Apoptosis and Cell Death: Comprehensive Overview

Intrinsic Pathway

- Mitochondrial pathway
- Cytochrome c release
- Apoptosome formation
- Triggered by DNA damage, stress

Extrinsic Pathway

- Death receptor activation
- FAS, TNF receptors
- DISC complex formation
- Immune-mediated

Caspase Cascade

- Initiator caspases (8, 9)
- Executioner caspases (3, 7)
- Proteolytic cleavage
- Irreversible commitment

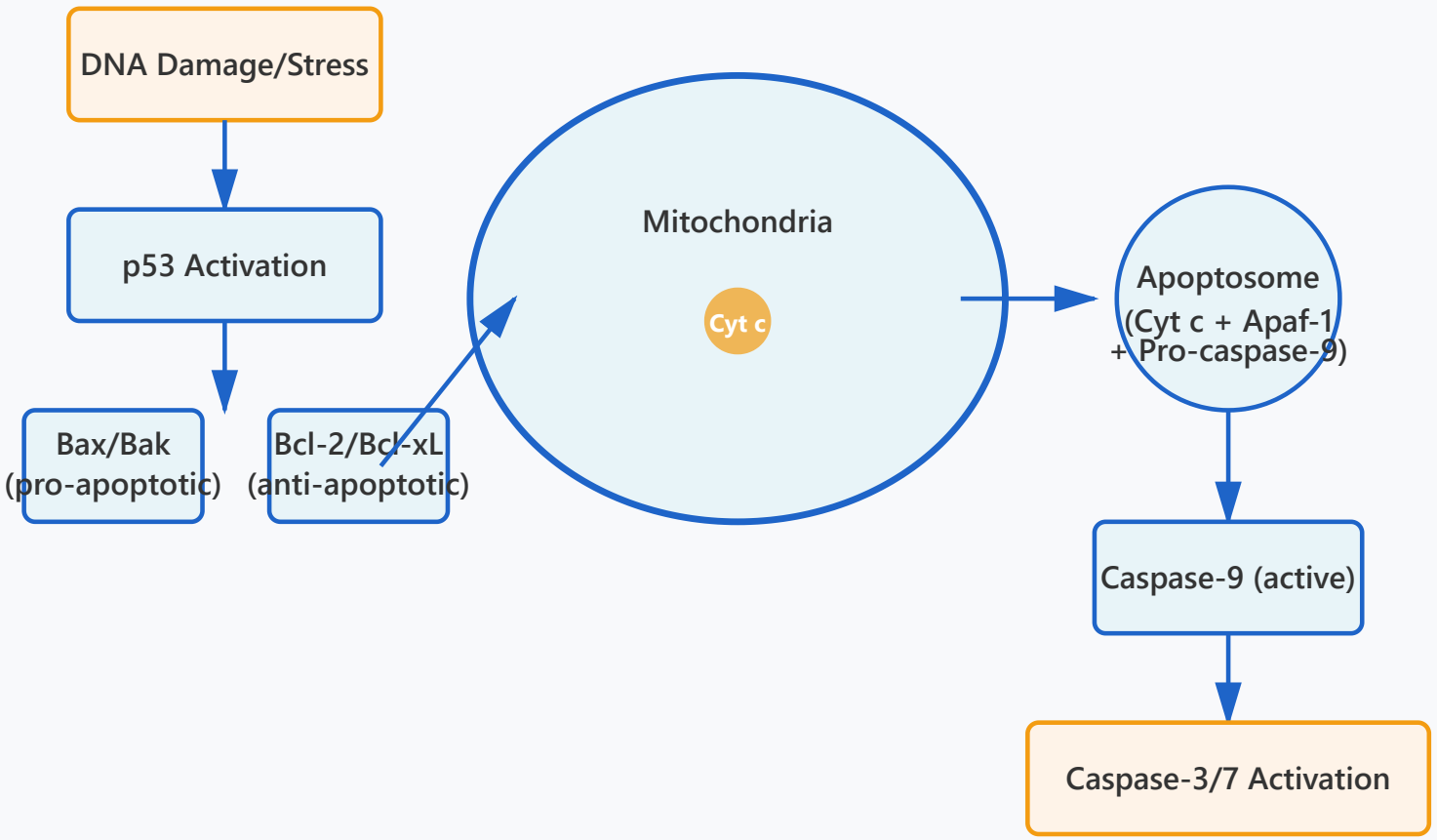
Regulation

- Bcl-2 family: pro and anti-apoptotic
- IAPs: caspase inhibitors
- p53: apoptosis inducer
- Cancer dysregulation

1 Intrinsic Pathway (Mitochondrial Pathway)

The intrinsic pathway is initiated by intracellular stress signals such as DNA damage, oxidative stress, growth factor deprivation, or endoplasmic reticulum stress. This pathway is centered around mitochondrial outer membrane permeabilization (MOMP), which is

the point of no return in the apoptotic process.



Key Mechanisms:

→ **MOMP (Mitochondrial Outer Membrane Permeabilization):** Controlled by the Bcl-2 family of proteins. Pro-apoptotic members (Bax, Bak) oligomerize to form pores in the outer mitochondrial membrane.

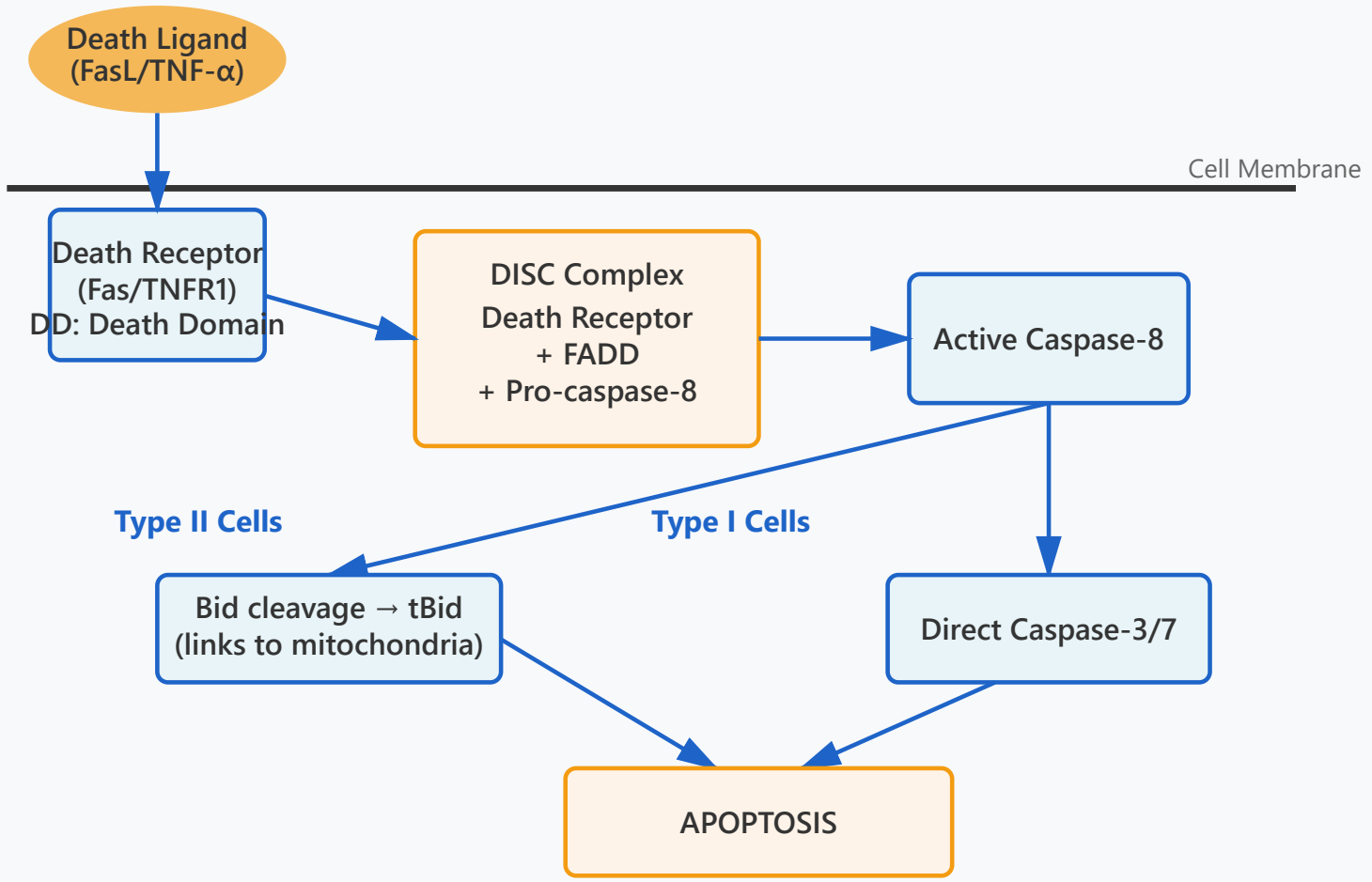
→ **Cytochrome c Release:** Once released into the cytosol, cytochrome c binds to Apaf-1 (apoptotic protease activating factor-1) in the presence of ATP/dATP.

→ **Apoptosome Formation:** A wheel-like heptameric complex that recruits and activates pro-caspase-9, initiating the caspase cascade.

→ **Clinical Relevance:** Many chemotherapy drugs work by inducing DNA damage that triggers the intrinsic pathway. Cancer cells often develop resistance by overexpressing anti-apoptotic Bcl-2 proteins.

2 Extrinsic Pathway (Death Receptor Pathway)

The extrinsic pathway is initiated by the binding of extracellular death ligands to their corresponding death receptors on the cell surface. This pathway is crucial for immune system function, allowing cytotoxic T cells and natural killer cells to eliminate infected or cancerous cells.



Key Mechanisms:

→ **Death Receptors:** Members of the TNF receptor superfamily containing intracellular death domains (DD). Main examples include Fas (CD95), TNFR1, DR4, and DR5 (TRAIL receptors).

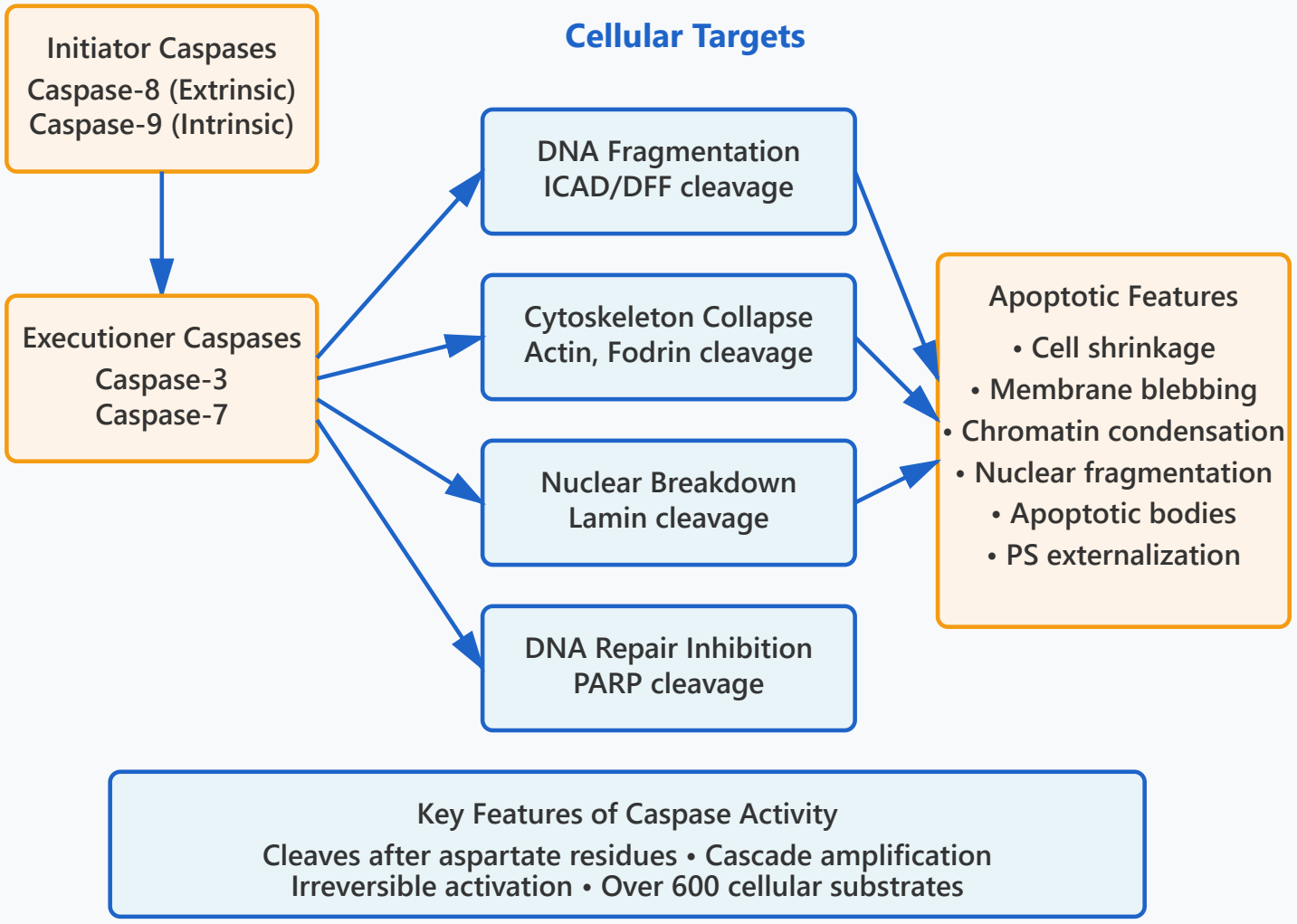
→ **DISC Assembly:** Upon ligand binding, death receptors trimerize and recruit adaptor protein FADD (Fas-Associated Death Domain), which then recruits pro-caspase-8 through death effector domains (DED).

→ **Type I vs Type II Cells:** Type I cells generate sufficient active caspase-8 to directly activate executioner caspases. Type II cells require amplification through the mitochondrial pathway via Bid cleavage.

→ **Immune Function:** Cytotoxic T lymphocytes use FasL and TRAIL to eliminate target cells. Defects in Fas signaling cause autoimmune lymphoproliferative syndrome (ALPS).

3 Caspase Cascade: The Execution Machinery

Caspases (cysteine-aspartic proteases) are the central executioners of apoptosis. They exist as inactive zymogens (pro-caspases) and are activated through proteolytic cleavage. The caspase cascade amplifies the death signal and ensures irreversible commitment to cell death.



Key Mechanisms:

→ **Caspase Structure:** All caspases contain a catalytic cysteine residue and cleave substrates after aspartate residues (hence "cysteine-aspartic protease"). They exist as dimeric zymogens requiring proteolytic processing for activation.

→ **Cascade Amplification:** One initiator caspase can activate multiple executioner caspases, and each executioner caspase can cleave hundreds of substrate proteins, creating a powerful amplification loop.

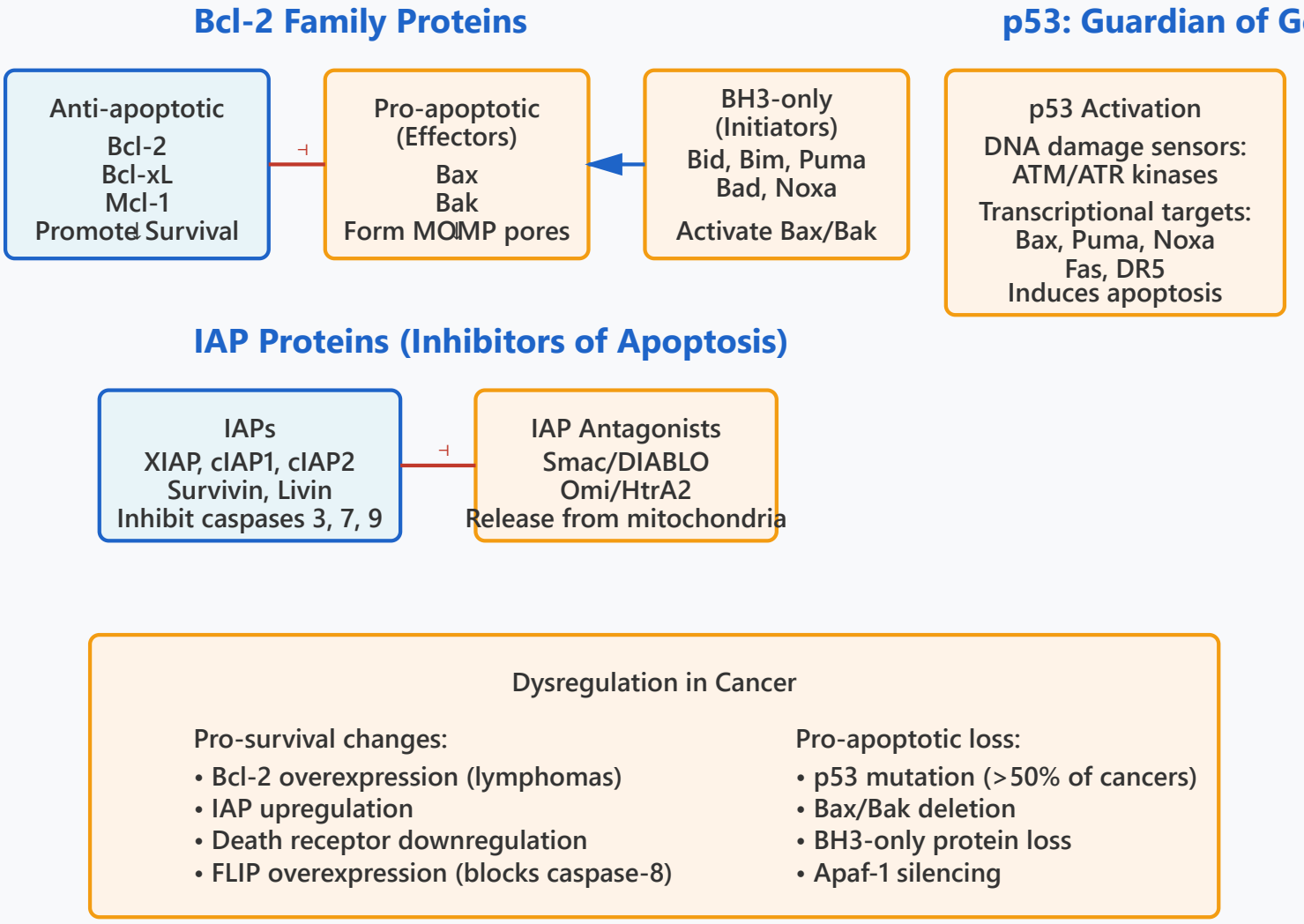
→ **Substrate Specificity:** Over 600 proteins are cleaved during apoptosis, including PARP (DNA repair), ICAD/DFF (DNA fragmentation), lamins (nuclear structure), and cytoskeletal proteins (fodrin, actin, gelsolin).

→ **Point of No Return:** Once executioner caspases are fully activated, the cell is irreversibly committed to death. This ensures that damaged or dangerous cells are completely eliminated.

→ **Clinical Applications:** Caspase activity can be measured using fluorogenic substrates (e.g., Ac-DEVD-AMC for caspase-3) and is used as a biomarker for apoptosis in research and drug development.

4 Regulation of Apoptosis: Balance Between Life and Death

Apoptosis is tightly regulated by multiple protein families that act as molecular switches between cell survival and death. Dysregulation of these pathways is a hallmark of cancer and many degenerative diseases. The balance between pro-apoptotic and anti-apoptotic signals determines cell fate.



Key Regulatory Mechanisms:

→ **Bcl-2 Family Balance:** The ratio of anti-apoptotic (Bcl-2, Bcl-xL, Mcl-1) to pro-apoptotic (Bax, Bak, BH3-only proteins) members determines the threshold for apoptosis. Anti-apoptotic members sequester BH3-only proteins and prevent Bax/Bak activation.

→ **IAP Function:** IAPs contain BIR (baculovirus IAP repeat) domains that directly bind and inhibit caspases. XIAP is the most potent, capable of inhibiting caspases-3, -7, and -9. Smac/DIABLO relieves this inhibition when released from mitochondria.

→ **p53 as Master Regulator:** Called the "guardian of the genome," p53 is activated by DNA damage, oncogenic stress, and hypoxia. It transcriptionally activates multiple pro-apoptotic genes and is mutated in over 50% of human cancers.

→ **Therapeutic Targeting:** BH3 mimetics (venetoclax/ABT-199) inhibit Bcl-2 and are FDA-approved for CLL and AML. Smac mimetics and IAP antagonists are in clinical trials. Restoring p53 function is a major goal of cancer research.

→ **Resistance Mechanisms:** Cancer cells evade apoptosis through: overexpressing anti-apoptotic proteins, mutating death receptors, upregulating IAPs, silencing pro-apoptotic genes via methylation, and losing p53 function through mutation or MDM2 amplification.