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AUTHOR'S VIEWS

BOK-MCL1 transmembrane interactions: a challenging target for cancer therapy

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ABSTRACT

Myeloid cell leukemia 1 (*MCL1*) gene amplification occurs in a wide range of human cancers and protein overexpression associates with malignant cell growth and evasion of apoptosis. We recently reported that disrupting the interaction between the transmembrane domains of MCL1 and BCL-2 related ovarian killer (BOK) induces cell death, thereby suggesting a new target site for anti-tumorigenic strategies.

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Members of the B-cell lymphoma 2 (BCL-2) protein family play crucial roles in controlling mitochondrial membrane permeability and modulating apoptosis. This family is divided into pro-apoptotic proteins that permeabilize the mitochondrial outer membrane, anti-apoptotic members that inhibit this process, and a BCL-2 containing *only* the BCL-2-Homology-3 (BH3) domain (BH3-only) subset of proteins that directly or indirectly activate the pro-apoptotic proteins. All BCL-2 proteins share from one to four BH conserved domains, with the BH3 domain being crucial for the homo- and hetero- interactions among BCL-2 family proteins and the modulation of apoptotic activity.

Tumor types that include hematological malignancies, breast, central nervous system, colon, lung, ovarian, prostate, and renal cancers, and melanoma[1] overexpress *MCL1*, an anti-apoptotic/pro-survival member of the BCL-2 protein family.[2] Studies have also identified *MCL1* overexpression as a mechanism of resistance to both conventional and targeted therapies in many tumors,[3] thereby contributing to the permanence of residual disease.

MCL1 interacts with the pro-apoptotic Bim, Puma, and truncated Bid *BH3-only* proteins and the pro-apoptotic effector Bak[4] through the BH3 domain. The hydrophobic groove present on the surface of MCL1 and other anti-apoptotic members accommodates the BH3 region of various pro-apoptotic proteins to avoid the induction of cell death. The disruption of protein-protein interactions between pro- and anti-apoptotic BCL-2 family members represents a significant challenge in the field of drug discovery. Small molecules that mimic BH3 binding (BH3 mimetics) block heterodimerization with pro-apoptotic members of the BCL-2 family, thereby increasing the number of free pro-apoptotic effectors and inducing apoptosis in cancer cells in which the overexpression of anti-apoptotic proteins provides a survival advantage and drug resistance. Years of intensive research have fostered the development of specific BH3 mimetics targeting MCL1 that induce cell death in tumors. Said mimetics are now under evaluation in clinical trials with the hope of soon reaching widespread use; however, the rapid appearance of mutations in the BH3 binding groove and the downregulation of BAX and BAK expression in specific tumors have highlighted a need for alternative approaches.[5]

We recently described a new interaction site between the transmembrane region of MCL1 (Mcl-1 TMD) and the pro-apoptotic effector BOK. Our study demonstrated that the introduction of the Mcl-1 TMD induces cell death, which is counteracted by full-length MCL1 overexpression but not by other anti-apoptotic BCL-2 proteins. In this case, cell death is dependent on BOK protein but independent of BAX and BAK.[6] These observations suggest the existence of transmembrane interaction specificity between different members of the BCL-2 protein family and highlight the disruption of Mcl-1 and Bok TMD interactions as an exciting new strategy to trigger apoptosis (Figure 1). The development of novel drugs directed to these interaction regions may provide a new means to combat those tumors that have lost BAX but retain MCL1 and BOK expression.

Can we now discover small molecules that act at transmembrane regions and display suitable pharmacological profiles to behave like drugs? Thanks to recent technological advances that have improved the structural resolution of membrane proteins, some examples of drugs whose mechanism of action involves interaction within the context of the membrane have been reported. As an example, ivacaftor was approved in 2012 by the Food and Drug Administration (FDA) as an enhancer molecule to increase the ion flux of the mutant cystic fibrosis transmembrane conductance regulator (CFTR) protein in cystic fibrosis patients. A recent study has demonstrated that ivacaftor binds to CFTR at the protein-lipid interface, docking into a cleft formed by the transmembrane regions of the CFTR channel,[7] thereby illustrating the feasibility of drugging transmembrane regions.

However, many pertinent questions remain unanswered. While we have observed the co-immunoprecipitation of MCL1 and BOK, this interaction remains a contentious point and it has been widely discussed in the literature. The classification of BOK as an apoptotic executor together with BAX and

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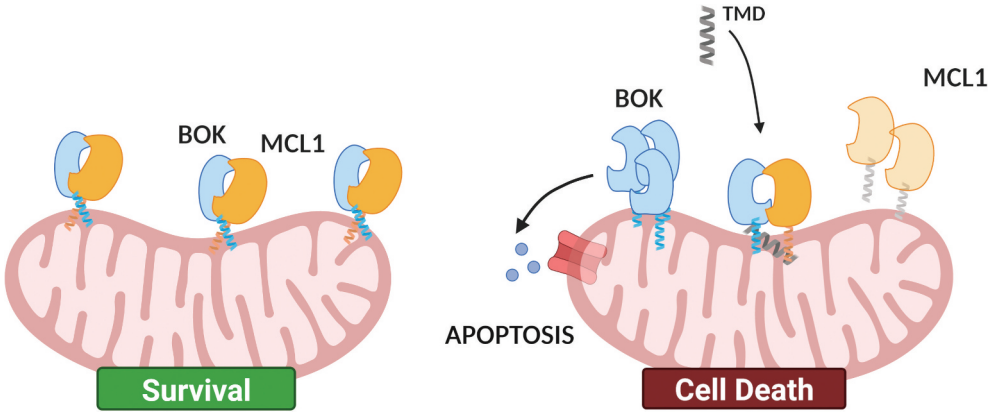

Figure 1. Interaction of MCL1 and BOK through transmembrane domains provides a new point of intervention to induce apoptosis in tumor cells. Disruption of the interaction between Mcl-1 and Bok TMD by a TMD or potential small molecule might release pro-apoptotic BOK inducing apoptosis. TMD refers to transmembrane domain. Created with BioRender.com.

BAK has also prompted significant discussion, with some reports demonstrating pro-apoptotic activity while others showing pro-survival effects.[8] In our hands, BOK behaves as a pro-apoptotic protein, and co-expression of both Mcl-1 and Bok TMDs increases the formation of hetero-oligomers in the mitochondrial membrane, promoting an increase in the number of endoplasmic reticulum-mitochondrial associated membranes (MAMs). Our results suggest that MCL1 and BOK transmembrane interactions could intervene in the modulation of MAMs and control the stability of both proteins; however, understanding the functional relevance of these processes will require further research. Interestingly, crosstalk between MCL1 and BOK proteins has been previously described; for example, the co-regulation of protein stability has been observed in preeclampsia models,[9] while BOK deficiency in neurons decreases survival due to MCL1 degradation.[10]

The evidence that MCL1 and BOK interactions occur through transmembrane regions opens new avenues of study that may explain many currently noted discrepancies and facilitate the development of new antitumor drugs.

Disclosure of potential conflicts of interest

The authors declare no competing interest.

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References

1. Beroukhim R, Mermel CH, Porter D, Wei G, Raychaudhuri S, Donovan J, Barretina J, Boehm JS, Dobson J, Urashima M, et al. The landscape of somatic copy-number alteration across human cancers. Nature. 2010;463(7283):899–905.
2. Kozopas KM, Yang T, Buchan HL, Zhou P, Craig RW. MCL1, a gene expressed in programmed myeloid cell differentiation, has sequence similarity to BCL2. Proc Natl Acad Sci U S A. 1993;90(8):3516–3520. doi:10.1073/pnas.90.8.3516.
3. Wood KC. Overcoming MCL-1-driven adaptive resistance to targeted therapies. Nat Commun. 2020;11(1):531. doi:10.1038/s41467-020-14392-z.
4. Zhai D, Jin C, Huang Z, Satterthwait AC, Reed JC. Differential regulation of Bax and Bak by anti-apoptotic Bcl-2 family proteins Bcl-B and Mcl-1. J Biol Chem. 2008;283(15):9580–9586. doi:10.1074/jbc.M708426200.
5. Hird AW, Tron AE. Recent advances in the development of Mcl-1 inhibitors for cancer therapy. Pharmacol Ther. 2019;198:59–67. doi:10.1016/j.pharmthera.2019.02.007.
6. Lucendo E, Sancho M, Lolicato F, Javanainen M, Kulig W, Leiva D, Duart G, Andreu-Fernandez V, Mingarro I, Orzaez M. Mcl-1 and Bok transmembrane domains: unexpected players in the modulation of apoptosis. Proc Natl Acad Sci U S A. 2020;117:27980–27988.
7. Liu F, Zhang Z, Levit A, Levring J, Touhara KK, Shoichet BK, Chen J. Structural identification of a hotspot on CFTR for potentiation. Science. 2019;364(6446):1184–1188. doi:10.1126/science.aaw7611.
8. Llambi F, Wang YM, Victor B, Yang M, Schneider DM, Gingras S, Parsons MJ, Zheng JH, Brown SA, Pelletier S, et al. BOK is a non-canonical BCL-2 family effector of apoptosis regulated by ER-associated degradation. Cell. 2016;165(2):421–433. doi:10.1016/j.cell.2016.02.026.
9. Kalkat M, Garcia J, Ebrahimi J, Melland-Smith M, Todros T, Post M, Caniggia I. Placental autophagy regulation by the BOK-MCL1 rheostat. Autophagy. 2013;9(12):2140–2153. doi:10.4161/auto.26452.
10. D'Orsi B, Engel T, Pfeiffer S, Nandi S, Kaufmann T, Henshall DC, Prehn JH. Bok is not pro-apoptotic but suppresses poly ADP-ribose polymerase-dependent cell death pathways and protects against excitotoxic and seizure-induced neuronal injury. J Neurosci. 2016;36(16):4564–4578. doi:10.1523/JNEUROSCI.3780-15.2016.