding to the FCgammaRIIIA receptor, even in patients with low-affinity polymorphisms. Some have augmented or decreased interaction with complement, and some appear to cause greater direct effects (in vitro) upon antibody binding. Early clinical trials are beginning to report clinical activity, largely in patients who are not rituximab-refractory. The final results from these trials are eagerly awaited.

Other B-cell surface proteins are also being targeted, both with naked monoclonal antibodies and with radiolabeled or drug/toxin conjugated antibodies. Results targeting CD19, CD22, CD23, CD40, and death receptors appear to be encouraging. Hopefully these new agents will continue to improve outcome for our patients with B-cell malignancies.

TARGETING TRAIL DEATH RECEPTORS IN LYMPHOMA

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TRAIL (Apo2 ligand) is a death protein that is predominantly expressed by activated T cells and natural killer cells. TRAIIL has four exclusive receptors: TRAIL-R1 (DR4), -R2 (DR5, KILLER, TRICK2), -R3 (DcR1, TRID, LIT), and -R4 (DcR2 TRUNDD).[1-3] TRAIL also binds to OPG, although with a lower affinity.[4] TRAIL-R1 and -R2 are death receptors that are preferentially expressed by cancer cells, whereas TRAIL-R3 and -R4

are decoy receptors.[5] Thus, TRAIL preferentially kills cancer cells as they express TRAIL-R1 and TRAIL-R2. TRAIL has demonstrated antitumor activity against most human cancer cell lines, including lymphoma.[6] Both TRAIL/Apo2L and anti TRAIL-R1 and TRAIL-R2 agonistic antibodies can induce cell death in cancer cells by recruiting caspase 8 and 10 and activating the extrinsic apoptotic pathways.[7] Several preclinical experiments demonstrated that activating TRAIL death receptors in lymphoma may have a potential therapeutic value.[7] Preliminary data from a phase 2 study usingthe therapeutic monoclonal antibody mapatumumab targeting TRAIL-R1 demonstrated excellent safety but with modest clinical activity in NHL.[8] Three patients (8%) with relapsed follicular lymphoma had a response to therapy, with one complete remission and two partial responses after a maximum of 6 cycles, with over 30% having stable disease. A phase 1 study using Apo2L/TRAIL recombinant protein also demonstrated an excellent safety profile. Based on preclinical experiments demonstrating synergy between TRAIL pathway and rituximab against B-cell lymphoma,[9,10] a randomized study is currently being conducted comparing TRAIL/Apo2L with rituximab and with the combination of rituximab plus Apo2L/TRAIL.[11]Results from this ongoing study will provide valuable information on the clinical value of this approach in patients with relapsed follicular lymphoma.

Selected References:

1. Younes A, Aggarwall BB. Clinical implications of the tumor necrosis factor family in benign and malignant hematologic disorders. Cancer. 2003;98:458-467.
2. Ashkenazi A. Targeting death and decoy receptors of the tumour-necrosis factor superfamily. Nat Rev Cancer. 2002;2:420-430.
3. Bhardwaj A, Aggarwal BB. Receptor-mediated choreography of life and death. J Clin Immunol. 2003;23:317-332.
4. Emery JG, McDonnell P, Burke MB, et al. Osteoprotegerin is a receptor for the cytotoxic ligand TRAIL. J Biol Chem. 1998;273:14363-14367.
5. Degli-Esposti M. To die or not to die–the quest of the TRAIL receptors. J Leukoc Biol. 1999;65:535-542.
6. Georgakis GV, Li Y, Humphreys R, et al. Activity of selective agonostic monoclonal antibodies to TRAIL death receptors R1 and R2 in primary and cultured tumor cells of hematological origin. Blood. 2003;102:228a (abstr 799).
7. Georgakis GV, Li Y, Humphreys R, et al. Activity of selective fully human agonistic antibodies to the TRAIL death receptors TRAIL-R1 and TRAIL-R2 in primary and cultured lymphoma cells: induction of apoptosis and enhancement of doxorubicin- and bortezomib-induced cell death. Br J Haematol. 2005;130:501-510.
8. Younes A, Vose JM, Zelenetz AD, et al. Results of a phase 2 trial of HGS-ETR1 (agonistic human monoclonal antibody to TRAIL receptor 1) in subjects with relapsed/refractory non-Hodgkin's lymphoma (NHL). Blood. 2005;106:146a (abstr 489).
9. Daniel D, Yang B, Lawrence DA, et al. Cooperation of the proapoptotic receptor agonist rhApo2L/TRAIL with the CD20 antibody rituximab against non-Hodgkin lymphoma xenografts. Blood. 2007;110:4037-4046.
10. Maddipatla S, Hernandez-Ilizaliturri FJ, Knight J, Czuczman MS. Augmented antitumor activity against B-cell lymphoma by a combination of monoclonal antibodies targeting TRAIL-R1 and CD20. Clin Cancer Res. 2007;13:4556-4564.
11. Yee L, Fanale M, Dimick K, et al. A phase IB safety and pharmacokinetic (PK) study of recombinant human Apo2L/TRAIL in combination with rituximab in patients with low-grade non-Hodgkin lymphoma. J Clin Oncol. 2007;25(18S):460s (abstr 8078).

IMMUNOTHERAPY OF LYMPHOMA

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Molecular Defined Target

Since B-cell lymphomas are the clonal proliferation of lymphocytes, the immunoglobulin they express is unique, and it can be regarded as a tumor-specific antigen. The first monoclonal antibodies used in human lymphoma were directed against this target and were successful.

Three phase 3 clinical trials have now been conducted (Biovest, Genitope, and Favrille). Two trials have finished accrual, and one has just announced results (Genitope). The Genitope trialshowed no difference in the progression-free survival between the arm vaccinated with idiotype (Id)-KLH plus GM-CSF vs the control arm, which was vaccinated with KLH plus GM-CSF. However, there was a highly significant difference among the patients on the Id-specific vaccine arm who mounted a positive immune response compared to those who did not make an immune response. This result confirms the observations made in past phase 2 trials and leads to the conclusion that the ability to make an anti-idiotype immune response is some kind of biomarker for clinical outcome in follicular lymphoma (FL). It now becomes important to identify this subset of patients in other ways so that the biology of the disease can be better understood.

Forced Presentation of Tumor Antigens

We have now described a new strategy of therapeutic vaccination against lymphoma that combines tumor cell death induction with local injection of CpG oligonucleotides, a ligand for the Toll-like receptor 9 ligand. As a result, antigen-presenting cells pick antigens from dying tumor cells and are stimulated to present them to T cells. The ensuing T-cell response then eliminates systemic disease. These T cells can be transferred together with hematopoietic stem cells into tumor-bearing donors and can expand to eliminate large, established tumors. In the animal model, we have shown that both cell-death induction and direct intratumoral injection of the CpG are necessary. We have now completed a pilot clinical trial and documented therapeutic efficacy of this maneuver in patients with low-grade lymphoma. In the future, direct intralesional injection of CpG could be combined with many different methods of tumor cell death induction.

ANGIOGENIC MEDIATORS SUPPORT GROWTH AND METASTASIS IN MYELOMA AND LYMPHOMA

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Angiogenesis, blood vessel formation, is required for tumor growth, and its importance is becoming increasingly evident in hematologic malignancies. Various studies have attempted to correlate the number and character of tumor-associated blood vessels with clinical outcome. Additionally, several angiogenic mediators appear to be important in tumor biology. Vascular endothelial growth factor (VEGF) is a signaling protein with several isoforms and is centrally important in vasculogenesis. Chemokines also play a role in tumor-associated angiogenesis. One example is SDF-1 alpha, a CXC chemokine that has been associated with tumor angiogenesis in multiple myeloma (MM) patients. Increased angiogenesis has been linked with decreased survival in MM and with relapse or resistance to chemotherapy in lymphoma. Therefore, it appears that antiangiogenic strategies may have a role in potentially improving outcomes for patients with MM or lymphoma. A relatively nonspecific strategy is the use of metronomic chemotherapy. Continuous or intermittent chronic use of low-dose oral cytotoxic agents can have antitumor effects in various settings, including those where higher dose, bolus chemotherapy is ineffective. An example of such a strategy is the PEP-C regimen, which has been explored by our group in a variety of settings and which is active and well-tolerated. Bevacizumab, a recombinant humanized antibody, inhibits angiogenesis by blocking VEGF receptors and is now being tested in lymphomas and multiple myeloma. Bevacizumab is undergoing testing with CHOP-based chemotherapy (+/- rituximab) in a variety of settings. In one study, diffuse large B-cell lymphoma (DLBCL) patients treated with a combination of R-CHOP (rituximab, cyclophosphamide, doxorubicin, vincristine, and prednisone) and bevacizumab had an 85% overall response rate. Immunomodulatory agents such as thalidomide and lenalidomide are also undergoing extensive evaluation in myeloma and lymphoma and clearly have important therapeutic activity. While they appear to work through a variety of pathways, one mechanism of action relates to their effects on tumor angiogenesis. Finally, there are a number of novel compounds, including small molecule VEGF receptor inhibitors and heat shock protein 90 inhibitors, with antiangiogenic effects that are currently being studied. Emerging data suggest that the incorporation of strategies targeting tumor vasculature along with standard treatments or other novel agents is likely to ultimately contribute to improved outcomes for patients with lymphoma and myeloma.