ERK substrate, is essential for the double-strand DNA break repair. The LEX-1 mRNA upregulation is dependent on NF-κB. IR induces enhancement of physical interaction between pERK and DNA-PK, which is mediated by LEX-1, leading to increased phosphorylation of DNA-PK. The conclusion based on their findings is that NHEJ DNA repair induced by TPO is mediated by an IEX-1/DNA-PK/ERK complex in HSCs. These findings provide novel insights into how TPO protects HSCs from DNA damage. Furthermore, manipulation of these molecules may help protect HSCs and megakaryocytes from genotoxic stress when cancer patients are treated with IR or chemotherapy.

How are these signaling molecules in DNA repair related to other signals under TPO stimulation? As previously mentioned, because TPO activates many signaling pathways, other downstream targets may affect DNA repair. ERK and NF-κB may be involved in other biological phenomena (eg, cell cycle or survival) in HSCs. One of the most intriguing issues is whether DNA repair response contributes to HSC self-renewal under the TPO signal. HSCs lacking TPO-MPL signaling exhibit a failure of dormancy of HSCs in vivo.[2,3] Although TPO is a potent stimulator of cell proliferation in vitro, HSCs that lose the TPO signal are actively cycling, associated with decreased negative cell cycle regulators, $p57^{KIP2}$ and $p19^{INK4D}$, which seems contradictory. One possible explanation is that there might be distinct types of HSC proliferation—"self-renewal" and "non–self-renewal"—and that TPO may stimulate the self-renewal type of cell proliferation, whereas it may suppress the non–self-renewal type of proliferation. Because TPO administration is reported to expand HSC numbers in vivo, the self-renewal type of HSC proliferation may be dominant in vivo. Alternatively, loss of the TPO signal reduces the self-renewal type of HSC proliferation but may stimulate the non–self-renewal type of proliferation, leading to failure of quiescence of HSCs. Is genomic integrity dependent on this differential signaling? Another question is whether NF-κB or ERK is involved in TPO-mediated HSC survival. Further dissection of the regulation of the TPO signal on maintenance of genomic integrity and elucidation of collaborative function with other TPO downstream molecules are required to understand the mechanism of HSC maintenance. A whole picture of the harmony of the TPO orchestra would lead to an understanding of the nature of HSCs, including self-renewal, expansion, aging, and tumorigenesis.

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

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● ● ● LYMPHOID NEOPLASIA

Comment on Fish et al, page 530

Not like a wrecking ball: EBV fine-tunes MYC lymphomagenesis

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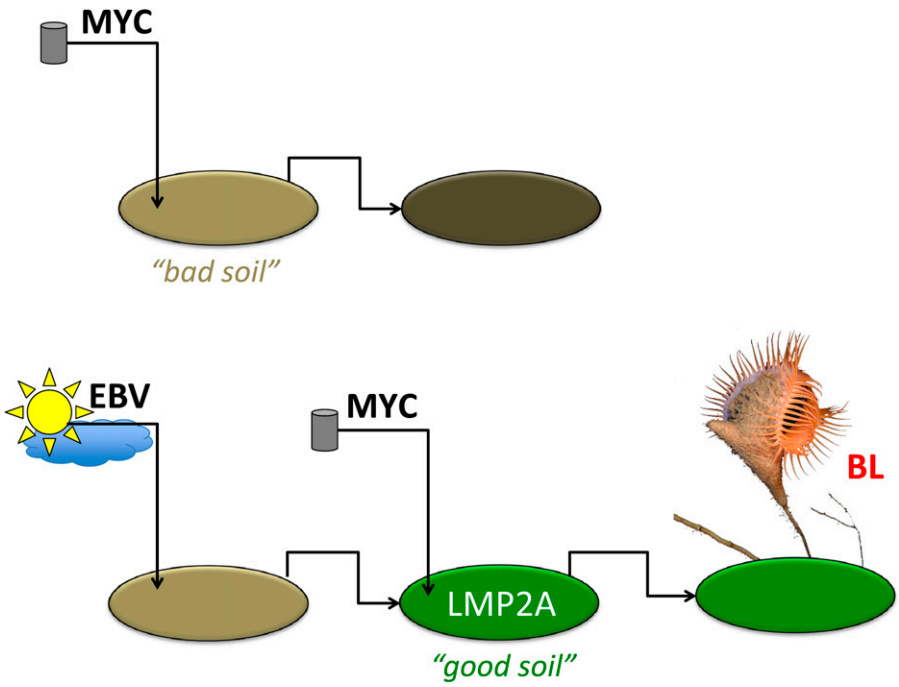
In this issue of *Blood*, Fish et al uncover how Epstein-Barr virus (EBV) enhances MYC-driven B-cell lymphoma by crossing EBV Eμ-EBV latent membrane protein 2A (LMP2A) transgenic mice with immunoglobulin-λ (Igλ)-MYC transgenic mice.[1]

Deregulated MYC function is a central driver in human cancer, cooperating with different cofactors to bring about tumor lineage-specific pathologies. MYC is the defining chromosomal translocation in Burkitt Lymphoma (BL). LMP2A accelerated cell cycle progression in the pretumor B cells by eliminating p27kip1 /CDKN1B post-transcriptionally, thus providing a permissive environment for MYC to act as a fully fledged oncogene. Normally, this would not happen, because deregulated MYC activates p53, which activates p27kip1 (among others), leading to MYC-induced apoptosis. Although p53 is mutated in most solid cancers with deregulated MYC, p53 is seldom mutated in EBV-positive, pediatric BL. P53 mutations tend to arise in EBV-negative, spontaneous BL or following cytotoxic chemotherapy.

In EBV-BL, MYC is not only translocated but also mutated.[2] Mutations in MYC itself or alterations in other pathways such as Bcl-2 are essential to prevent MYC-induced apoptosis. LMP2A cooperates with MYC in transgenic mice at the premalignant stage. Like bcl-2 Eμ transgenic mice, LMP2A Eμ transgenic mice do not form lymphomas in the C57BL/6 background. Another EBV oncogene, LMP1, also modulates the premalignant B-cell state. This predisposes the mice to lymphomas at old age.[3] The EBV LMP1 and LMP2A genes cooperate in reprogramming normal B-cell function, rather than driving lymphoma development. In fact, LMP2A counteracts LMP1's hyperproliferative phenotype,[4] leading to reduced lymphoma incidence in the double transgenic mice. This approximates the human pathology. Neither LMP1 nor LMP2 is consistently expressed in the fully malignant state of human BL; they are part of an EBV multigene driver set that promotes EBV infection in naïve B cells,[5] initial expansion, reprogramming, and germinal center traversal before EBV establishes latency in memory B cells. Teleologically speaking, γ-herpesviruses in general are not intent on



Model of EBV LMP2A and MYC interactions.

causing lymphomas; rather, they want to create a pseudo-germinal center environment that supports lifelong latency.[6,7] The report by Fish et al suggests that this is the very same molecular environment that MYC needs to unfold its oncogenic potential.

What features does a premalignant B cell have to exhibit to become susceptible to MYC-dependent transformation? First, such a cell has to have activated B-cell receptor (BCR)[8] and/or ligand-independent coreceptor signaling such as that generated by LMP1 or CD40. It is no coincidence that hyperendemic BL is associated with childhood EBV infection, as well as constant exposure to an environmental antigen, namely malaria. By contrast, spontaneous EBV-negative BL develops later in life or in immune-compromised patients, and we may surmise that somatic driver mutations substitute for hyperactive BCR signaling. Second, such a cell has to circumvent MYC-induced apoptosis, through bcl-2, through p52 mutation, through specific gain of function mutations in MYC itself, or, as Fish et al now introduce, through reducing the expression and activity of the p27Kip tumor suppressor protein. p27Kip is regulated at the protein level, signaled out by phosphorylation and then targeted for ubiquitin-mediated proteosomal degradation. LMP2A could overcome the tumor protective phenotype of wild-type p27Kip and even that of a p27Kip super-repressor allele (S10A), attesting to the importance of this molecule in premalignant B-cell hyperplasia.

Many types of MYC transgenic mice have been generated over the years. All develop B-cell lymphoma, but few of them are as potent and penetrant as the initial Eμ-MYC transgenic mice. Fish et al used the more accurate, Igλ-MYC transgenic model.[9] Only for their LMP2A × p27Kip experiments did they have to move to C57BL/6 × 129. This allowed them to study the premalignant B-cell environment in the context of intermediate MYC activation. Perhaps it would be informative if one was to try and match MYC and EBV transgene expression even more closely, eg, by targeting MYC and viral protein expression to limited, more mature B-cell developmental stages. This would mimic more closely the preneoplastic state after human EBV infection. Unfortunately, the experience with EBV LMP1 has been sobering in this regard: CD19-CRE–directed tissue-specific expression of LMP1 in mature B cells led to rapid clearance of the LMP1 transgenic cells by the mouse T-cell response.[10]

Fidler has popularized the "seed and soil" hypothesis in tumor metastasis. Metastasis only happens if tumor cells (the "seeds") find a receptive tissue microenvironment (the "soil"). The same concept may help us understand the role of MYC activation (see figure). The MYC translocation/activation event has to happen in the right cellular environment at the right stage of B-cell differentiation to be transforming, otherwise apoptosis ensues. MYC needs a lot of help to function as an oncogene, and γ herpesviruses such as EBV are poised to lend a helping hand.

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

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