CASPASE ACTIVATION & APOPTOSIS

Extrinsic & Intrinsic Pathways of Caspase Activation

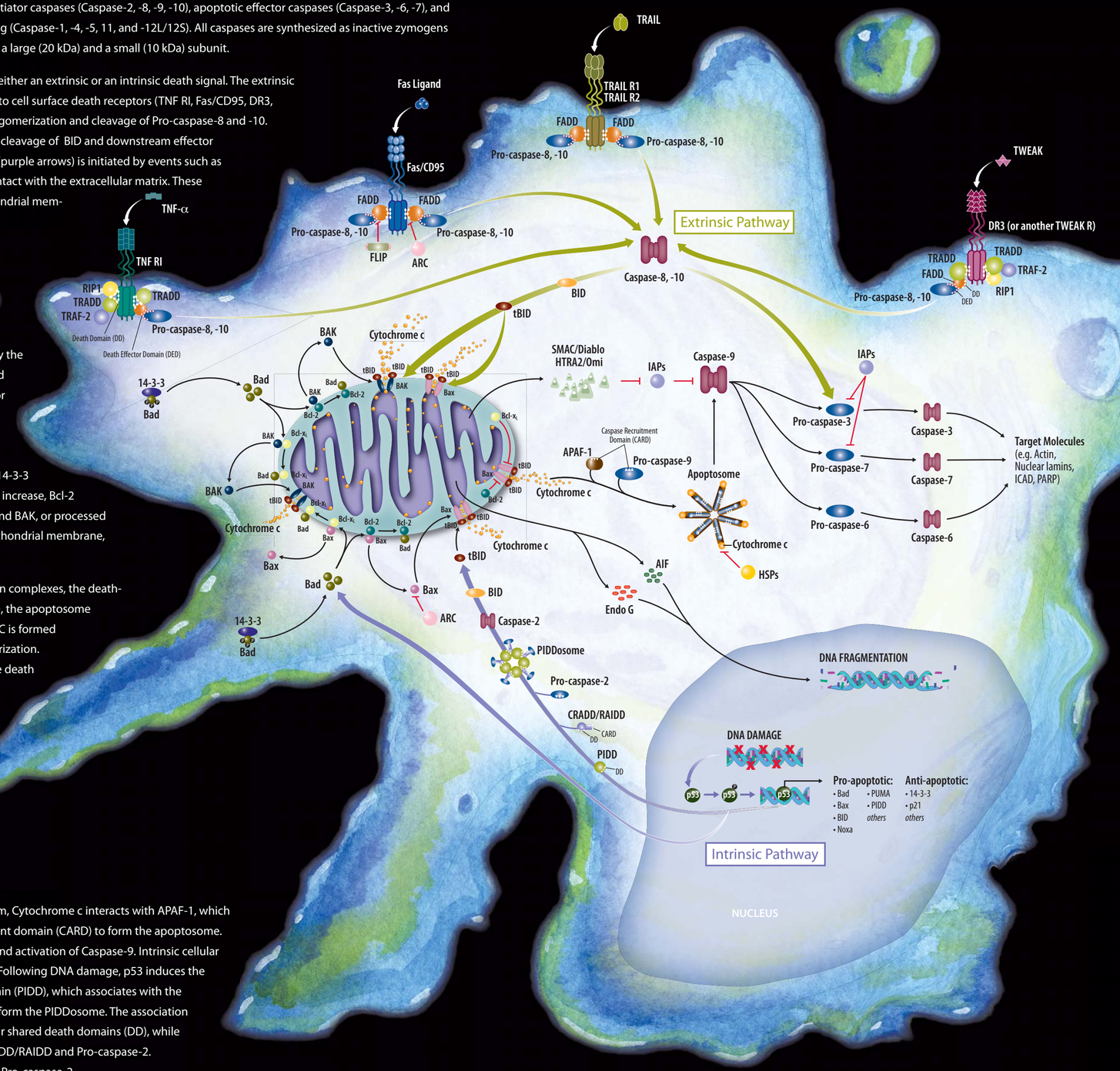
Caspases are a family of aspartate-specific, cysteine proteases that serve as the primary mediators of apoptosis. Mammalian caspases can be subdivided into three functional groups, apoptotic initiator caspases (Caspase-2, -8, -9, -10), apoptotic effector caspases (Caspase-3, -6, -7), and caspases involved in inflammatory cytokine processing (Caspase-1, -4, -5, 11, and -12L/12S). All caspases are synthesized as inactive zymogens containing a variable length pro-domain, followed by a large (20 kDa) and a small (10 kDa) subunit.

Apoptotic caspases are activated upon the receipt of either an extrinsic or an intrinsic death signal. The extrinsic pathway (green arrows) is initiated by ligand binding to cell surface death receptors (TNF RI, Fas/CD95, DR3, TRAIL R1/DR4, TRAIL R2/DR5) followed by receptor oligomerization and cleavage of Pro-caspase-8 and -10. Activation of Caspase-8 and Caspase-10 results in the cleavage of BID and downstream effector caspases. The intrinsic pathway of caspase activation (purple arrows) is initiated by events such as DNA damage, growth factor withdrawal, or loss of contact with the extracellular matrix. These events lead to changes in the integrity of the mitochondrial membrane that result in the release of pro-apoptotic proteins including Cytochrome c, Smac/Diablo, HTRA2/Omi, Apoptosis-Inducing Factor (AIF), and Endonuclease G.

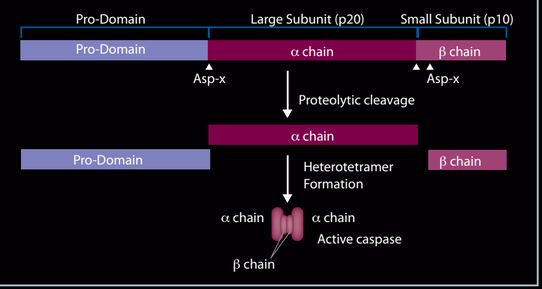
Mitochondrial membrane permeability is regulated by the Bcl-2 family of proteins. The balance between pro- and anti-apoptotic family members determines whether or not a cell will undergo apoptosis. In healthy cells, Bcl-2 and Bcl-x_L inhibit apoptosis by binding to the pro-apoptotic Bax and BAK proteins. Bad is also phosphorylated and sequestered in the cytoplasm by the 14-3-3 protein in these cells. If cytoplasmic levels of free BAD increase, Bcl-2 and Bcl-x_L bind to Bad and release Bax and BAK. Bax and BAK, or processed forms of these proteins, can then insert into the mitochondrial membrane, compromising its integrity.

Initiator caspases are activated in three distinct protein complexes, the death-inducing signaling complex (DISC; Caspase-8 and -10), the apoptosome (Caspase-9), and the PIDDosome (Caspase-2). The DISC is formed following ligand binding and death receptor oligomerization. Pro-caspase-8 and Pro-caspase-10 are recruited to the death receptors through their interactions with the adaptor protein, FADD. This interaction is mediated by their shared death effector domains (DED). Clustering of pro-caspases near the death receptors results in their cleavage and activation. In contrast, Pro-caspase-9 is activated following an intrinsic change associated with the release of Cytochrome c from the mitochondria. In the cytoplasm, Cytochrome c interacts with APAF-1, which recruits Pro-caspase-9 by way of its caspase recruitment domain (CARD) to form the apoptosome. Formation of the apoptosome leads to the cleavage and activation of Caspase-9. Intrinsic cellular changes can also lead to the activation of Caspase-2. Following DNA damage, p53 induces the expression of p53-induced protein with a death domain (PIDD), which associates with the CRADD/RAIDD adaptor protein and Pro-caspase-2 to form the PIDDosome. The association between CRADD/RAIDD and PIDD is mediated by their shared death domains (DD), while CARD domains mediate the interaction between CRADD/RAIDD and Pro-caspase-2. Formation of the PIDDosome leads to the cleavage of Pro-caspase-2.

Autocatalytic cleavage of the initiator pro-caspases occurs at aspartic acid residues located after the pro-domain, and in between the large and the small subunits. Upon cleavage, mature caspases form a proteolytically active heterotetramer consisting of two small and two large subunits. Once activated, initiator caspases cleave downstream effector caspases that promote the ordered disassembly of the cell by targeting a number of critical cellular proteins including structural proteins, DNA repair proteins, and proteins involved in signal transduction pathways.

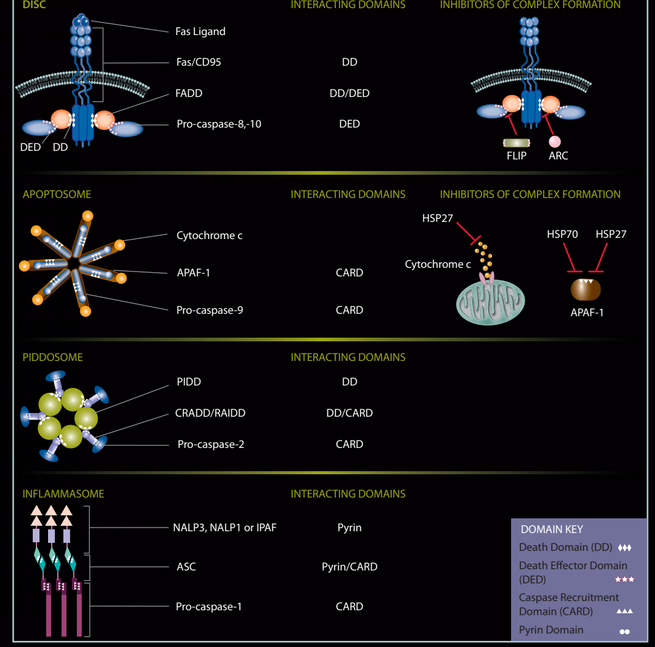


CASPASE CLEAVAGE & ACTIVATION



MAMMALIAN CASPASE DOMAINS & CLEAVAGE SITES

CASPASE ACTIVATING COMPLEXES



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Note: This poster conveys a general overview and should be considered neither comprehensive nor definitive. The details of the process are understood to be subject to interpretation.