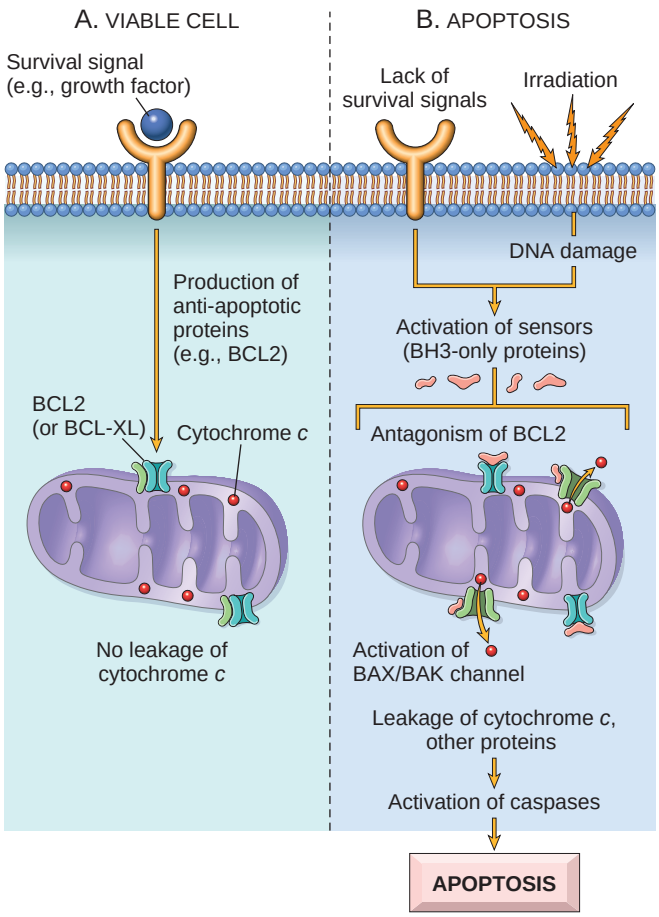

Figure 2-24 The intrinsic (mitochondrial) pathway of apoptosis. **A,** Cell viability is maintained by the induction of anti-apoptotic proteins such as BCL2 by survival signals. These proteins maintain the integrity of mitochondrial membranes and prevent leakage of mitochondrial proteins. **B,** Loss of survival signals, DNA damage, and other insults activate sensors that antagonize the anti-apoptotic proteins and activate the pro-apoptotic proteins BAX and BAK, which form channels in the mitochondrial membrane. The subsequent leakage of cytochrome c (and other proteins, not shown) leads to caspase activation and apoptosis.

molecules from the mitochondrial intermembrane space into the cytoplasm (Fig. 2-24). Mitochondria are remarkable organelles in that they contain proteins such as cytochrome c that are essential for life, but some of the same proteins, in particular cytochrome c, when released into the cytoplasm (an indication that the cell is not healthy), initiate the suicide program of apoptosis. The release of mitochondrial pro-apoptotic proteins is tightly controlled by the BCL2 family of proteins This family is named after *BCL2*, which is frequently overexpressed due to chromosomal translocations and resulting rearrangements in certain B cell lymphomas (Chapter 13). There are more than 20 members of the *BCL* family, which can be divided into three groups based on their pro-apoptotic or anti-apoptotic function and the BCL2 homology (BH) domains they possess.

- *Anti-apoptotic.* BCL2, BCL-XL, and MCL1 are the principal members of this group; they possess four BH domains (called BH1-4). These proteins reside in the outer mitochondrial membranes as well as the cytosol and ER membranes. By keeping the mitochondrial outer membrane impermeable they prevent leakage of cytochrome c and other death-inducing proteins into the cytosol (Fig. 2-24*A*).
- *Pro-apoptotic.* BAX and BAK are the two prototypic members of this group. Like their anti-apoptotic cousins they also have four BH domains. Upon activation, BAX and BAK oligomerize within the outer mitochondrial protein and promote mitochondrial outer membrane permeability. The precise mechanism by which Bax-Bak permeabilize membranes is not settled. According to one model illustrated in Fig 2-24*B*, they form a channel in the outer mitochondrial membrane, allowing leakage of cytochrome c from the intermembranous space.
- *Sensors.* Members of this group, including BAD, BIM, BID, Puma, and Noxa, contain only one BH domain, the third of the four BH domains, and hence are sometimes called BH3-only proteins. BH3-only proteins act as sensors of cellular stress and damage, and regulate the balance between the other two groups, thus acting as arbiters of apoptosis.

Growth factors and other survival signals stimulate the production of anti-apoptotic proteins such as BCL2, thus preventing the leakage of death-inducing proteins from the outer mitochondrial membrane. When cells are deprived of survival signals or their DNA is damaged, or misfolded proteins induce ER stress, the BH3-only proteins "sense" such damage and are activated. These sensors in turn activate the two critical (pro-apoptotic) effectors, BAX and BAK, which form oligomers that insert into the mitochondrial membrane and allow proteins from the inner mitochondrial membrane to leak out into the cytoplasm. BH3-only proteins may also bind to and block the function of BCL2 and BCL-XL. At the same time, the synthesis of BCL2 and BCL-XL may decline because of the relative deficiency of survival signals. The net result of BAX-BAK activation coupled with loss of the protective functions of the anti-apoptotic BCL2 family members is the release into the cytoplasm of several mitochondrial proteins that can activate the caspase cascade (Fig. 2-24). As already mentioned, one of these proteins is cytochrome c, well known for its role in mitochondrial respiration.

Once released into the cytosol, cytochrome c binds to a protein called APAF-1 (apoptosis-activating factor-1), which forms a wheel-like hexamer that has been called the *apoptosome*. This complex is able to bind caspase-9, the critical initiator caspase of the mitochondrial pathway, and the enzyme cleaves adjacent caspase-9 molecules, thus setting up an autoamplification process. Cleavage activates caspase-9, which triggers a cascade of caspase activation by cleaving and thereby activating other pro-caspases, and the active enzymes mediate the execution phase of apoptosis (discussed later). Other mitochondrial proteins, with arcane names like Smac/Diablo, enter the cytoplasm, where they bind to and neutralize cytoplasmic proteins that function as physiologic inhibitors of apoptosis (called IAPs). The normal function of the IAPs is to block the activation of caspases, including executioners like caspase-3, and keep cells alive. Thus, the neutralization of these IAPs permits the initiation of a caspase cascade.