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# Apoptosis-targeted therapies for cancer

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## Introduction

Defects in programmed cell death (apoptosis) mechanisms play important roles in tumor pathogenesis, allowing neoplastic cells to survive beyond their normally intended lifespans, subverting the need for exogenous survival factors, providing protection from hypoxia and oxidative stress as tumor mass expands, and allowing time for accumulative genetic alterations that deregulate cell proliferation, interfere with differentiation, promote angiogenesis, and increase cell motility and invasiveness during tumor progression (Reed, 1999). In fact, apoptosis defects are recognized as an important complement to protooncogene activation, as many deregulated oncoproteins that drive cell division also trigger apoptosis (e.g., Myc, E1a, Cyclin-D1) (Green and Evan, 2002). Similarly, defects in DNA repair and chromosome segregation normally trigger cell suicide as a defense mechanism for eradicating genetically unstable cells, and thus apoptosis defects permit survival of genetically unstable cells, providing opportunities for selection of progressively aggressive clones (Ionov et al., 2000). Apoptosis defects also facilitate metastasis by allowing epithelial cells to survive in a suspended state, without attachment to extracellular matrix (Frisch and Screaton, 2001). They also promote resistance to the immune system, inasmuch as many of the weapons cytolytic T cells (CTLs) and natural killer (NK) cells use for attacking tumors depend on integrity of the apoptosis machinery (Tschopp et al., 1999). Finally, cancer-associated defects in apoptosis play a role in chemoresistance and radioresistance, increasing the threshold for cell death and thereby requiring higher doses for tumor killing (Makin and Hickman, 2000). Thus, defective apoptosis regulation is a fundamental aspect of the biology of cancer.

When it comes to the successful eradication of cancer cells by nonsurgical means, ultimately, all roads lead to apoptosis. Essentially all cytotoxic anticancer drugs currently in clinical use, when they work, induce apoptosis of malignant cells. While microtubule binding drugs, DNA-damaging agents, and nucleosides are important weapons in the treatment of cancer, a new class of targeted therapeutics may soon be forthcoming based on strategies that have emerged from a deeper understanding of the molecular mechanisms that underlie the phenomenon of apoptosis.

Apoptosis is caused by proteases known as "caspases," for *c*ysteine *asp*artyl-specific prote*ases* (Cryns and Yuan, 1999; Thornberry and Lazebnik, 1998). Caspases constitute a family of intracellular cysteine proteases (n = 11 in humans), which collaborate in proteolytic cascades where caspases activate themselves and each other. Within these proteolytic cascades, caspases can be positioned as either upstream "initiators" or downstream "effectors" of apoptosis. Several pathways for activating caspases exist (Figure 1). First, of the ~30 members of the tumor necrosis factor (TNF)-family receptors, eight contain a so-called death domain (DD) in their cytosolic tail (Locksley et al., 2001). Several of these DD-containing TNF-family receptors use caspase activation as a signaling mechanism, including TNFR1/CD120a, Fas/APO1/CD95, DR3/Apo2/Weasle, DR4/TrailR1, DR5/TrailR2, and DR6. Ligation of these receptors at the cell surface results in the recruitment of several intracellular proteins, including certain procaspases, to the cytosolic domains of these receptors, forming a "death-inducing signaling complex" (DISC) that triggers caspase activation—constituting the so-called "extrinsic" pathway for apoptosis (Wallach et al., 1999). The specific caspases summoned to the DISC are caspase-8 and, in some cases, caspase-10. These caspases contain so-called death effector domains (DEDs) in their N-terminal prodomains that bind to a corresponding DED in the adaptor protein, FADD, thus linking them to the TNF-family death receptor complexes. Second, mitochondria induce apoptosis by releasing cytochrome-c (cyt-c) into the cytosol, which then causes assembly of a multiprotein caspase-activating complex, referred to as the "apoptosome" (Green and Reed, 1998). The central component of the apoptosome is Apaf1, a caspase-activating protein that oligomerizes upon binding cyt-c, then binds procaspase-9 via interaction with its caspase recruitment domain (CARD). The mitochondrial pathway for apoptosis, also known as the "intrinsic pathway," is activated by myriad stimuli, including growth factor deprivation, oxidants, $Ca^{2+}$ overload, oncogene activation, DNA-damaging agents, and microtubule-attacking drugs. In addition to cyt-c, mitochondria also release several other proteins, including endonuclease G, AIF (a death-modulating flavoprotein), and IAP antagonists SMAC (DIABLO) and OMI (HtrA2) (see below), some of which may promote caspase-independent (nonapoptotic) cell death (Kroemer and Reed, 2000). A third pathway for apoptosis induction is specific to CTL and NK cells, which spray apoptosis-inducing protease, granzyme B (GraB), onto target cells. GraB then piggybacks into cells via mannose-6-phosphate receptors (IGFR2) and enters effective cellular compartments via perforin channels (Motyka et al., 2000). GraB is a serine protease, but similar to the caspases, it cleaves substrates at Asp residues, including several caspases and some caspase substrates. Fourth, a caspase activation pathway linked to endoplasmic reticulum (ER)/Golgi stress has been proposed (Cryns and Yuan, 1999), though many details are lacking. Finally, a nuclear pathway for apoptosis regulation may exist which centers on discrete nuclear organelles, called Pml oncogenic domains (PODs) or nuclear bodies (NBs). Targeted ablation of the *pml* gene in mice results in general resistance to apoptosis through unknown mechanisms. Several proteins that can promote apoptosis have been localized to PODs, including Daax, Zip kinase, and Par4, and defects in assembly of these nuclear structures are documented in cancers (Salomoni and Pandolfi, 2002). How PODs are linked to caspase activation pathways is unknown.

Several endogenous antagonists of the caspase-activation pathways have been discovered and examples of dysregulation of their expression or function in cancers have been obtained. These apoptosis suppressors have become targets for drug dis-

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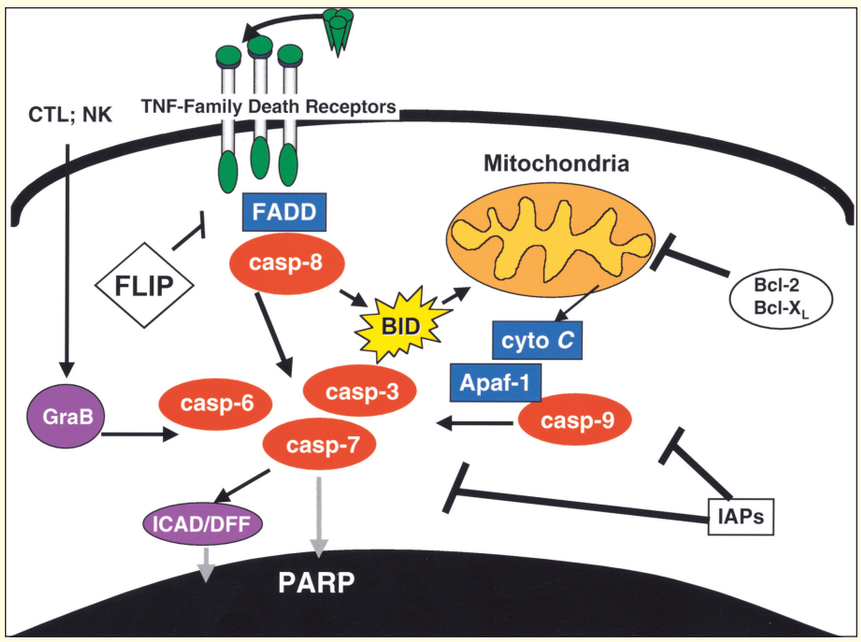


**Figure 1.** Apoptosis pathways

Some of the better defined apoptosis pathways are depicted. FLIP, Bcl-2/Bcl-XL, and IAPs suppress specific steps in the extrinsic, intrinsic, and convergence pathways, respectively.

covery, with the idea of abrogating their cytoprotective functions and restoring apoptosis sensitivity to tumor cells. Alternatively, one can envision directly activating agonists of apoptosis in cancer cells. A summary of some of the more promising strategies for modulating the activity of apoptosis genes and proteins for cancer therapy is provided below, according to the apoptosis pathways they affect.

**Intrinsic pathway**

Bcl-2 family proteins (n = 24 in humans) are central regulators of the intrinsic pathway, which either suppress or promote changes in mitochondrial membrane permeability required for release of cyt-c and other apoptogenic proteins (Green and Reed, 1998; Gross et al., 1999). Overexpression of antiapoptotic Bcl-2 or Bcl-$X_L$ probably occurs in more than half of all cancers (Amundson et al., 2000), rendering tumor cells resistant to myriad apoptotic stimuli, including most cytotoxic anticancer drugs. Attempts to overcome the cytoprotective effects of Bcl-2 and Bcl-$X_L$ in cancer include three strategies: (1) shutting off gene transcription, (2) inducing mRNA degradation with antisense oligonucleotides, and (3) directly attacking the proteins with small-molecule drugs.

Though not components of the "core" apoptosis machinery, some members of the steroid/retinoid superfamily of ligand-activated transcription factors (SRTFs) represent potentially "drugable" modulators of *BCL-2* and *BCL-X$_L$* gene transcription. In the mammary gland, for example, expression of *BCL-2* is estrogen-dependent. Consequently, antiestrogens, such as tamoxifen, inhibit endogenous *BCL-2* expression in breast cancer cell lines, promoting sensitivity to cytotoxic anticancer drugs such as doxorubicin. Enforced expression of Bcl-2 protein from plasmid vectors, in contrast, abrogates sensitivity to the apoptosis-promoting effects of antiestrogens in breast cancer lines, while antisense *BCL-2* prevents estrogen-mediated apoptosis suppression, thus establishing a direct functional connection between ER, Bcl-2, and suppression of apoptosis (Teixeira et al., 1995). In addition to antiestrogens in estrogen receptor (ER)-positive breast cancers, expression of Bcl-2 or Bcl-$X_L$ can be downregulated in specific types of cancer and leukemia cells by small-molecule drugs that modulate the activity of retinoic acid receptors (RAR), retinoid X receptors (RXR), PPARγ, vitamin D receptors (VDR), and certain other members of the SRTF superfamily. RAR and RXR ligands are already approved for treatment of some types of leukemia and lymphoma, and are in advanced clinical testing for solid tumors. PPARγ modulators have demonstrated antitumor activity in xenograft models of breast and prostate cancer (Kubota et al., 1998), sometimes displaying synergy with retinoids, probably due in part to the fact that PPARγ binds DNA as a heterodimer with RXR. Interestingly, PPARγ agonist troglitazone can lower serum PSA in men with advanced prostate cancer, though it is unknown whether this reflects a proapoptotic mechanism in vivo. Compounds that inhibit histone deactylases (HDACs), transcriptional repressors that interact with retinoid receptors and other transcription factors, can also favorably modulate expression of Bcl-2 or Bcl-$X_L$ in some tumor lines (Reed, 2002). These observations portend opportunities for exploiting endogenous transcriptional pathways for suppressing expression of antiapoptotic *BCL-2*-family genes in cancer, with the idea of employing them as chemo- or radiosensitizers, rather than relying on their antitumor activity as single agents. However, much remains to be learned about the genetic characteristics of cancer cells that dictate responsiveness versus resistance to SRTF-ligands, as well as the complex pharmacological interplay between this class of agents and conventional cytotoxic drugs.

Nuclease-resistant (phosphorothioate) antisense oligonucleotides targeting the *BCL-2* mRNA have advanced to Phase III clinical trials for melanoma, myeloma, CLL, and AML, with Phase II activity underway for a variety of solid tumors (Table 1). Because *BCL-2* antisense enhances sensitivity to cytotoxic anticancer drugs in vitro and in xenograft models, most clinical trials combine the antisense agent with conventional chemotherapy (Chi et al., 2001). Uncontrolled Phase II data suggest a benefit to adding *BCL-2* antisense to conventional therapy, but definitive proof awaits the Phase III results. Also at issue is whether phosphorothioate-based oligonucleotides work entirely through antisense mechanisms.

Strategies for attacking the Bcl-2 and Bcl-$X_L$ proteins using small-molecule drugs have emerged from an understanding of how our own cells keep these antiapoptotic proteins under control. In this regard, a large family of endogenous antagonists of Bcl-2 and Bcl-$X_L$ has been revealed (so-called "BH3-only proteins"), possessing a conserved amphipathic α-helical domain (BH3) that binds a hydrophobic crevice on the surface of Bcl-2

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**Table 1.** Summary of drug discovery activity targeting core components of the apoptosis machinery

| Agent/strategy | Group/company | Current status |
|---|---|---|
| A. Bcl-2/Bcl-XL antagonists | | |
| Bcl-2 antisense (G3139) | GENTA/Aventis | Phase II and III |
| Bcl-2/Bcl-XL small-molecule antagonists | IDUN/Abbott | Preclinical |
| Bcl-2 small-molecule antagonist (GX-01) | Gemin-X | Preclinical |
| Bcl-2 small-molecule antagonist | Structural Bioinformatics | Preclinical |
| Bcl-2 small-molecule antagonists | Thomas Jefferson University; Harvard University Washington University | Preclinical |
| Bcl-2 inhibitory natural product (Tetrocarcin-A) | Kyowa-Hakko Kogyo | Preclinical |
| Bax adenovirus | MD Anderson Cancer Center; University of South Carolina | Phase I |
| BH3 mimetics (peptides) | Biomeasure | Preclinical |
| B. TRAIL-R agonists | | |
| Anti-DR4 monoclonal agonist | Human Genome Sciences (Cambridge Antibody) | Phase I |
| Anti-DR5 monoclonal agonist | Sankyo | Preclinical |
| TRAIL extracellular domain | Genentech-Immunex (Amgen) | Preclinical |
| C. IAP-antagonists | | |
| SMAC-mimicking small molecules | IDUN/Abbott | Preclinical |
| Survivin antisense | ISIS/Lilly | Preclinical |
| XIAP antisense | Agera | Preclinical |

Some of the current activity toward discovery and development of apoptosis-based therapies for cancer is summarized for (A) Bcl-2/Bcl-XL antagonists, (B) TRAIL-Receptor agonists, and (C) IAP-antagonists. Data are based on a combination of published reports, patent applications, meeting reports, press releases, and internet-derived information.

and Bcl-$X_L$ (Huang and Strasser, 2000). Proof-of-concept experiments using synthetic BH3 peptides have confirmed the validity of strategies based on generating small-molecule drugs that occupy the BH3 binding site on Bcl-2 or Bcl-$X_L$, abrogating their cytoprotective functions (Letai et al., 2002). Indeed, several groups have presented preclinical data regarding BH3-mimicking compounds (Table 1A). The optimal structure-activity relation (SAR) profile of these compounds remains to be determined, with respect to the issue of selective versus broad-spectrum activity against the various antiapoptotic members of the Bcl-2-family (Bcl-2, Bcl-$X_L$, Mcl-1, Bcl-W, Bfl-1, Bcl-B), in terms of balancing antitumor efficacy against side effects.

Other strategies for countering Bcl-2 and Bcl-$X_L$ in cancer include inducing expression of opposing proapoptotic family members such as Bax with p53 adenovirus (in Phase III trials) or with Mda7 (IL-24) adenovirus gene therapy (completed Phase I trials), as well as Bax adenovirus gene therapy for locoregional cancer control (Table 1A). Alternatively, it might be possible to activate the Bax and Bak proteins using drugs that mimic agonistic BH3 peptides (Letai et al., 2002).

### Extrinsic pathway

The extrinsic pathway is activated in vivo by TNF family ligands that engage DD-containing receptors, resulting in activation of DED-containing caspases. The "death ligands" are expressed on CTLs, NK cells, and other types of immune-relevant cells (activated monocytes/macrophages and dendritic cells) and are used as weapons for eradication of transformed cells (Locksley et al., 2001). Various studies using mice with genetic alterations in genes encoding death ligands or their receptors, as well as use of neutralizing antibodies and Fc-fusion proteins, have provided evidence of important roles in tumor suppression by cellular immune mechanisms. For example, Fas-Ligand (FasL) is important for CTL-mediated killing of some tumor targets, and TRAIL (Apo2-Ligand) is critical for NK-mediated tumor suppression.

Interest has emerged in developing therapeutics that kill cancer cells via the extrinsic pathway, particularly since chemorefractory cells tend to have defects in their intrinsic pathway, given the predominant reliance of cytotoxic drugs and x-irradiation on the mitochondrial pathway for cell death (Green and Reed, 1998). Attempts to apply TNF as a biological agent for cancer treatment were stymied by the proinflammatory effects of this cytokine, due to its simultaneous induction of both caspase activation pathways and NFκB. In contrast, the TNF family cytokines FasL and TRAIL trigger activation of the caspase pathway without concomitant induction of NFκB—raising hopes that they might be successfully exploited for cancer therapy, where TNF failed due to toxicity. While agonistic antibodies that trigger the receptor Fas (CD95) are unfortunately highly toxic to liver (Ogasawara et al., 1993), TRAIL and agonistic antibodies that bind TRAIL receptors appear to be well tolerated in vivo. Indeed, a Phase I trial in humans was recently completed using an agonistic antibody directed against TRAIL-receptor-1 (TRAIL-R1) (also known as death receptor-4 [DR4]) (Table 1B). In mouse xenograft models using selected tumor human cells lines, TRAIL and agonistic antibodies directed against TRAIL receptors have been demonstrated to possess potent antitumor activity (Ashkenazi et al., 1999), raising hopes of using these biological agents as a novel approach to cancer treatment and thereby mimicking some of the effector mechanisms normally employed by the immune system in its defense against transformed cells, and potentially bypassing the roadblocks to apop-

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tosis within the intrinsic (mitochondrial) pathway.

Unfortunately, many tumor cell lines display intrinsic resistance to TRAIL, despite expressing the necessary cell-surface receptors. This observation implies that during evolution of tumors in vivo, selection occurs for malignant clones capable of withstanding immune attack, and thus, successful biological therapy depends on restoring competency of the extrinsic pathway. A possible mechanism for accomplishing this goal has recently been revealed by studies of synthetic triterpenoids, such as CDDO and CDDOm, that trigger a poorly defined pathway for ubiquitination and degradation of FLIP (Kim et al., 2002; Pedersen et al., 2002), an apoptosis-modulating protein that binds procaspase-8 and -10 (Tschopp et al., 1999). At least in vitro, these compounds sensitize solid tumor cell lines to TRAIL, and induce apoptosis as single agents in leukemia cells, through a caspase-8-dependent mechanism that remains operative even in chemorefractory cells (Ito et al., 2000; Kim et al., 2002; Pedersen et al., 2002). It remains to be determined whether these promising observations will hold up in vivo.

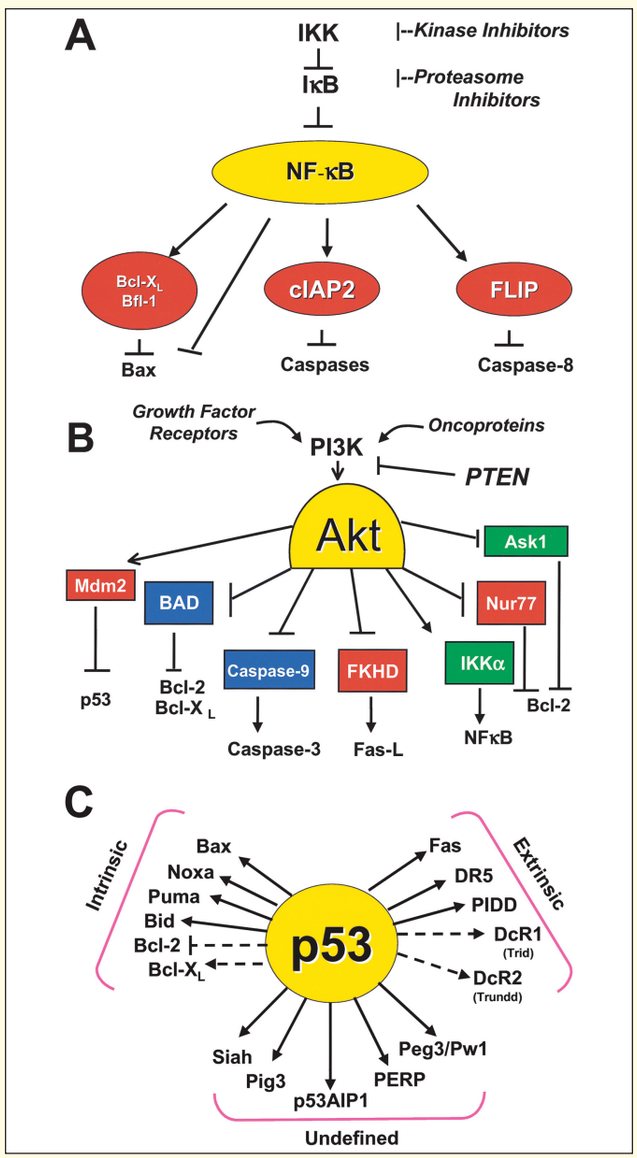


**Figure 2.** Signal transduction and apoptosis regulation

Some protein kinases and transcription factors are well known for their ability to modulate activity or expression of apoptosis-relevant proteins. The figure shows examples for (**A**) NFκB, (**B**) Akt, and (**C**) p53.

Other routes to engaging the extrinsic pathway have recently been revealed that may apply at least to leukemia. For example, the protein kinase C (PKC) modulator byrostatin induces myeloid leukemia cell lines to produce TNF, resulting in autocrine engagement of TNF-receptors and apoptosis induction through a mechanism that is suppressible by TNFR-Fc fusion protein and caspase-8 dominant-negative (Cartee et al., 2002). Similarly, all-*trans*-retinoic acid (ATRA) induces TRAIL production and triggers autocrine induction of apoptosis in acute promyelomonocytic leukemia (APML) cells that harbor the RARα-PML fusion protein resulting from t(15;17) chromosomal translocations (Altucci et al., 2001). It remains to be determined whether similar autocrine loops can be activated by these agents in solid tumors.

**Convergence pathway**

Ultimately, the intrinsic and extrinsic pathways for caspase activation converge on downstream effector caspases (Figure 1). Mechanisms for suppressing apoptosis at this distal step in cell death pathways have been revealed, and their relevance to cancer is becoming progressively obvious. In this regard, the inhibitor of apoptosis proteins (IAPs) represent a family of evolutionarily conserved apoptosis suppressors (n = 8 in humans), many of which function as endogenous inhibitors of caspases (Deveraux and Reed, 1999). All members of this family, by definition, contain at least one copy of a so-called BIR (*baculovirus iap repeat*) domain, a zinc binding fold, which is important for their antiapoptotic activity, present in 1–3 copies. Among the caspases directly inhibited by human IAP family members XIAP, cIAP1, and cIAP2 are effector caspases-3 and -7, as well as initiator caspase-9 (intrinsic pathway). For XIAP, the second BIR domain and the linker region between BIR1 and BIR2 are required for binding and suppressing caspases-3 and -7, while the third BIR domain binds caspase-9. Thus, different domains in the multi-BIR containing IAPs are responsible for suppression of different caspases. IAP family member Livin (ML-IAP) contains a single BIR and inhibits caspase-9 but not caspase-3 and -7. Survivin also contains a single BIR and reportedly associates with caspase-9, though its mechanism of caspase suppression is poorly defined.

Pathological overexpression of IAPs occurs in many cancers, though more detailed information is needed about which of the 8 members of this gene family is deregulated in specific types of cancer. Antisense-mediated reductions in IAP-family members XIAP, cIAP1, Survivin, or Apollon can induce apoptosis of tumor cell lines in culture, or sensitize cells to cytotoxic anticancer drugs, thus providing proof-of-concept evidence that the pathological elevations of IAPs found in cancers are important for maintaining tumor cell survival and resistance to chemotherapy (Table 1C).

Endogenous antagonists of IAPs help to keep these apoptosis suppressors in check, promoting apoptosis. Two of these naturally occurring IAP-antagonists, SMAC (Diablo) and HtrA2 (Omi), are sequestered inside mitochondria, becoming released into the cytosol during apoptosis (Reed, 2002). SMAC and HtrA2 have N-terminal leader sequences that are removed

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by proteolysis upon import into mitochondria, exposing a novel tetrapeptide motif that binds the BIR domains of IAPs. These IAP antagonists compete with caspases for binding to IAPs, thus freeing caspases from the grip of the IAPs and promoting apoptosis. Synthetic peptides that mimic SMAC and HtrA2 trigger apoptosis or sensitize tumor cell lines to apoptosis induced by cytotoxic anticancer drugs or TRAIL in vitro and even in some tumor xenograft models (Fulda et al., 2002). Structural information derived from high-field NMR and X-ray crystallography indicates that a tetrapeptide motif is sufficient to bind the BIRs of IAPs, suggesting a path to drug discovery where small-molecule chemical compounds that occupy the same tetrapeptide binding motif on BIRs can be envisioned as new apoptosis-sensitizing agents for cancer (Wu et al., 2000). However, many issues remain unresolved, including the questions of which IAP-family members, and which BIR domains within those IAPs, should be targeted by drugs.

**Multipathway regulators**

Though not core components of the cell death machinery, certain signaling proteins have emerged as potential drug targets for modulating apoptosis pathways, based on their ability to influence the expression or function of other proteins. Some of these apoptosis-modulating proteins may impact several apoptosis pathways simultaneously, possibly at multiple points. For example, NFκB family transcription factors can induce expression of antiapoptotic proteins that oppose the intrinsic (Bcl-$X_L$, Bfl-1), extrinsic (FLIP), and convergence (cIAP2) pathways (Figure 2A), as well as suppressing expression of the death inducer Bax (intrinsic pathway) in some types of tumor cells (Karin and Lin, 2002). Further, hyperactivity of NFκB is now well documented in certain cancers (Karin et al., 2002). Consequently, agents that inhibit NFκB activation are desired, and are currently being pursued, based on strategies that seek to maintain levels of endogenous NFκB-inhibiting IκB-family proteins, by either suppressing the IκB kinases (IKKs) responsible for phosphorylation of IκBα (which induces its polyubiquitination) or blocking the proteolytic activity of the 26s proteasome responsible for degrading IkB following ubiquitination (Figure 2A).

Similarly, Akt (PKB) directly phosphorylates multiple protein targets of relevance to apoptosis (Figure 2B), suppressing cell death clearly within the intrinsic pathway (e.g., BAD inactivation, human caspase-9 suppression) and possibly also the extrinsic pathway (e.g., FasL expression) in some types of cells. Pathological elevations in Akt activity are a common occurrence in tumors, due to loss of tumor suppressor PTEN, hyperactivity of PI3K-activating growth factors receptors and oncoproteins, and other mechanisms (Testa and Bellacosa, 2001). Small-molecule drugs that occupy the ATP binding pocket of the catalytic domain of Akt thus represent a highly attractive approach to restoration of apoptosis sensitivity in cancers, and would also be expected to have additional benefits on cell proliferation and cell growth. While the PH domain of Akt that binds second-messenger phospholipids involved in Akt activation could theoretically be targeted by drugs, the hydrophilic interactions associated with coordinating polyphosphorylated lipids do not lend themselves well to membrane permeable compounds.

The transcription factor p53 reportedly regulates the expression of multiple apoptosis-regulating genes that affect either the intrinsic or extrinsic pathways (Figure 2C), though the specific targets of p53 that are most relevant probably differ widely among tissues and tumor types (Vogelstein et al., 2000). Clinical trials are underway using gene therapy-based approaches for restoring p53 activity in deficient cancers, which might permit better loco-regional control of cancer in combination with radiotherapy.

In addition to NFκB, Akt, and p53, multiple cancer-relevant proteins that operate upstream of these factors could to some extent be viewed as apoptosis modulators, though acting rather indirectly on the core cell death machinery. Therapies targeting these upstream inducers include many biological agents, such as: (1) monoclonal antibodies recognizing cell surface receptors (e.g., anti-HER2 [Herceptin], anti-EGFR [Erbitux]), (2) small molecule drugs that target the kinase domains of these receptors and other oncogenic protein tyrosine kinases (e.g., EGFR/Iressa, BCR-ABL/Gleevec), which presumably suppress PI3K/Akt activation, and (3) anti-CD40L antibodies and CD40L adenovirus gene therapy, which modulate NFκB activity. Monoclonal antibodies targeting the tetra-span membrane protein CD20 (Rituximab) also can downregulate expression of Bcl-2-family member Mcl-1 (intrinsic pathway) and XIAP (convergence pathway) in certain leukemias, providing yet another example of unexpected links to apoptosis pathways through receptor-mediated signaling, in a context relevant to cancer treatment (Byrd et al., 2002). Thus, abundant opportunities may exist for exploiting receptor targeting agents for indirectly restoring apoptosis sensitivity in cancer, representing an area worthy of further in-depth exploration.

**Conclusions**

From an improved understanding of apoptosis mechanisms, multiple novel strategies have emerged for restoring apoptosis sensitivity in cancer, and clinical testing of several of these approaches is underway. Much remains to be learned about how best to exploit these new potential therapies, matching genetic lesion in cancers to the optimal agent. It remains to be determined to what extent toxicity to normal tissues will limit application of apoptosis-based therapies for cancer treatment. Cancer cells, however, may be relatively more dependent on apoptosis suppression than normal cells, due to aberrations in protooncogene activity and cell cycle checkpoint control, wandering of cancer cells from their normal sites of trophic support, and hypoxic conditions caused by inadequate vascular supply. Exploiting this differential dependence on apoptosis-suppressing mechanisms offers hope that improved clinical outcomes may not be far from realization.

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