PROGRAMMED CELL DEATH

LECTURE OUTLINE

1. Introduction

2. Programmed Cell Death
   a. Apoptosis vs. Necrosis
   b. Apoptosis is caused by a proteolytic cascade
   c. Initiation of apoptosis

3. Integration of Internal and External Cues

4. Programmed Cell Death, Cell Cycle, and Cancer

DISEASES/GENETIC DISORDERS RELEVANT TO LECTURE

Cancer of all types; Retinoblastoma
1. INTRODUCTION

In prosperous countries, 1 in 5 people will die of cancer (discussed in previous lecture).

Organisms maintain an optimal number of cells both by regulating the production of cells (cell division) and by regulating the destruction of cells (programmed cell death). [Fig. 20-14, p. 1216]

Destruction of unneeded cells occurs by a variety of types of programmed cell death. Apoptosis is the best-understood of these pathways. Other pathways include autophagic cell death and mitotic catastrophe.

Apoptosis is actually a specific type of Programmed Cell Death, but the two terms are sometimes used as if they are interchangeable.
2. PROGRAMMED CELL DEATH

When it undergoes programmed cell death, a cell kills itself by activating an intracellular cell death program. Apoptosis is the most common and best-understood form of programmed cell death.

a. Apoptosis vs. Necrosis

*apo* = from  
*ptosis* = falling

1. Apoptosis is most visible during development, but it also occurs in the adult organism.
   - One example of apoptosis is the elimination of autoreactive B cells and T cells (Immunology lecture).
   - Development of hands and paws requires the programmed death of the cells between the digits [Fig. 18-2, p.1116]

APOPTOSIS IN THE DEVELOPING MOUSE
PAW SCULPTS THE DIGITS

2. Apoptosis is more like induced suicide than murder (Immunology lecture) [Fig. 18-1, p.1116 (next page of notes)].
   - *Necrosis* is usually caused by major insults, such as trauma or lack of blood supply.
     - Cells swell, burst, and spill their contents into the surrounding tissue.
     - Inflammation occurs.
   - *Apoptosis* is usually mediated by internal or external cues.
     - Nuclear envelope disintegrates; chromatin condenses; DNA is chopped into nucleosome-sized pieces.
     - Phosphatidylserine flips to the outside of the plasma membrane, marking the cell.
     - The apoptotic cell is frequently consumed by another cell, such as a macrophage.
     - Inflammation does not occur.
APOPTOSIS vs. NECROSIS

(A)  

(B) engulfed dead cell  

(C) phagocytic cell

Apoptosis

[Cell Biology Interactive video “GCCC”]
b. **Apoptosis is accomplished by the actions of a proteolytic cascade.**

The proteases responsible for apoptosis are members of a family of proteases called caspases.

- Caspases are cysteine proteases that recognize tetrapeptide motifs and cleave their substrates at sites on the carboxyl side of aspartate residues.
- Caspases exist in an inactive procaspace form until they are cleaved.

**PROCASPASE ACTIVATION**

- The first caspases to be activated are the “initiator” caspases. Each initiator caspase molecule cleaves and activates several “executioner” caspase molecules.
- In mammalian cells, for example, active Caspase 9 molecules each cleave and activate several Caspase 3 molecules [Fig. 18-5, p.1119].

**PROTEOLYTIC CASCADE**
The protease activity of the caspases causes the changes observed during apoptosis.

- The nuclear membrane, for example, is broken down by proteolysis of the nuclear lamins.
- The inhibitor of a specific endonuclease is cleaved by caspase activity, allowing the endonuclease to chop up DNA.

c. Initiation of Apoptosis.

The apoptotic pathway can be initiated by either external or internal stimuli.

1. Cell surface death receptors activate the extrinsic pathway of apoptosis. [Fig. 18-16, p. 1120]
   - For example, killer lymphocytes induce apoptosis in their target cells using this pathway (Immunology lecture).

EXTERNAL STIMULI CAN TRIGGER APOPTOSIS
2. The intrinsic pathway of apoptosis depends on mitochondria.

- The intrinsic pathway of apoptosis arises in response to cellular stresses such as severe DNA damage or lack of oxygen.
- The intrinsic pathway of apoptosis begins with release of cytochrome c from the mitochondria. [Fig 18-7, p. 1121]

RELEASE OF CYTOCHROME C FROM THE MITOCHONDRIA INITIATES THE INTRINSIC PATHWAY

- Cytochrome c release leads to the activation of initiator caspases, which themselves activate executioner caspases [Fig. 18-8, p. 1122]

INTERNAL STIMULI CAN TRIGGER APOPTOSIS
CANCER: The most common non-Hodgkin’s lymphoma (follicular lymphoma) is characterized by a chromosomal translocation (t(14;18)) between an immunoglobulin heavy chain locus and the Bcl2 gene. This fusion causes increased expression of Bcl2 (an anti-apoptotic protein) in these cells.

- The balance of activities of Bcl2-related proteins (some of which reside in the mitochondrial membrane) determines whether the intrinsic pathway of apoptosis is triggered.
  
  - Bcl2-related proteins fall into two classes: pro-apoptotic and anti-apoptotic
  
  - Bcl2-related proteins each contain between 1 and 4 regions of Bcl2-homology (“BH”) domains
  
  - Bcl2-related proteins interact directly with one another through their Bcl2-homology domains
  
  - Normally, cells remain viable because the anti-apoptotic activity prevails. When there is an excess of pro-apoptotic activity, the cell undergoes apoptosis.

3. INTEGRATION OF INTERNAL AND EXTERNAL CUES

The body is a collection of cooperative cells. The cells coexist peacefully by responding to cues from one another.

a. Growth factors

Growth factors were first discovered when scientists were trying to grow cells in culture.

- Successful with platelets or whole serum, but not with plasma
- Platelets release PDGF during clotting
- Serum deprivation prevents passage through G1 checkpoint

Growth factor table (Dr. Wang lecture)

<table>
<thead>
<tr>
<th>Growth Factor</th>
<th>Related Family Members</th>
<th>Broad or Narrow Specificity</th>
<th>Representative Actions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Platelet-derived growth factor (PDGF)—three subtypes</td>
<td>Transforming growth factor α (TGF-α); lin-3 protein (in C. elegans)</td>
<td>Broad</td>
<td>Stimulate proliferation of connective-tissue cells and some neuroglial cells</td>
</tr>
<tr>
<td>Epidermal growth factor (EGF)</td>
<td></td>
<td>Broad</td>
<td>Stimulate proliferation of many cell types; act as inductive signal in embryonic development</td>
</tr>
<tr>
<td>Insulinlike growth factor I (IGF-I)</td>
<td>Insulinlike growth factor II (IGF-II); insulin</td>
<td>Broad</td>
<td>Promote cell survival; stimulate cell metabolism; collaborate with other growth factors to stimulate cell proliferation</td>
</tr>
<tr>
<td>Interleukin-2 (IL-2)</td>
<td></td>
<td>Narrow</td>
<td>Stimulate proliferation of activated T lymphocytes</td>
</tr>
<tr>
<td>Nerve growth factor (NGF)</td>
<td>Brain-derived neurotrophic factor (BDNF); neurotrophin-3 (NT-3); neurotrophin-4 (NT-4)</td>
<td>Narrow</td>
<td>Promote survival and nerve process outgrowth of specific classes of neurons</td>
</tr>
</tbody>
</table>
b. **Internal and external cues are integrated in each cell.**
   - This integration is how the cells of the body are normally kept in balance.
   - Cells divide only when new cells are needed; cells die when they are no longer needed.
   - Each normal cell must be in the correct place, receiving the correct signals from its neighbors, or it will undergo apoptosis. [Fig. 15-8, p. 885; Fig 20-37, p. 1243]
**CANCER:** Epidermal growth factor (EGF in table on earlier page) normally binds to the EGF receptor to stimulate cell proliferation. Some mutations of the receptor delete the extracellular domain such that the stimulatory signal is produced constantly by the receptor, even if EGF is not present.

4. **PROGRAMMED CELL DEATH, THE CELL CYCLE, AND CANCER**

Many gene products that are normally involved in controlling the cell cycle and programmed cell death are found to be abnormal in some cancers. This brings up the possibility of either repairing them or substituting for their function as a potential future therapy.

Cancer cells usually contain multiple mutations. The mutations cause cells to divide when they should not divide, and to fail to undergo programmed cell death when they should die.

There are two broad categories of cancer genes: **proto-oncogenes** and **tumor suppressor** genes (Dr. Wang lecture).

- The normal products of **proto-oncogenes** help to tell the cell when to divide or when not to undergo apoptosis.
- The normal products of **tumor suppressor** genes help to tell the cell when not to divide or when to undergo apoptosis.
- It's the balance between these activities that normally keeps cell numbers appropriate.

a. **Proto-oncogenes**

A proto-oncogene is like an accelerator in a car; it can get stuck ON.

An “oncogene” is a proto-oncogene that carries such an activating mutation (i.e., it is a mutant allele of the proto-oncogene).

A single mutation of this type pushes the cell to proliferate or survive even when it should not do so. These mutations are “dominant” at the cellular level. They cause a gain of function that is not found in normal cells.
PROTO-ONCOGENES CAN MUTATE TO CAUSE PROLIFERATION

CANCER Examples:

1. The EGF receptor is encoded by the proto-oncogene erbB (Dr. Wang lecture). Mutations of the extracellular domain produce an oncogene that stimulates cell proliferation (discussed in the integration of external cues section of this lecture).

2. B cell type chronic lymphocytic leukemias sometimes have an immunoglobulin locus (14q32) fused to cyclin D1, which is an important G1 cyclin (discussed in cyclin-dependent kinase section of previous lecture).

3. Bcl2 (anti-apoptotic) fusions are characteristic of a common type of non-Hodgkin’s lymphoma (discussed in apoptosis section of this lecture).

b. Tumor suppressor genes

Tumor suppressor mutations cause the loss of functions that would normally be found in cells. Tumor suppressor genes are like the brakes in a car. If they’re broken, you can’t stop.

The best-known tumor suppressor genes are p53 (discussed in checkpoint section of previous lecture) and Rb.

CANCER: The Rb gene was originally identified through a rare inherited predisposition to childhood cancer (Retinoblastoma).
The normal function of the Rb gene is to keep some genes involved in cellular proliferation turned off [Fig. 20-38, b and c; p.1244].

**TUMOR SUPPRESSOR MUTANTS CAN FAIL TO STOP DIVISION**

In patients who are heterozygous for a mutation in a tumor suppressor gene such as Rb, only one genetic event is required to produce homozygous mutant cells, which then divide inappropriately. This is called “loss of heterozygosity” (discussed in previous lecture). [Fig. 20-30, p. 1235]

**LOSS OF THE ONE NORMAL RB GENE LEADS TO HEREDITARY RB**

Figure 20-30  Molecular Biology of the Cell (© Garland Science 2008)