Hox genes have the interesting property of specifying both spatial identity along the rostrocaudal axis of the developing embryo and differentiation status within the hierarchy of hematopoietic cell fates. Their importance to hematopoiesis is underscored by the overexpression of various Hox genes in hematologic malignancies, alone, as fusion proteins, or as downstream targets of deregulated MLL mutants. HOXA9, for example, is the gene most strongly correlated with poor prognosis in acute myeloid leukemia. The Hox gene family is large, comprising 39 transcription factors, each containing a DNA-binding homeodomain, and efforts to understand their differential effects mechanistically have made very slow progress. When expressed in a deregulated manner in bone marrow hematopoietic stem cells, most Hox family members cause leukemia, with the conspicuous exception of HOXB4, which does promote dramatic expansion of the hematopoietic stem cell compartment, but only up to a point limited by normal regulatory mechanisms. This property has generated considerable interest in the use of HOXB4 to derive or expand repopulating hematopoietic stem cells that could be used in therapeutic settings. Cusan et al now show that this property of regulated expansion requires a proline-rich domain (PRD) in the N-terminal half of the protein. When that domain was deleted, HOXB4 became oncogenic, and mice transplanted with bone marrow cells overexpressing HOXB4ΔPRD succumbed to a myeloid leukemia.

More interestingly, when the PRD of HOXB4 was transferred to the C-terminus of HOXA9, a Hox gene that promotes an aggressive leukemia in combination with MEIS1, it significantly mitigated the leukemic activity, suggesting that the PRD is a modular “antileukemic” domain. Understanding how this antileukemic regulation works would give much-needed insight into what underlies the oncogenic properties of the Hox gene family.

At a basic level, the first question to ask is whether the presence of the PRD changes the output of HOXB4 in a quantitative or qualitative manner, that is, are HOXB4 and HOXA9 simply less potent or perhaps less stable in the presence of this domain? The investigators ruled out effects on stability and went on to perform gene expression and chromatin immunoprecipitation (ChIP) sequencing analyses, which showed that the 2 versions of HOXB4 bound to very divergent sets of downstream target genes in 32D cells. Fewer than 10% of the targets of the ΔPRD version of HOXB4 were in common with those of wild-type (WT) HOXB4. It will be important to determine whether this remarkably altered specificity is mediated in cis, by modulating the sequence preference of the HOXB4 homeodomain, or in trans, perhaps by specifying different cofactors. Peculiarly, many target loci of HOXB4 identified in other studies were actually present within the set bound by HOXB4ΔPRD. Because these other studies used more primitive cell types for ChIP than did the 32D cells used in the Cusan et al study, it may be that the PRD renders HOXB4 more sensitive to chromatin environment and able to bind to its targets only if they happen to be in accessible chromatin. When PRD is removed, HOXB4 would be unleashed from this restriction and able to probe inaccessible chromatin, functioning more as a pioneer factor. Alternatively, perhaps in 32D cells, a myeloid-specific factor interacts with the PRD, dragging HOXB4 away from those targets that it prefers in stem cells.

The most interesting aspect of the PRD is its modularity—the fact that it can be transferred to HOXA9, where it imparts, superficially at least, a similar function. It will be important to round out the analysis of this chimera by evaluating whether HOXA9 + PRD exhibits a different profile of genomic targets than does WT HOXA9 and by determining how much this set has in common with WT HOXB4. Extended proline repeats often form polyproline helices, which can function as protein–protein interaction domains, with the moderately hydrophobic proline repeats attracting hydrophobic pockets of interactors of relatively low affinity and high reversibility. Proof of the modularity of this domain would be the eventual identification of such interacting factors and the determination of the functional effects of recruiting PRD-interacting factors to Hox complexes.

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LYMPHOID NEOPLASIA

Most were at an early stage (82%), and 76% were treated successfully. Although orbital MZL is almost always limited to the conjunctiva or lacrimal gland, selected cases, particularly when the MZL is symptomatic, may require special attention regardless of stage. The investigators from the University of Miami highlight the issue of radiation dose. They claim that their data show a correlation between inferior progression-free survival (mostly related to nonorbital events) and lower orbital radiation dose. This assertion, if accepted, could potentially influence the RT dose prescribed to the orbit and, in fact, to the eye, with its sensitive components. The dose they imply is higher than what international and national dose guidelines recommend; thus, the quality of the observed dose-outcome data requires further scrutiny.

It appears that in the more than 3 decades of patient inclusion in the study, there has not been a consistent RT dose policy. It is unclear how dose decisions were made, yet, in general, the dose hovered around 30 Gy. The more recent patients (since 2003) have received 30.6 Gy in 1.8 Gy per fraction doses, whereas earlier there was more RT dose variance. The investigators selected 2 dose groups: <30.6 Gy and ≥30.6 Gy. Notably, there was no evidence the RT dose influenced disease control (even in advanced stages) was highly successful. When RT was avoided in 15 patients, nearly one-half had orbital relapse or progression.

Advances in tumor imaging and radiation treatment planning, targeting, and delivery as well as a larger menu of RT systems (including intensity-modulated RT, electrons, and protons) provide an opportunity for more conformal treatment and a possibility of safely reducing the irradiated volume in the orbit in selected cases, particularly when the MZL is limited to the conjunctiva or lacrimal gland. Yet, the standard recommendation for most cases is to still include the full orbital contents unless the disease is confined to the conjunctiva (see figure).

The investigators from the University of Miami report a large retrospective analysis from the University of Miami including 182 patients with primary ocular adnexal mucosa-associated lymphoid tissue (MALT) lymphoma (orbital marginal zone lymphoma [MZL]).1 Most were at an early stage (82%), and 76% were treated successfully with orbital irradiation. Treatment and follow-up data spanned 31 years. This study reaffirms the excellent long-term control of orbital MZL with localized radiation therapy (RT; complete remission [CR] rate >97%); only 3 deaths were attributed to lymphoma, and 2 of those occurred following systemic transformation.

In this issue of Blood, Desai et al report a large retrospective analysis from the University of Miami including 182 patients with primary ocular adnexal mucosa-associated lymphoid tissue (MALT) lymphoma (orbital marginal zone lymphoma [MZL]).1 Most were at an early stage (82%), and 76% were treated successfully with orbital irradiation. Treatment and follow-up data spanned 31 years. This study reaffirms the excellent long-term control of orbital MZL with localized radiation therapy (RT; complete remission [CR] rate >97%); only 3 deaths were attributed to lymphoma, and 2 of those occurred following systemic transformation.

A 64-year-old man with MZL of the left orbit.

(A) A 64-year-old man with MZL of the left orbit. (B) An intensity-modulated RT plan of 24 Gy was designed, but only 4 Gy was delivered with near-CR obtained. (C) Seven months’ follow-up after only 4 Gy with no progression.
Modulating the malignancy of Hox proteins

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