underlying rationale exists where miRNA depletion causes particular proliferative or survival advantages in these cancers. Indeed, it seems rather mysterious that a pathway that is considered important to provide robustness to gene expression programs would display itself so little robustness to mutational assault. It will therefore be of particular interest to examine whether haploinsufficiency is a general feature of miRNA pathway genes across various cancer sites and types or more closely restricted to MSI^+ tumors.

Finally, it remains to be seen whether the new findings can be exploited therapeutically. At this point, short of gene therapy, there seems to be little that can be done to target the *XPO5* defect directly so that restoring miRNA accumulation by alternative routes might be a more realistic approach. Provided that only one or few of the deregulated miRNAs are responsible for the tumor-promoting effect of *XPO5* mutation, it may be possible to supply them exogenously as miRNA duplexes that would not need to undergo nuclear export. Alternatively, it may be possible to identify a subset of key targets of the deregulated miRNAs that might be amenable to inactivation through classical pharmacological approaches or novel biologics.

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T-Lineage Lymphoblastic Lymphoma and Leukemia—a MASSive Problem

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T cell precursor malignancies may present as T-lymphoblastic lymphoma (T-LBL) with marked enlargement of lymph nodes or acute T-lymphoblastic leukemia (T-ALL) with little lymph node enlargement. In this issue of *Cancer Cell*, Feng et al. show that dysregulation of *BCL2*, AKT signaling, and cell adhesion pathways are hallmarks of T-LBL.

T-lineage lymphoblastic lymphoma (T-LBL) and acute lymphoblastic leukemia (T-ALL) represent up to 15%–25% of cases of ALL in children and adults and exhibit a remarkable spectrum of clinical, pathologic, and genetic features. In contrast to B-progenitor ALL, in which extensive bone marrow involvement at presentation is a near universal feature and significant lymph node enlargement is uncommon, patients with T-lineage disease frequently present with marked lymph node enlargement in the chest (mediastinal masses), often with minimal or absent bone marrow involvement (Figure 1). Traditionally, patients with a mediastinal mass and less than 25% leukemic cells (blasts) in the bone marrow are deemed to have T-LBL; those with a high marrow burden, T-ALL. While this distinction may appear somewhat arbitrary, a notable observation is that patients with large mediastinal masses frequently exhibit little, if any, evidence of tumor dissemination and marrow involvement, and the basis for this is unknown.

Both T-LBL and T-ALL cases commonly harbor chromosomal rearrangements, submicroscopic DNA copy number alterations, and sequence mutations. These alterations commonly dysregulate or disrupt genes with key roles in hematopoietic development, lymphoid differentiation, cell cycle regulation, and tumor suppression and are key events in leukemogenesis (e.g., rearrangements of T cell antigen receptor genes, *HOX11L1* and *HOX11L2*, *TAL1*, *LYL1,* mutation of *NOTCH1* and *FBXW7*, and deletion or mutation of *PTEN* and *WT1*) (Aifantis et al., 2008). However, in contrast to B-progenitor ALL, in which specific genetic alterations such as mutation of the lymphoid transcription factor *IKZF1* are strongly associated with poor prognosis (Mullighan et al., 2009), identification of features that predict clinical

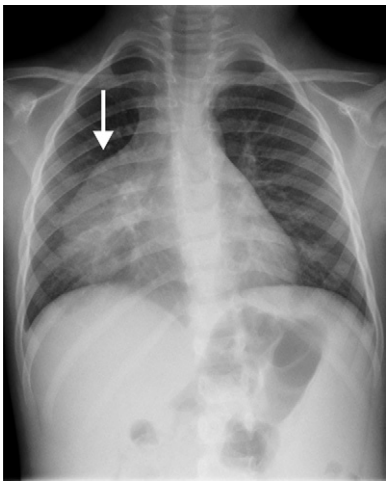

Figure 1. Chest Radiograph Showing a Mediastinal Mass, Indicated by an Arrow, in T-Lineage Lymphoblastic Lymphoma

features, behavior, or outcome in T-lineage disease has been frustratingly difficult. Peripheral blood leukocyte count, early responsiveness to therapy (Campana, 2010), and aberrant "early thymic progenitor" immunophenotype are associated with poor outcome (Coustan-Smith et al., 2009), but associations between individual genetic alterations and clinical features are, at best, weak (Karrman et al., 2009). Several studies have compared and contrasted clinical features and genetic alterations between T-LBL and T-ALL (reviewed in Burkhardt, 2010). While several differences have been reported, such as somewhat higher relapse rate in T-ALL, these differences are often subtle. Similarly, several studies have examined differences in cytogenetic alterations and submicroscopic genetic alterations in ALL, with no unique alterations in either T-ALL or T-LBL identified. Intriguingly, gene expression profiling has identified differences in expression of cell adhesion, cell cycle, and apoptosis genes between the two diseases, but the mechanistic basis of these differences has not been determined (Raetz et al., 2006).

In the current issue, Feng, Look, and colleagues have used a zebrafish model of T-LBL/ALL to investigate the basis of this dichotomy (Feng et al., 2010). Both T-ALL and T-LBL cells commonly harbor activating mutations of *NOTCH1* (Weng et al., 2004), which result in activation of MYC-regulated transcriptional networks (Palomero et al., 2006). In the current study, the authors used a transgenic *Myc*-driven zebrafish model of T-LBL, in which fish expressing *Myc* and *Bcl2* exhibited an increased penetrance and reduced latency of T-LBL than did fish transgenic for *Myc* alone. The authors found that while T-lineage disease developed in both *Myc:Bcl2*- and Myc-only fish, there was little systemic dissemination from *Myc:Bcl2* T-LBL, either from the primary tumors or after transplantation into secondary recipients. The Bcl2-expressing tumors also had a survival advantage and the propensity to form aggregates in long-term culture. Careful examination of the tumors showed local invasion in the *Myc:Bcl2* tumors, but a notable lack of vascular invasion.

The authors also observed increased *BCL2* expression in primary human T-LBL, but not T-ALL samples, thus implicating *BCL2* expression as an important determinant of the behavior of T-lineage disease. To investigate the underlying mechanism, the authors examined the ultrastructural and signaling characteristics of their tumors. They observed evidence of autophagy in both fish and human T-LBLs, but no evidence of dissemination in T-LBL fish treated with the autophagy inhibitor chloroquine, indicating that while autophagy may be a hallmark of T-LBL, it is not a critical determinant of dissemination. As AKT activation has been implicated in T cell migration and suppression of autophagy, the authors examined AKT activation in their tumors and noted markedly higher levels of phosphorylation of Ser473 of Akt, indicating Akt activation, in T-LBL tumors that disseminated. Introduction of myristoylated, constitutively active *Akt2* accelerated tumorigenesis and dissemination in *Myc:bcl2* tumors and inhibited in vitro aggregate formation, implicating Akt signaling in the ability to disseminate.

While these findings suggest a role of *BCL2* in influencing tumor behavior in T-lineage leukemia, they do not directly explain why *Bcl2*-overexpressing tumors display a singular lack of vascular invasion and dissemination. To further address this, the authors examined published gene expression profiling data comparing human T-ALL and T-LBL (Raetz et al., 2006). This analysis failed to detect enrichment of leukocyte adhesion and migration gene sets in human T-LBL but did identify increased expression of the adhesion molecule sphingosine-1-phosphate receptor 1 (S1PR1, or S1P1) and the adhesion molecule (and putative downstream target of S1PR1) ICAM1. S1PR1 expression was noted to be high in human T-LBL cells and immature cortical thymocytes in normal thymi, but low in more mature thymocytes in the thymic medulla and human T-ALL cells. As S1PR1 signaling has been reported to promote T cell adhesion and inhibit thymocyte emigration, this suggested a possible mechanism for retention of T-LBL cells in the thymus. Accordingly, the authors observed increased intravasation of T-LBL cells treated with the S1PR1 inhibitor W416.

Together, these data provide an important advance in our understanding of the biology of T-LBL. The results of Feng et al. (2010) strongly suggest that the accumulation of lymphoblasts in mediastinal masses is at least in part determined by the activation of signaling and adhesion pathways that inhibit the intravasation and subsequent dissemination of leukemic cells. This elegant model has proven highly informative but leaves several important questions to be answered in future studies. While the data for increased expression of BCL2, reduced activation of AKT, and activated S1PR1 signaling in both fish and human T-LBL are compelling, the nature of the upstream events dysregulating and linking these pathways is unknown. Existing genetic and genomic analyses have failed to identify alterations unique to T-LBL, and the nature of the genetic or epigenetic events activating these pathways are unclear. As the expression of S1PR1 by T-LBL cells is similar to that of immature cortical thymocytes, it is possible that the phenotype of T-LBL may in part reflect maturational stage, although this is not supported by differences in immunophenotype between T-LBL and T-ALL in clinical samples. Moreover, the mechanisms responsible for activation of BCL2 expression and aberrant AKT activation in T-LBL remain unclear. It is likely that future studies interrogating the genomic alterations in T-LBL in greater detail will provide important additional insights into this disease. However, the current study represents an important advance and raises the possibility that therapeutic inhibition of BCL2 and AKT may be beneficial in patients with the lymphomatous form of this disease.

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Epigenetics and MicroRNAs Combine to Modulate the MDM2/p53 Axis in Myeloma

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Avoiding the inhibitory effects of p53 on cell growth is important for tumor progression. In this issue of *Cancer Cell*, Picchiorri et al. describe epigenetic silencing of MDM2-targeting microRNAs in multiple myeloma (MM), which generally appears to have intact p53 function. This provides the rationale for microRNA-targeted therapy for MM.

Genetically, multiple myeloma (MM) can be divided into tumors with recurrent immunoglobulin heavy chain gene translocations or hyperdiploidy. Superimposed on this are a variety of presumably secondary genetic events, including activating mutations of *Ras*, rearrangements of *Myc*, mutations that activate the NFkB pathway, and late inactivating mutations of p53 (Fonseca et al., 2009). As noted by Pichiorri et al., (2010), mutations of p53 are rare in untreated MM and the tumors appear to have intact, if perhaps suppressed, p53 function, suggesting that therapeutic modulation of the p53/MDM2 pathway holds promise to help the majority of patients.

Deletion of one copy of p53 by FISH has been uniformly found to be an adverse prognostic factor with all therapies used in the treatment of MM: alkylating agents, proteasome inhibitors, and immune modulator-based therapies (Lode' et al., 2010; Kapoor et al., 2010). In contrast to patients with other poor-prognostic genetic lesions [e.g., t(4;14)], patients with deletion of p53 have seen less improvement in their survival with the addition of proteasome inhibitors and immune modulators. Nevertheless, data concerning loss of heterozygosity of p53 due to deletion of chromosome 17p and/or p53 mutation in MM have been confusing, perhaps at least in part because of old literature based on the analysis of unpurified bone marrow samples. In an analysis of CD138-purified myeloma cells from 716 patients, deletion of one copy of p53 was detected by FISH in 3% of MGUS (monoclonal gammopathy of undetermined significance), 1% of SMM (smoldering multiple myeloma), and 10% of MM (Chiecchio et al., 2009). There is a paucity of data on patients at the time of relapse; however, in another recent study, deletion of p53 was seen in 56% of primary plasma cell leukemia and 83% of the more aggressive secondary plasma cell leukemia (Tiedemann et al., 2008).

Somewhat surprising, given the strong correlation between loss of one copy of p53 deletion and survival, is the fact that in a cohort of 92 untreated MM patients, only 37% of patients with p53 deletions by FISH were found to have mutations on the remaining allele, although all those with mutations were found to also have p53 deletions (Lode' et al., 2010). It would be very informative to characterize whether in terminal samples from patients that initially presented with a 17p deletion by FISH, a p53 mutation has emerged under the selective pressure of therapy and disease progression. In the absence of such a study, we can examine the status of p53 in a panel of human MM cell lines derived from patients at the very end stages of the disease. It is notable that the majority of MM cell lines have p53 mutation (Mazars et al., 1992). Although one has to consider that cell lines have been under the selective pressure of being