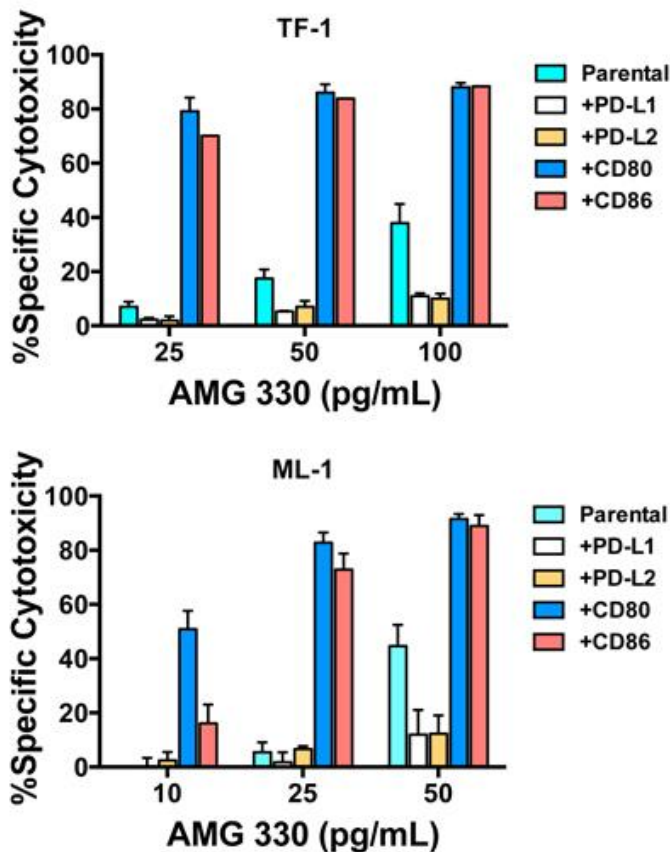# T-cell ligands take the teeth out of BiTE antibodies

October 19, 2015

JA Herman

AMG 330-induced cytotoxicity displays dose-dependent response in CD33+ human AML cells. Expression of inhibitory T-cell ligands, PDL-1 and PD-L2, reduces the activity of AMG 330, while stimulatory T-cell ligands, CD80 and CD86, sensitize cells to AMG 330 activity.

Image provided by Dr. George Laszlo

Antibody based therapeutics have heralded major advances in cancer treatment. A long-pursued therapeutic approach in this class is bi-functional antibodies that mediate the interaction between tumor and immune cells. The goal of such a therapy is to induce cytotoxicity, but more importantly to do so in targeted manner as an antigen-specific immunotherapy. Such bispecific antibodies exhibited limited clinical success until miniaturized molecules containing only minimal binding domains were generated. These streamlined molecules are termed BiTE (Bispecific T-cell engaging) antibodies and have already seen clinical success. These molecules generally bind T-cells by targeting the invariant epsilon subunit, CD3, however the tumor-specific marker can be vastly different depending on the cancer to be targeted. For example, Blinatumomab has seen clinical success in refractory and relapsed acute lymphoblastic leukemia (ALL) using the tumor-specific

marker, CD19. AMG 330 is another BiTE therapy, which was developed due to Blinatumomab's success in patients. AMG 330 is designed to bind CD33 rather than CD19 because it targets tumors in patients with acute myeloid leukemia (AML). Patients are currently being enrolled for a phase I clinical trial thanks, in part, to preclinical successes. In their recent study in *Blood Cancer Journal*, Dr. George Laszlo and colleagues in the Walter Laboratory (Clinical Research Division), demonstrate that the entire composition of T-cell receptors expressed in AML patients affects response to AMG 330.

Two AML cell lines were engineered to amplify expression of either inhibitory (PD-L1/2) or activating (CD80/86) T-cell ligands. Using these derived cell lines, Dr. Laszlo and colleagues found that AMG 330 cytotoxicity was strongly repressed by either PD-L1 or PD-L2 expression. The resistance to AMG 330 activity is due to specific PD-L1/2 activity as antibodies targeting those ligands sensitized cells to AMG 330 treatment. This finding is important both for understanding heterogeneity in patient responses to AMG 330 and for addressing mechanisms of drug resistance that may evolve. Already, it appears, the question of drug resistance will be a more pertinent application of this study, " Our PD-L1 phenotyping studies on patient specimens revealed little in the way of PD-L1 expression in previously frozen AML specimens, however another study recently published in *Leukemia* demonstrated PD-L1 expression due to prolonged exposure of AML patient specimens to AMG 330" , said Dr. Laszlo.

Understanding the mechanism of AMG 330 resistance and patient response in AML may provide the answer for overcoming those challenges. Dr. Roland Walter hopes this and future work will impact the application of BiTE antibodies " 2-fold: first, expression of T-cell ligands could be explored as biomarkers for response (e.g. to select patients most appropriate to receive drug); second: if ligands can be documented, then combination therapies could be explored."

Laszlo GS, Gudgeon CJ, Harrington KH, Walter RB. 2015. T-cell ligands modulate the cytolytic activity of the CD33/CD3 BiTE antibody construct, AMG 330. Blood Cancer J. 5:e430.