tissues, including the spleen, lymph nodes, and the kidneys. Accordingly, VQ mice present a more profound impairment of hematopoiesis and ensuing anemia and increased immunoglobulin deposition in the kidneys, when compared with the Vκ*MYC model. Attesting to the aggressive nature of the disease in the VQ mouse, plasma cells are highly proliferative, and, intriguingly, present a gene expression pattern reminiscent of the high-risk myeloma signature (UAMS-70). As expected, the survival of the VQ mice is severely affected, with the animals living only for a few months.

The translational relevance of the VQ model has also been explored by Wen et al and is quite remarkable. For example, as it has been for the Vκ*MYC model,[4] the VQ mouse was successfully tested as a drug screening in vivo platform, with drugs already used in the clinic to treat myeloma patients or more experimental compounds. In addition, VQ myeloma cells preserve both the PD1 and the TIGIT immune checkpoint pathways, which mediate T-cell suppression in myeloma patients. As such, the VQ mice could be exploited for the preclinical evaluation of immunotherapeutic strategies in an aggressive myeloma setting. Finally, tumor cells derived from this model could be transplanted into sublethally irradiated syngeneic recipients for several passages and, importantly, could be transduced with both retro- and lentiviruses, thus enabling additional genetic manipulations.

Thus, are we there yet? Is this the end of the journey? The model presented herein represents an important step forward, toward the generation of a reliable mouse model of aggressive myeloma. However, in solid tumors, the knowledge of the underlying genetic lesions has led to the generation of increasingly refined mouse models, as a result of the accrual of layers of genetic engineering.[2] Although MYC and NRAS certainly represent crucial genetic events in the development of the disease,[4,5] myeloma is a genetically heterogeneous disease, with several other genes and pathways that are engaged in its development, in specific patient subsets.[5] These lesions have not yet been properly modeled. There is indeed the possibility that the platform described in this study may represent the foundation for models where additional lesions could be engineered, to capture more closely the puzzling intra- and interpatient heterogeneity of human myeloma.

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MYELOID NEOPLASIA

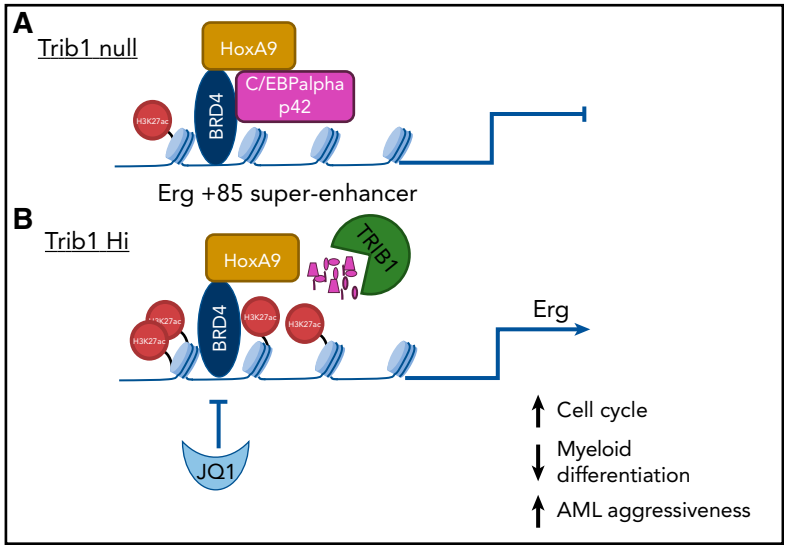
Comment on Yoshino et al, page 75

Superenhancing AML with Trib1

Karen Keeshan | University of Glasgow

In this issue of *Blood*, Yoshino et al use the genetic Trib1-ROSA26-Cre–knockout murine model and retroviral-mediated HoxA9 overexpression, together or not with Trib1 reexpression (Trib1 Hi and Trib1 null), to investigate the mechanistic basis for HoxA9-mediated acute myeloid leukemia (AML).[1] They have deciphered the mechanism involved in HoxA9-associated AML, linking an Erg-specific superenhancer with the Trib1-C/EBPα axis. It has previously been shown that TRIB proteins, including TRIB1 and TRIB2, are AML oncoproteins capable of driving disease development. Degradation of the p42 isoform of C/EBPα (an important myeloid transcription factor) and increased MAPK/ERK signaling (a proliferation and survival indicator) are key molecular mechanisms for Trib oncogenic activity.[2] Cooperation with HoxA9 leads to accelerated AML development,[3,4] which is important because HoxA9 overexpression alone is unable to drive AML in vivo. The role that TRIB proteins may have across the wide heterogeneity of AML phenotypes is still underappreciated; however, given that deregulated Hox signaling is associated with ∼70% of all AMLs, it is important to understand the involvement of Trib oncogenic activities in HoxA9-mediated AML. It remained an open question how TRIB proteins cooperate with Hoxa9 at the mechanistic level.

Previous data have shown that HoxA9 is organized into enhanceosomes containing lineage-restricted transcription factors, including C/EBPα.[5] Together with what is known about TRIB1 oncoprotein function, this led the investigators to hypothesize that



Schematic depiction of Erg +85 superenhancer in Trib1 null and Trib1 Hi HoxA9-expressing leukemia cells. (A) Trib1 null; (B) Trib1 Hi.

Trib1-mediated degradation of C/EBPα p42 isoform would lead to enhanced remodeling at HoxA9-associated genomic loci and, thus, be a key event in HoxA9-Trib1 cooperation in AML. The investigators tested this using microarray gene expression and chromatin immunoprecipitation sequencing analysis and could show that, in the absence of Trib1 (Trib1 null) in HoxA9-expressing cells, C/EBPα p42 is expressed, and a HoxA9-C/EBP complex and H3K27ac are present at distal enhancers. The HoxA9-C/EBP complex is associated with gene activation and repression and is known to contribute to the aberrant proliferative phenotype in an AML model that required coexpression of MEIS1.[6] In this article, the investigators now advance our understanding of HoxA9 leukemogenesis (see figure); HoxA9 DNA binding peaks and H3K27ac peaks were not significantly different between TRIB1 hi and TRIB1 null cells, but HoxA9-associated superenhancers specific to TRIB1 hi AML cells were enriched, most notably at the Erg +85 superenhancer, together with H3K27ac. Importantly, it was the removal of C/EBPαa p42 isoform mediated by Trib1 that occurred at these specific HoxA9-associated superenhancer genomic loci leading to enhanced H3K27ac and elevated Erg expression. There is a requirement for C/EBPalpha expression in HoxA9-associated and TRIB-mediated AML,[6,7] which can seem counterintuitive. However, this is explained in the literature by the expression and opposing functions of C/EBPα p42 and p30 isoforms. The degradation of p42 isoform, while p30 expression remains intact, is a feature of AML[8] and is consistent with the literature; here, it is shown to be a driving force for enhancer modification at the HoxA9-binding specific Erg superenhancer.

The investigators tested the therapeutic targeting of this mechanism with the use of the BRD4 inhibitor JQ1, because BRD4 is abundant at superenhancers. JQ1 suppressed the leukemic superenhancer activity of TRIB1 hi AML cells in vitro and in vivo, consistent with a block in Erg target gene expression. It remains an open question to what degree the expressions of Hox proteins and Trib proteins are linked across AML. Using messenger RNA expression has not revealed strong correlations among HoxA9, Trib1 or Trib2, and Erg; however, as elegantly shown in this article, oncoprotein activity and protein isoform expression are the key factors distinguishing AML with highly aggressive features that can be therapeutically targeted. Erg expression is associated with poor prognosis in AML,[9] as well as a more aggressive phenotype[10]; therefore, its elevated expression as a result of Trib1 function in HoxA9-associated leukemia has important implications for patient prognostication and, potentially, AML stratification.

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