benefit from HSCT vs those who would be better served by participating in clinical trials with newer antileukemic therapies.

Abnl(17p) has emerged as one of the worst cytogenetic aberrations in AML. Although abnl(17p) may be conveniently identified by conventional metaphase cytogenetics or fluorescence in situ hybridization (FISH), it is only a part of the mutational spectrum in the tumor protein 53 (*TP53*) gene. TP53 is highly conserved across vertebrates and, in humans, is located on chromosome 17p. Most mutations in *TP53* remain cryptic and are only identified by gene sequencing. What do we know about *TP53* mutations in AML? Whereas cytogenetically defined abnl(17p) is seen in ~5% of AML, the frequency of *TP53* mutations found by sequencing is 8% in de novo AML.[2] *TP53* mutation is the hallmark genetic aberration, rising to ~30%, in secondary AML.[3] Significantly, *TP53* mutations are driver mutations, in most cases mutually exclusive with mutations involving the transcription factor fusion genes *NPM1*, *RUNX1*, *Flt3-ITD*, or *CEBPA* and are highly correlated with a complex karyotype.[2,4–6]

In this retrospective analysis of pooled registry data, Middeke et al have reported on the outcomes of 201 AML subjects with abnl (17p) by cytogenetics or FISH, all of whom underwent allogeneic HSCT. This is the first large retrospective study to derive definitive conclusions about the value of HSCT with this cytogenetic abnormality. Overall survival (OS) was only 22% even in CR1, and early relapse was the greatest contributor to mortality with a cumulative incidence estimate of 49%.[1] Such dismal outcomes are comparable to the monosomal karyotpye[7] and *Flt-3 ITD*[8] mutations that confer OS of ~20% despite prompt HSCT in CR1. Although the OS with abnl(17p) after HSCT is certainly better than the OS of 0% described in another large series,[4] it is clear that HSCT produces disappointing outcomes.

The poor prognosis of this subtype of AML is not at all surprising. *TP53* is frequently referred to as the "guardian of the genome" based upon its crucial role as a tumor suppressor gene in protecting against mutations in the genome. *TP53* mutations may occur infrequently in the germline (Li-Fraumeni syndrome) but more commonly as acquired somatic mutations that are involved in ~50% of all malignant conditions.[9] More than 1000 different somatic mutations have been described, defying drug development. Mutational effects range from recessive loss of function (with an intact functional copy of the wild-type [WT] allele) to dominant-negative mutations (which dimerize to inactivate WT TP53). TP53 works as the critical mediator of a network that senses cellular stress and, in turn, regulates many other genes. TP53 activation either halts cell proliferation in order to facilitate DNA repair or kills the cell when damage is irreparable (see figure). Loss of TP53 function has been associated with invasion, proliferation, and metastasis. Importantly, *TP53* mutation also confers resistance to genotoxic agents, which translates into refractoriness to cytotoxic chemotherapy and irradiation. Conventional small-molecule approaches to restore the functional loss of a defective tumor suppressor have not been successful but there are several promising therapies in development.[10] It is therefore notable that the modest benefit of HSCT in abnl(17p) AML probably suggests that there is residual susceptibility to the graft-versus-leukemia (GVL) effect. Definitive evidence for GVL against abnl(17p) AML, such as responses to donor lymphocyte infusions, is still lacking. However, this does support exploring immunotherapeutic approaches against what is currently an "undruggable" target. Given the critical importance of *TP53* mutation in oncogenesis and the absence of a US Food and Drug Administration (FDA)–approved therapeutic, there is an urgent need for strategies that may restore the "guardian of the genome" to its guard post.

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

REFERENCES

1. Middeke JM, Fang M, Cornelissen JJ, et al. Outcome of patients with abnl(17p) acute myeloid leukemia after allogeneic hematopoietic stem cell transplantation. *Blood.* 2014;123(19):2960-2967.

2. Cancer Genome Atlas Research Network. Genomic and epigenomic landscapes of adult de novo acute myeloid leukemia. *N Engl J Med.* 2013;368(22):2059-2074.

3. Wong TN, Ramsingh G, Young A, et al. The role of early TP53 mutations on the evolution of therapy-related AML [abstract]. *Blood.* 2013;122(21). Abstract 5.

4. Grossmann V, Schnittger S, Kohlmann A, et al. A novel hierarchical prognostic model of AML solely based on molecular mutations. *Blood.* 2012;120(15): 2963-2972.

5. Bowen D, Groves MJ, Burnett AK, et al. TP53 gene mutation is frequent in patients with acute myeloid leukemia and complex karyotype, and is associated with very poor prognosis. *Leukemia.* 2009;23(1): 203-206.

6. Seifert H, Mohr B, Thiede C, et al; Study Alliance Leukemia (SAL). The prognostic impact of 17p (p53) deletion in 2272 adults with acute myeloid leukemia. *Leukemia.* 2009;23(4):656-663.

7. Cornelissen JJ, Breems D, van Putten WL, et al. Comparative analysis of the value of allogeneic hematopoietic stem-cell transplantation in acute myeloid leukemia with monosomal karyotype versus other cytogenetic risk categories. *J Clin Oncol.* 2012;30(17): 2140-2146.

8. Sengsayadeth SM, Jagasia M, Engelhardt BG, et al. Allo-SCT for high-risk AML-CR1 in the molecular era: impact of FLT3/ITD outweighs the conventional markers. *Bone Marrow Transplant.* 2012;47(12): 1535-1537.

9. Hollstein M, Sidransky D, Vogelstein B, Harris CC. p53 mutations in human cancers. *Science.* 1991;253(5015): 49-53.

10. Prabhu VV, Allen JE, Hong B, Zhang S, Cheng H, El-Deiry WS. Therapeutic targeting of the p53 pathway in cancer stem cells. *Expert Opin Ther Targets.* 2012;16(12): 1161-1174.

● ● ● IMMUNOBIOLOGY

Comment on Steward-Tharp et al, page 2978

The right "Job" for STAT3 mutant mice!

Stuart G. Tangye[1,2] [1]GARVAN INSTITUTE OF MEDICAL RESEARCH; [2]UNIVERSITY OF NEW SOUTH WALES

In this issue of *Blood*, Steward-Tharp et al report the generation of a murine model of the human primary immunodeficiency autosomal dominant hyper-immunoglobulin E syndrome (AD-HIES) and reveal novel insights and therapeutic outcomes for this fascinating human monogenic disorder.[1]

"So Satan went forth from the presence of the Lord, and smote Job with sore boils from the sole of his foot unto his crown." This quote from the book of Job prefaced the subject of a clinical report by Davis et al that, in 1966, provided the first description of a new primary immunodeficiency. It was named Job's syndrome after the biblical figure Job who

was covered in boils, not unlike the patients detailed in that report.[2] Job's syndrome is characterized by recurrent staphylococcal infections of the lung and skin, as well as chronic mucocutaneous candidiasis.[2-5] Strikingly, viral susceptibility is not a common clinical complication of Job's syndrome patients, although recent reports suggest problems controlling reactivation of some herpes viruses.[6] Following the discovery of immunoglobulin E (IgE) in the early 1970s, Buckley noted that these patients had extremely high levels of IgE,[3] which led to renaming Job's syndrome as autosomal dominant hyper-immunoglobulin E syndrome (AD-HIES). Buckley also reported defective humoral immune responses in patients with Job's syndrome, despite normal levels of serum IgM, IgG, and IgA.[3] Additional features of AD-HIES include eosinophilia, susceptibility to B-cell lymphoma, and nonimmunologic defects affecting musculoskeletal, connective tissue, dental, and circulatory systems.[4,5]

Despite increased understanding and awareness of the clinical features of AD-HIES over the ensuing decades, it was not until 2007—41 years after the initial description[2]—that the genetic defect underlying this condition was revealed. Elegant studies by Minegishi et al[7] and Holland et al[8] discovered that heterozygous mutations in *STAT3* caused AD-HIES. Subsequent studies of lymphocytes from patients with AD-HIES identified critical roles for STAT3 in generating Th17 cells, Tfh cells, and memory B and $CD8^+$ T cells and the ability of naïve B cells to differentiate into plasmablasts in response to STAT3-activating cytokines.[5] These findings provided a cellular basis for several key clinical features of AD-HIES, namely susceptibility to recurrent fungal and possibly staphylococcal infections (Th17 deficit), impaired humoral immunity (Tfh and B-cell defects), and susceptibility to B-cell lymphoma and latent herpes viruses (reduced memory $CD8^+$ T cells).[5,6] However, studies in humans are limited by ethical and logistical constraints; thus, an animal model of STAT3 deficiency would be a valuable resource for further exploring molecular and cellular mechanisms underlying disease pathogenesis in AD-HIES.

Although *Stat3*-gene targeted mice were generated in 1997, their utility as an experimental model was curtailed by embryonic lethality following constitutive Stat3 deletion.[9] This finding had at least two important outcomes. First, Stat3-null mice provided little indication that *STAT3* mutations could underlie a primary immune-deficient condition in humans. Second, there were substantial differences between Stat3-null mutations in mice and heterozygous mutations in humans, with the former being fatal and the latter, although compatible with life, were deleterious for human health. This reflected residual activity of the wild-type *STAT3* allele that permitted placental implantation and embryogenesis but was clearly insufficient for immune cell function. However, the generation of mice lacking Stat3 in specific lineages did reveal defects in Th17 cells, Tfh cells, and antigen-specific antibody (Ab) responses when *Stat3* was selectively deleted from $CD4^+$ T cells or B cells.[5] Even with the knowledge that *STAT3* mutations caused AD-HIES, Stat3 conditionally deficient mice had limited application as an in vivo model because loss of Stat3 in individual cell lineages failed to recapitulate the full phenotype of disease.

Thus, a new mouse model was required that would be more faithful to the human condition. To achieve this, Steward-Tharp et al[1] generated BAC-transgenic mice expressing a deletion mutation (V463del) in *Stat3* that is relatively common in AD-HIES patients.[7,8] Strategically, the authors selected 1 mouse strain expressing 2 copies of the transgene, thus recreating the heterozygous and dominant negative nature of the human *STAT3* mutation. Detailed analysis revealed significant similarities between these mice and AD-HIES patients, including normal lymphocyte development but impaired generation of Th17 cells, which resulted in susceptibility to infection with the gut-tropic bacteria *Citrobacter rodentium*, poor Ab responses and, importantly, hyper-IgE.[1] Although it could be argued that susceptibility to *C rodentium* is a feature of mice rather than the human condition, it is consistent with poor immunity in AD-HIES to infections at mucocutaneous sites which, like protection against *C rodentium*, require interleukin-17 (IL-17) and IL-22 production by $CD4^+$ T cells and IL-22–induced STAT3-dependent expression of antimicrobial proteins in epithelial cells. *Stat3*-mutant mice also developed severe *C rodentium*–induced inflammatory bowel disease and significant mortality following lipopolysaccharide challenge[1]; both of these were attributed to increased production of proinflammatory cytokines (tumor necrosis factor α [TNF-α], IL-12, interferon gamma [IFN-γ]) and reduced IL-10, which signals through STAT3. A curious feature of AD-HIES is a marked lack of inflammation,[2,4] which may reflect a predominant role for IL-6/STAT3 signaling in human inflammation that is not phenocopied in mice or may be unique to *C rodentium* infection in rodents.

Although AD-HIES is not a fatal immunodeficiency, it is nonetheless life shortening.[4] Thus, insights into therapeutic options would be valuable. Although stem cell transplantation (SCT) of AD-HIES patients has been attempted, results have been inconsistent.[10] Given the ubiquitous expression of STAT3 and the nonimmunologic features of AD-HIES, it is perhaps not surprising that SCT has had mixed outcomes. Reconstitution of *Stat3*-mutant mice with wild-type bone marrow restored Th17 responses, but this provided only partial protection against *C rodentium*, demonstrating that, although transplanted mice could generate appropriate cytokines for protection against this pathogen, these effector cytokines—probably IL-22—need to exert their function via STAT3 in nonhematopoietic cells (ie, epithelia) to provide complete protection against infection. These findings suggest that SCT of AD-HIES patients may have only a partial impact on infectious susceptibility, with additional prophylactic treatments required. However, SCT will probably cure defects in Ab responses in AD-HIES.

Overall, this is a very exciting model that, unlike germline *Stat3* deletion, mimics the genetic lesion in AD-HIES, thereby allowing detailed dissection of cellular defects and disease in affected individuals. And although this model may not tell us what causes retention of primary teeth in AD-HIES patients, it may provide answers to the age-old question of what causes hyper-IgE and may identify mechanisms causing nonimmunologic defects. Furthermore, it could represent a preclinical model to test drugs considered for treating AD-HIES. So, there are plenty of "Job's" in store for these mice!

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

REFERENCES

1. Steward-Tharp SM, Laurence A, Kanno Y, et al. A mouse model of HIES reveals pro- and anti-inflammatory functions of STAT3. *Blood*. 2014;123(19):2978-2987.

2. Davis SD, Schaller J, Wedgwood RJ. Job's Syndrome. Recurrent, "cold", staphylococcal abscesses. *Lancet*. 1966; 1(7445):1013-1015.

3. Buckley RH, Wray BB, Belmaker EZ. Extreme hyperimmunoglobulinemia E and undue susceptibility to infection. *Pediatrics*. 1972;49(1):59-70.

4. Grimbacher B, Holland SM, Puck JM. Hyper-IgE syndromes. *Immunol Rev*. 2005;203:244-250.

5. Kane A, Deenick EK, Ma CS, Cook MC, Uzel G, Tangye SG. STAT3 is a central regulator of lymphocyte differentiation and function. *Curr Opin Immunol*. 2014;28C: 49-57.

6. Chandesris MO, Melki I, Natividad A, et al. Autosomal dominant STAT3 deficiency and hyper-IgE syndrome: molecular, cellular, and clinical features from a French national survey. *Medicine (Baltimore)*. 2012;91(4):e1-e19.

7. Minegishi Y, Saito M, Tsuchiya S, et al. Dominant-negative mutations in the DNA-binding domain of STAT3 cause hyper-IgE syndrome. *Nature*. 2007;448(7157): 1058-1062.

8. Holland SM, DeLeo FR, Elloumi HZ, et al. STAT3 mutations in the hyper-IgE syndrome. *N Engl J Med*. 2007;357(16):1608-1619.

9. Takeda K, Noguchi K, Shi W, et al. Targeted disruption of the mouse Stat3 gene leads to early embryonic lethality. *Proc Natl Acad Sci U.S.A*. 1997;94(8):3801-3804.

10. Goussetis E, Peristeri I, Kitra V, et al. Successful long-term immunologic reconstitution by allogeneic hematopoietic stem cell transplantation cures patients with autosomal dominant hyper-IgE syndrome. *J Allergy Clin Immunol*. 2010;126(2):392-394.

LYMPHOID NEOPLASIA

Comment on Iqbal et al, page 2915, and on Wang et al, page 3007

Cellular origin of T-cell lymphomas

Laurence de Leval[1] and Philippe Gaulard[2] [1]UNIVERSITY HOSPITAL OF LAUSANNE; [2]UNIVERSITY HOSPITAL HENRI MONDOR

In this issue of *Blood*, Iqbal et al,[1] having compiled gene expression profiles from >300 peripheral T-cell lymphomas, expand previous findings on the diagnostic value of molecular signatures that correlate with different histological types of T-cell lymphomas. They report the discovery of 2 molecular subgroups of peripheral T-cell lymphomas, not otherwise specified (PTCL, NOS), characterized by high expression of either GATA-binding protein 3 (GATA-3) or t-box 21 (TBX21) transcription factors and corresponding target genes, with the GATA3 subgroup being associated with distinctly worse prognosis. In an independent study, Wang et al[2] also show that GATA3 expression in a subset of PTCL, NOS identifies a subgroup of patients with inferior survival.

PTCLs—a term collectively designating malignancies derived from mature T cells and natural killer cells—comprise a heterogeneous group of disease entities that are overall rare, accounting for <15% of all non-Hodgkin lymphomas worldwide.[3] PTCLs pose challenges in several respects. Clinically, most entities are aggressive diseases with overall poor response to classical treatments and carry a dismal prognosis.[4] With respect to classification and diagnosis, in contrast with the main B-cell lymphomas entities that are defined by a combination of morphologic, immunophenotypic, genetic, and clinical features and are linked to a normal cellular counterpart, many T-cell lymphoma entities are pathologically heterogeneous, most of them lack defining genetic aberrations, and their classification relies on less well-characterized diagnostic criteria.[5] The paradigm is represented by PTCL, NOS, which represents the largest PTCL entity and is defined "by default" as encompassing cases not fulfilling criteria allowing categorization in a more specific entity. Not unexpectedly, PTCL, NOS is heterogeneous, both pathologically and clinically, and although it is has been repeatedly stated that it may encompass several distinct entities, various attempts to identify biologically relevant subgroups have yet mostly failed.[6]

The discovery of 2 distinct molecular subgroups of PTCL, NOS identified by unsupervised analysis of genome-wide molecular profiles reported by Iqbal et al represents a major step forward in deciphering the heterogeneity of PTCL, NOS. Interestingly, the 2 subgroups, defined by overexpression of GATA3 or TBX21 (*t*-bet) and associated target genes, are biologically meaningful. Both GATA3 and TBX21 are transcription factors that are master regulators of gene expression profiles in T helper (Th) cells, skewing Th polarization into Th2 and Th1 differentiation pathways, respectively. The molecular signatures of lymphomas associated with high expression of these transcription factors were also enriched in other Th2- or Th1-associated transcripts, and GATA-3–positive PTCLs are associated with eosinophilia that is typically mediated by Th2 cytokines. These novel findings add to the increasing evidence that cell lineage is a major determinant of PTCL biology and defining factor for the delineation of PTCL entities or subgroups. Considering Th cell subsets (see figure), it is already known that the follicular helper subset represents the cellular origin of angioimmunoblastic T-cell lymphoma,[7] whereas the neoplastic cells in human T-lymphotropic virus 1–associated T-cell lymphoma/leukemia generally exhibit a T-regulatory phenotype with expression of FOXP3 transcription factor.[8] The new data published by Iqbal and Wang and colleagues now suggest that a large proportion of PTCL, NOS is related to either Th1 or Th2 lineage derivation. There is, however, no perfect overlap between the signatures derived from the lymphoma subgroups and those of the normal Th subsets, likely reflecting the plasticity of the T-cell system. In addition, the TBX21 group might be more heterogeneous as it also comprises a subset of cases with a cytotoxic profile, and using the molecular classifier developed to distinguish between the GATA3 and TBX21 subgroups, there remains a proportion of cases whose gene expression signature is indeterminate and cannot be assigned to one or the other category. Thus, additional studies and analysis of larger cohorts of patients may be necessary to validate the current findings and possibly refine the molecular classification.

The data published by Iqbal et al suggest that the GATA3 signature associates with distinctly less favorable clinical outcome and shorter overall survival than the TBX21 signature. There appears to be a good correlation between the molecular signatures and protein expression; hence, immunohistochemistry for GATA3 and TBX21 is a reliable surrogate to the