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have relevance with regard to nonspecific toxicity.

There is a distinct need in the field for CD33-based ADCs to quickly fill the void left by the withdrawal of GO from the market, and the rapid preclinical and clinical development of agents such as SGN-CD33A is needed.

Conflict-of-interest disclosure: The author declares no competing financial interests. ■

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● ● ● MYELOID NEOPLASIA

Comment on Kung Sutherland et al, page 1455

Precision 're'arming of CD33 antibodies

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In this issue of *Blood*, Kung Sutherland et al report on the preclinical activity of SGN-CD33A, a humanized anti-CD33 antibody conjugated to a pyrrolbenzodiazepine (PBD) dimer via a protease-cleavable linker, against acute myeloid leukemia (AML) cells in vitro and in vivo.¹

At this time, there is a void in the field of antibody–drug conjugates (ADC) targeting CD33 as a therapeutic option for treatment of AML after the voluntary withdrawal of gemtuzumab ozogamicin (GO) from the market.

Even though CD33 is an antigen that is both considered to be expressed in more committed myeloid precursors^{2,3} and is not present in AML stem cells, the clinical relevance of CD33 as a target is validated by survival benefit with GO in subgroups of patients with AML in randomized clinical trials.⁴⁻⁶ The reliable efficacy of drug conjugation with antibody and the greater stability of conjugate to avoid the exposure of non-target-expressing tissue to drugs can improve CD33 ADCs as therapeutic agents. The PBD dimer released after protease cleavage of SGN-CD33A causes DNA cross-linking and is capable of inducing cell cycle arrest and apoptosis. Engineered cysteine moieties at linker attachment sites allow more precise PBD dimer loading of antibody, thus improving predictability of payload delivery. The activity of SGN-CD33A requires CD33 expression, but its activity does not correlate with levels of CD33 surface expression. In vitro studies with SGN-CD33A showed approximately 3-fold more potency than GO shows against primary AML cells. What makes SGN-

CD33A potentially interesting is that this ADC appears to have activity irrespective of the multidrug resistance phenotype and p53 status of AML cells. Additional in vivo studies using immune-competent isogenic mouse models of disseminated AML with relevant translocations/mutations and variable p53 backgrounds are needed to add valuable information to the current report. In addition, data regarding levels of “naked” PBD dimer in plasma after infusions are important, as this may

● ● ● TRANSFUSION MEDICINE

Comment on Stowell et al, page 1494

A mouse model of hemolytic disease of the newborn

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In this issue of *Blood*, Stowell et al describe a novel mouse model of hemolytic disease of the fetus and newborn (HDFN) that recapitulates many of the key features of human disease.¹ Recently, this same group of researchers described a transgenic mouse that expresses the human KEL2 (Chellano) red cell surface protein from the Kell system on red cells,² and subsequently demonstrated that Kell differences on transfused blood induce antibody responses and hemolytic transfusion reactions similar to those seen in patients.³ In this latest report,

Stowell et al¹ demonstrate that similar to some patients, Kell differences between mother and father can lead to maternal antibody generation and hemolytic disease in utero. In so doing, they provide experimental confirmation of a long sought after animal model of HDFN.

HDFN can be thought of as a clinically relevant natural experiment in human immunology, whereby allogenic paternal proteins expressed on red blood cells in the fetus are capable of elucidating a class switched, IgG alloantibody response in the mother.⁴ Exposure to red cell antigens typically occurs at the time of delivery, sparing the initial pregnancy. However, with subsequent pregnancies, transfer of maternal IgG to the developing fetus leads to antibody binding and destruction of developing fetal red blood cells. Alternatively, exposure of women of child-bearing age to paternal red cell antigens via transfusion can lead to antibody production capable of impacting even the first pregnancy. Classically, antibody titers and disease severity both increase with subsequent pregnancies; with lower titers associated with neonatal anemia and hyperbilirubinemia and higher titers associated with erythroblastosis fetalis and interuterine death. Stowell et al¹ convincingly demonstrate in their Kell-expressing transgenic mouse model that repeated exposure of Kell negative moms, via

either multiple pregnancies or transfusion, leads to increasing anti-Kell antibody titers and worsening of HDFN of their offspring (see figure). By using mothers that are genetically incapable of antibody production, they definitively demonstrate that these antibodies drive the observed pathology (see figure).

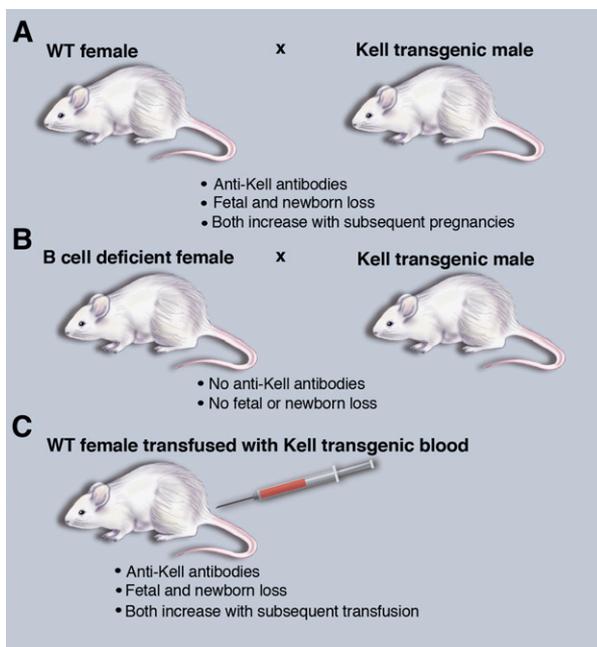
Despite the initial description of HDFN in 1609 by a French midwife and the discovery of the antibody-mediated disease mechanism underlying it in the 1940s,⁴ our understanding of the basic immune mechanisms that govern induction of antibody responses in the relatively immunosuppressive environment of pregnancy remains unclear. The new Kell transgenic system described by Stowell et al¹ provides an experimentally tractable model in which to address basic questions regarding the molecular mechanisms of initial immune sensitization. Perhaps more importantly, their work provides a clinically relevant model in which to investigate novel methods for prevention and treatment of HDFN going forward. Although polyclonal Rh(D) antisera (RhoGam) has proven

tremendously successful in the prevention of the most common setting of HDFN driven by the Rh-D antigen, the mechanism of action of RhoGam remains a mystery.⁵ There are also many other RBC antigen systems capable of inducing severe HDFN for which there is no RhoGam equivalent, including the Kell, Kidd, and the non-D Rh antigens.⁶ In these non-D Rh mismatch settings, there is no proven prevention strategy and clinical management often requires interuterine transfusion strategies that themselves come with significant risk of fetal loss. Given its continued clinical importance, there have been several attempts throughout the years to generate animal models of HDFN. These attempts have focused on Rh(D) expression in one form or another, and they have universally proved unsuccessful, likely secondary to the combined effects of the genetic complexity of the Rh system⁷ and differences between mouse and human MHC class II peptide presentation. By focusing on the genetically less complicated, yet clinically relevant Kell system, Stowell et al¹ have finally overcome this important hurdle. In so doing, they have provided a pre-clinical model in which development and testing of novel therapeutic approaches for the prevention and/or treatment of this devastating disease is now possible.

Conflict-of-interest disclosure: The authors declare no competing financial interests. ■

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A mouse model of HDFN that recapitulates human disease. (A) Breeding wild-type (WT) female mice with Kell transgenic male mice leads to HDFN. (B) HDFN depends on B-cell production of antibodies. (C) Transfusion of Kell transgenic blood into wild-type females also leads to HDFN. Professional illustration by Marie Dauheimer.