

Key References

1. Muchmore S.W., et al. (1996) X-ray and NMR structure of human Bcl-X_L, an inhibitor of programmed cell death. *Nature*, **381**, 335-341. Sattler, M., et al. (1997)
2. Structure of Bcl-X_L/Bak peptide complex: Recognition between regulators of apoptosis. *Science*, **275**, 983-986. Shuker, S.B., et al. (1996) Discovering high-affinity ligands for proteins: SAR by NMR. *Science* **274**, 1531-1534.
3. Hajduk P.J., et al. (1997) Discovering high-affinity ligands for proteins. *Science* **278**, 497-499.
4. Petros, A.M., et al. (2006) Discovery of a potent inhibitor of the antiapoptotic protein Bcl-X_L from NMR and parallel synthesis. *J. Med. Chem.* **49**, 656-663.
5. Wendt, M.D., et al. (2006) Discovery and structure-activity relationship of Bcl-X_L antagonists with chemopotential activity *in vitro* and *in vivo*. *J. Med. Chem.* **49**, 1165-1181.
6. Petros, A.M., et al. (2001) Solution structure of the anti-apoptotic protein Bcl-2. *Proceed. Natl. Acad. Sci., USA*, **98**, 3012-3017.
7. Oltersdorf, T., et al. (2005) An Inhibitor of Bcl-2 family proteins induces regression of solid tumors. *Nature* **435**, 677-681.
8. Tahir, S.K., et al. (2006) Influence of Bcl-2 family members on the cellular response of small cell lung cancer cell lines to ABT-737. *Cancer Res.* **67**, 1176-1183 (2007).
9. Lin, X., et al. (2006) Seed analysis of off-target siRNAs reveals an essential role of Mcl-1 resistance to the small molecule Bcl-2/Bcl-X_L inhibitor ABT-737. *Oncogene* **26**, 3972-3979 (2007).



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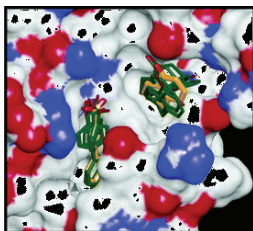
Dr. Steve Fesik
VP, Abbott

Discovery of Bcl-2 Family Inhibitors for the Treatment of Cancer



Date: 07/11/2007
(Wednesday)
Time: 12:00 Noon

Venue: COMRB auditorium
Following seminar, there will
be a reception in the lobby.
All are welcome!!!!!!



Bcl-x_L Discovery of a Potent Inhibitor of the Antiapoptotic Protein Bcl-x_L from NMR and its Parallel Synthesis
J. Med. Chem.; 2006; 49(2) pp 656 - 663.

Apoptosis is a natural process for eliminating unwanted cells that is deregulated in many cancers. One of the major mechanisms for circumventing programmed cell death is by the overexpression of anti-apoptotic proteins. The **Bcl-2** family of proteins regulate programmed cell death and can either inhibit or promote apoptosis. Anti-apoptotic Bcl-2 proteins have been shown to contribute to tumor initiation, tumor progression, and resistance to therapy. Thus, members of the Bcl-2 family of proteins that inhibit programmed cell death such as Bcl-2 and Bcl-XL represent promising targets for the treatment of cancer.

As a first step in designing small molecule inhibitors of Bcl-2 and Bcl-XL that bind tightly to these proteins, we determined the three-dimensional structure of Bcl-XL alone (1) and when complexed to a peptide derived from the pro-apoptotic protein Bak (2). These structures gave us

XL binding pocket. Although we were targeting protein/protein interaction, the structures revealed that the binding pocket consisted of a hydrophobic groove that might be amenable to small molecule drug discovery. In order to obtain a small molecule that binds to Bcl-XL and other anti-apoptotic members of the Bcl-2 family, we applied an NMR-based method for discovering lead compounds called **SAR by NMR** (3,4). Using this approach, we screened a library of small molecules by NMR and identified a molecule (4'-fluoro-biphenyl-4-carboxylic acid) that bound to Bcl-XL with a dissociation constant of 0.3 mM and 5,6,7,8-tetrahydro-naphthalen-1-ol that bound (K_d=4.3 mM) to a second nearby site (5). Interestingly, these compounds identified from the NMR-based screen bind to the same sites on Bcl-XL where the key Bak peptide residues bind. Based on structural information that indicated how the two molecules bind to Bcl-XL, they were linked together to form an initial lead that was subsequently improved by preparing a library of acylsulfonamides. Although the binding affinity was improved, the lead compounds were found to bind tightly to human serum albumin (HSA) which severely attenuated their activity when measured in the presence of serum. To overcome this hurdle, a structure-based approach was utilized. Structures were compared of the compounds when bound to Bcl-XL and when bound to domain III of HSA. On the basis of the structures of the complexes, compounds were designed that retained binding to Bcl-XL but reduced binding to HSA (6). Finally, to improve binding to Bcl-2, a lipophilic group was added to access a hydrophobic pocket identified in the structure of Bcl-2 (7). These efforts resulted in a compound, **ABT-737**, that potently (K_i ≤ 1nM) inhibits Bcl-2, Bcl-XL, and Bcl-w (8).

In cell-based assays, ABT-737 was shown to

be synergistic with multiple chemotherapeutic agents and radiation for killing a wide variety of tumor cells. In addition, ABT-737 was active as a single agent in tumor cells derived from small cell lung carcinomas, lymphomas and leukemias. In tumor xenograft mouse models, ABT-737 caused the regression of SCLC and exhibited a survival benefit in lymphoma models (8).

Recently, we have conducted preclinical experiments to further understand the biological activities of our Bcl-2 family inhibitors that may provide information to aid our clinical trials. We found that cancer cells that are sensitive to ABT-737 overexpress Bcl-2 and/or Bcl-XL and have low levels of Mcl-1, an antiapoptotic protein that is not inhibited by ABT-737. In addition, those cells that are resistant to ABT-737 have high levels of Mcl-1 (9). When Mcl-1 levels are reduced by siRNA or by the addition of chemotherapeutic agents, the resistant cells become sensitive to ABT-737 treatment. These studies were extended to include other types of cancers. For many different types of cancer, if Mcl-1 levels are low or reduced, these cells are sensitive to ABT-737 treatment (10). These results suggest that our Bcl-2 family inhibitors may be useful as a single agent for the treatment of cancers that contain a low level of Mcl-1 and many other cancers when given in combination with chemotherapy and radiation when the expression of Mcl-1 is reduced. Currently, we are testing an orally active Bcl-2 family inhibitor that is structurally similar to ABT-737 in phase I

