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Editorial

Seeking a MCL-1 inhibitor

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The development of small molecule drugs that mimic the actions of BH3-only proteins is one of the major triumphs of many years of apoptosis research. They are built upon a platform of an understanding the mechanisms of action of BCL-2 family proteins, and particularly a structural appreciation of the interactions between BCL-2 family proteins. These BH3-mimetic drugs function by slotting into the hydrophobic groove on the surface of BCL-2 and BCL-X_L, so blocking their capacity to inhibit apoptosis.[1] They are a leading example, along with the Smac/DIABLO inhibitor drugs, of structure-based design of small molecule drugs. Both classes of drug are negotiating their way through early phase clinical trials with some promise.

The BH3-mimetic drugs, ABT-263 and ABT-199, primarily inhibit antiapoptotic BCL-2 proteins BCL-2 and BCL-X_L, and their therapeutic potentials are being tested in malignancies characterized by overexpression of these proteins, such as non-Hodgkin's lymphoma and CLL. Additionally, these drugs may increase the sensitivity of tumors such as small-cell lung cancer to conventional chemotherapeutics, by removing blocks on the activation of apoptosis pathways. Their specificity of action is an important design feature, as these drugs are modeled on the BH3 domain of BAD, which binds to BCL-2 and BCL-X_L but not to MCL-1.[2] This specificity limits the potential side effects and increases utility by selectively killing malignant cells dependent on the overexpression of BCL-2.

However, it is also clear that MCL-1 is a very attractive drug target, and a small molecule that specifically inhibited MCL-1 would have significant therapeutic potential in the many malignancies in which it is overexpressed.[3] In AML, for example, it is clear that MCL-1 expression is required for AML to develop and for disease to manifest in secondary transplants.[4] The only way around this block was for cells to silence the machinery that permitted MCL-1 deletion. Moreover, because expression of MCL-1 is an important mechanism of resistance to the BH3-mimetic drugs,[2] expanding the range of BCL-2 inhibitors to include at least some activity against MCL-1 would potentially be a significant advantage.

Specific targeting MCL-1 has proved a hard task. In this issue of *Cell Death and Differentiation*, Varadarajan and colleagues[5] have put to the test a range of lead compounds that may have some claim to MCL-1 specificity. In designing their experiments, they have considered just what might characterize a specific MCL-1 inhibitor. Such a drug should not kill cells lacking both the pro-apoptotic proteins Bax and BAK, as these molecules are absolutely required for apoptosis pathways inhibited by MCL-1 and BCL-2. MCL-1 has some binding specificity for BAK,[6] so one might expect a MCL-1 blocker to require BAK for full activity. Further, a MCL-1-specific inhibitor should induce apoptosis in MCL-1-dependent cells, and significantly less death in cells protected by other antiapoptotic BCL-2 family members.

Having set these criteria, Varadarajan *et al* have applied them to a range of molecules and cancer cell lines. Although none of the compounds satisfies all the benchmarks, one molecule, TW-37, manages to meet at least some, suggesting it has some promise as a lead molecule from which to build greater specificity for MCL-1 (Figure 1). TW-37 is a non-peptidic small molecule, which in preclinical studies could block BAK-MCL-1 binding.[7] Varadarajan *et al* show that TW-37 does not kill cells with a deficient intrinsic apoptosis pathway, and has some specific requirement for BAK to induce apoptosis. TW-37 also induces apoptosis in at least two MCL-1-dependent systems, IL-3-dependent cells and H23 cells. Further, TW-37 killing is blocked by BCL-2 and BCL-X_L, suggesting this drug is less effective at inhibiting these molecules than it is at inhibiting MCL-1.

One intriguing finding is that at least some of the activity of TW-37 results from the induction of expression, by an unknown mechanism, of the BH3-only protein NOXA. This was particularly noted in the non-small-cell lung carcinoma H1299 cells, and when TW-37 was used in combination with ABT-737. Noxa has specificity for MCL-1 binding, and so these results suggest the possibility that TW-37 targets MCL-1 through an indirect mechanism. This may not be so bad if, in the end, the result is inhibition of MCL-1 and the death of the cancer cell. TW-37 may thus act indirectly to alter the 'primed for death' status of the malignant cell.[8] On the other hand, the finding that Noxa is required for the TW-37 and ABT-737 combination to kill H1299 cells highlights at least one potential escape route to drug resistance. The crystal structure of MCL-1 bound to TW-37, or more specific derivatives as they are developed, would clarify many questions regarding function and specificity.

An important concern surrounding a potential MCL-1 inhibitor drug is the potential for serious side effects. MCL-1 deletion is embryonic lethal,[9] and MCL-1 is required for, among other things, normal haematopoiesis.[10] One might imagine that using a MCL-1 inhibitor to kill malignant cells

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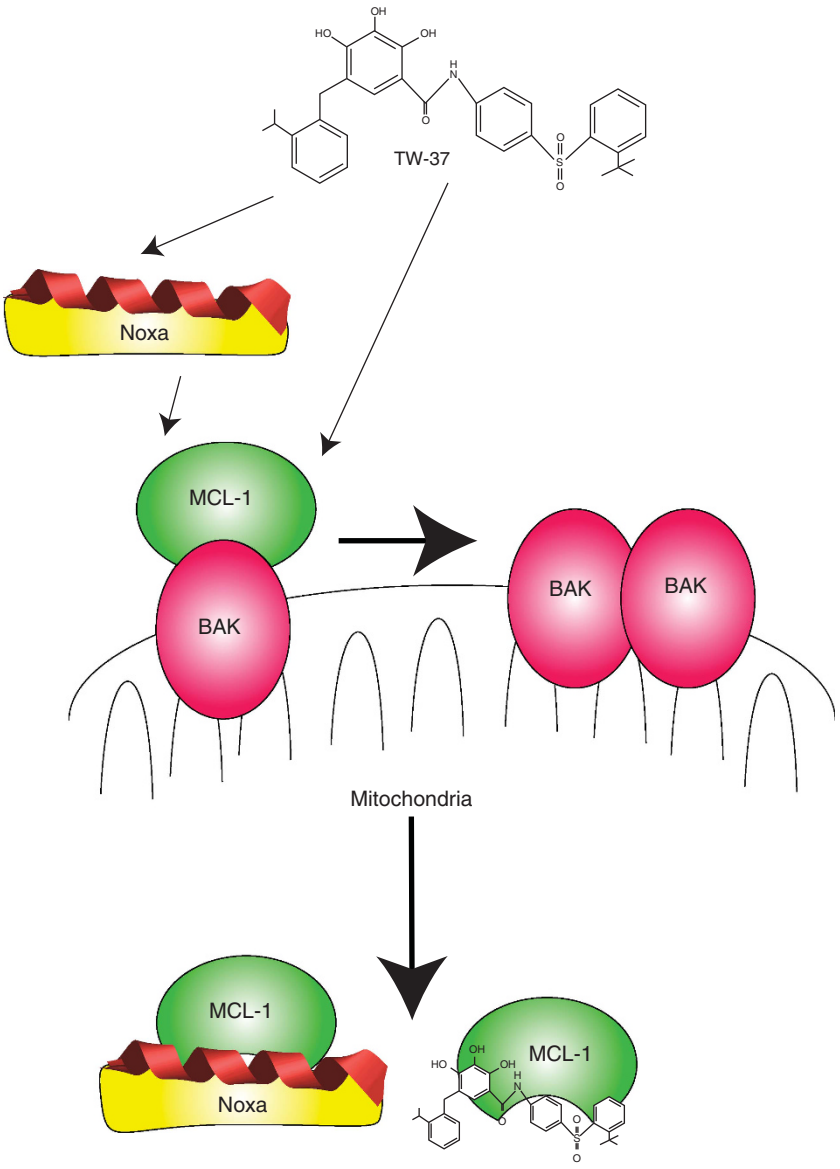

Figure 1 TW-37 has MCL-1 specific activity to induce apoptosis. The small-molecule TW-37 may directly inhibit MCL-1 and disrupt MCL-1 binding to BAK (or to BAX). TW-37 also induces transcription and expression of the BH3-only protein NOXA, which binds via its BH3-domain (represented by the helix) to MCL-1. The disruption of MCL-1-BAK binding permits BAK to oligomerise (which may or may not be driven by another BH3-only protein) and commits cells to undergo apoptosis. MCL-1 itself is targeted for proteasomal degradation

might take out a large number of innocent bystanders, including haematopoietic progenitor cells. If this were the case, a MCL-1 inhibitor, although fulfilling some of the promise of targeted therapies, could result in toxicities not dissimilar to conventional chemotherapeutic drugs. The hope is that malignant cells are uniquely dependent on MCL-1, permitting a therapeutic window for an inhibitor to kill many more malignant cells than normal cells. Alternatively, as pointed out by Varadarajan and colleagues, an inhibitor of BCL-2-like molecules with at least some anti-MCL-1 activity may be sufficient to gain a therapeutic advantage for a minimal cost in toxicity. Thus, the therapeutic potential of inhibiting MCL-1 is such that concerns about potential side effects should not stand in the way of developing a clinical compound. The assessment of side effects will be more meaningful when there is true inhibitor to test in preclinical models. Perhaps TW-37 is the basis on which just such a drug can be built.

Conflict of Interest

The authors declare no conflict of interest.

1. Oltersdorf T *et al. Nature* 2005; **435**: 677–681.
2. van Delft MF *et al. Cancer Cell* 2006; **10**: 389–399.
3. Beroukhim R *et al. Nature* 2010; **463**: 899–905.
4. Glaser SP *et al. Genes Dev* 2012; **26**: 120–125.
5. Varadarajan S *et al. Cell Death Differ* 2013; **20**: 1475–1484.
6. Willis SN *et al. Genes Dev* 2005; **19**: 1294–1305.
7. Mohammad RM *et al. Clin Cancer Res* 2007; **13**: 2226–2235.
8. Certo M *et al. Cancer Cell* 2006; **9**: 351–365.
9. Rinkenberger JL. *Genes Dev* 2000; **14**: 23–27.
10. Opferman JT *et al. Science* 2005; **307**: 1101–1104.