

Morphologic and biochemical changes in apoptosis

A pathology teaching lecture

Morphology • Biochemical execution • Efferocytosis

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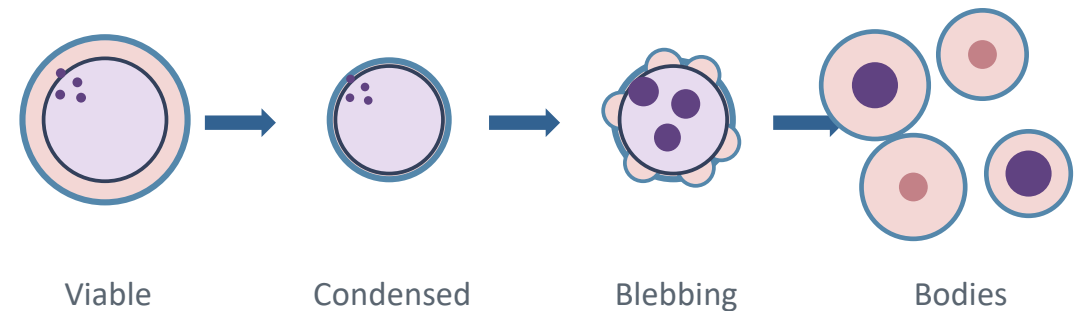
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Learning objectives

- Recognize the four cardinal morphologic features of apoptotic cells.
- Relate caspase-dependent biochemical events to cellular morphology.
- Explain apoptotic-body removal and the limited inflammatory response.
- Distinguish apoptotic changes from necrotic injury.

Reading the phenotype of apoptosis

- Apoptosis affects cells individually and produces an orderly, recognizable sequence.
- Morphology and biochemistry describe different aspects of the same process.
- Cells can disappear rapidly, so a single tissue section may underestimate apoptosis.

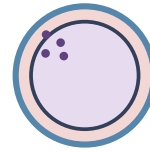


Schematic sequence; Stages may overlap

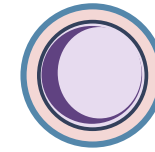
Schematic

Four defining morphologic findings

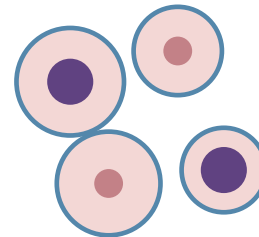
- Cell shrinkage and increased cytoplasmic density.
- Peripheral chromatin condensation, often with nuclear fragmentation.
- Membrane blebs followed by membrane-bound apoptotic bodies.
- Prompt ingestion and degradation by phagocytes.



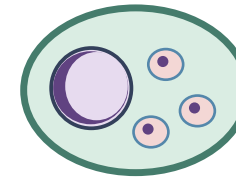
Shrinkage



Chromatin



Bodies

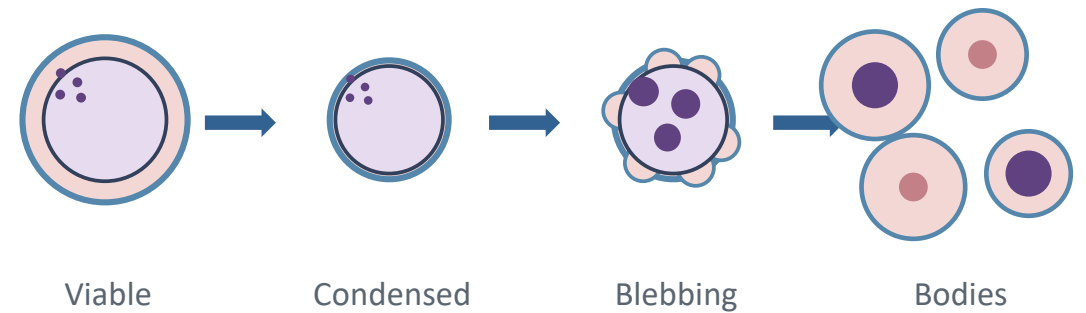


Removal

Schematic

The phenotype evolves, rather than appearing all at once

- Early: The cell contracts and chromatin becomes condensed.
- Intermediate: Surface blebs emerge, and the nucleus may fragment.
- Late: Apoptotic bodies are engulfed and degraded.
- The steps overlap; Not every snapshot displays all features.

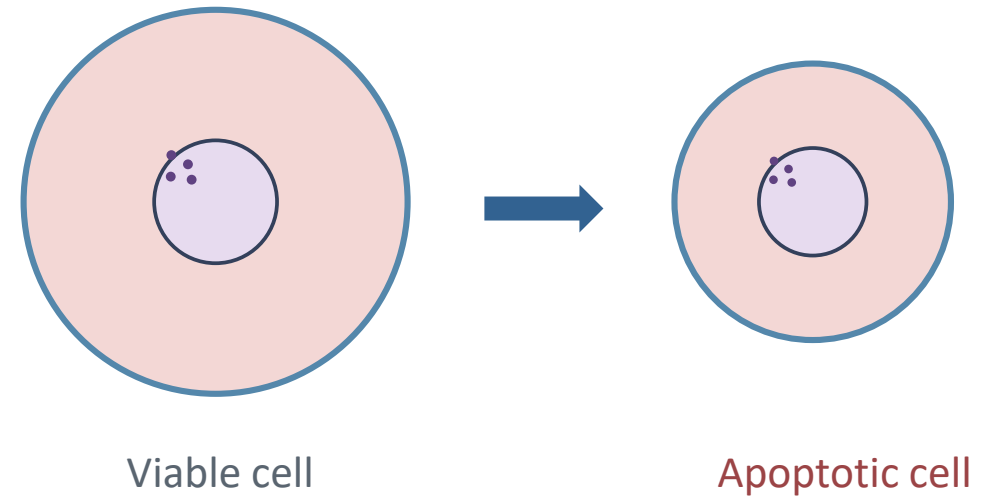


Schematic sequence; Stages may overlap

Schematic

Cell shrinkage is a cardinal early change

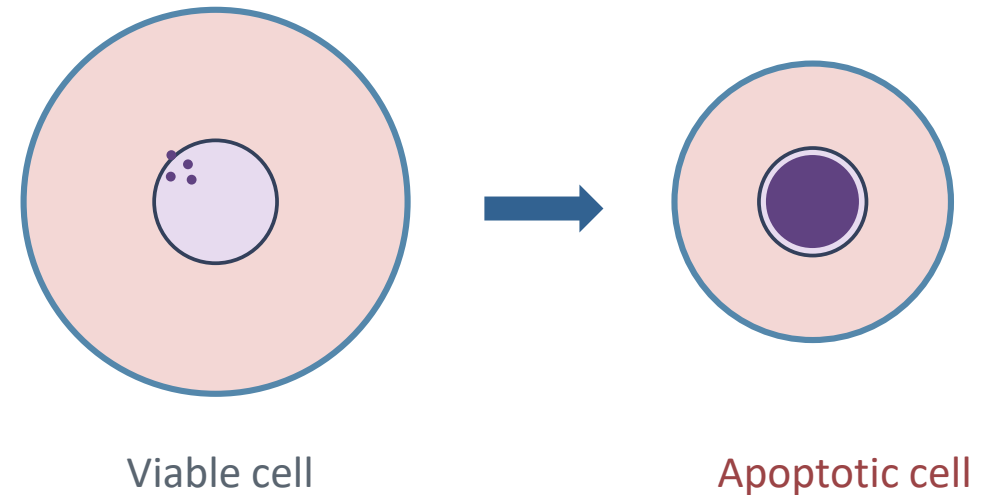
- The apoptotic cell becomes smaller than its neighbors.
- Loss of volume makes cellular constituents more concentrated.
- This is opposite to the early swelling characteristic of necrosis.



Schematic

Cytoplasm becomes dense and eosinophilic

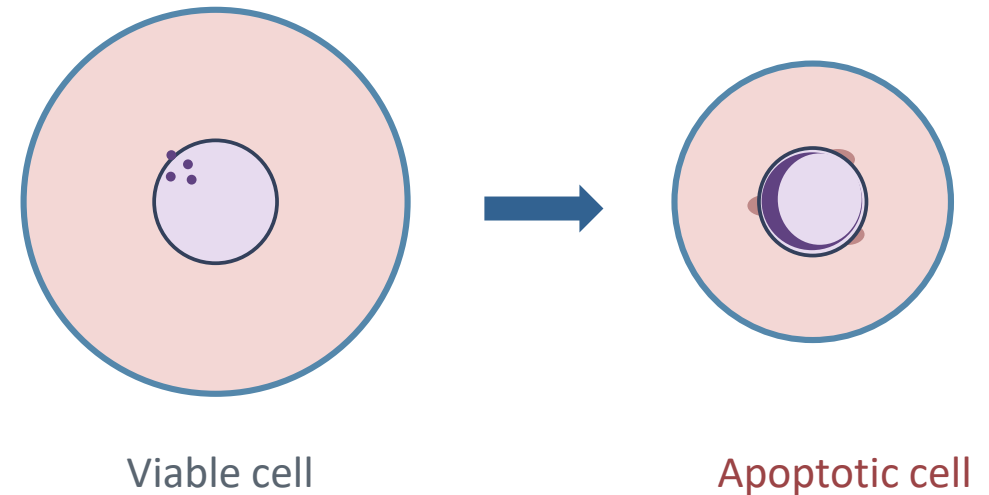
- Cytoplasmic compaction intensifies eosin staining in H&E sections.
- A round or oval, strongly pink cell profile may stand out from adjacent cells.
- Eosinophilia is a clue, not diagnostic alone.



Schematic

Organelles are tightly packed but relatively intact

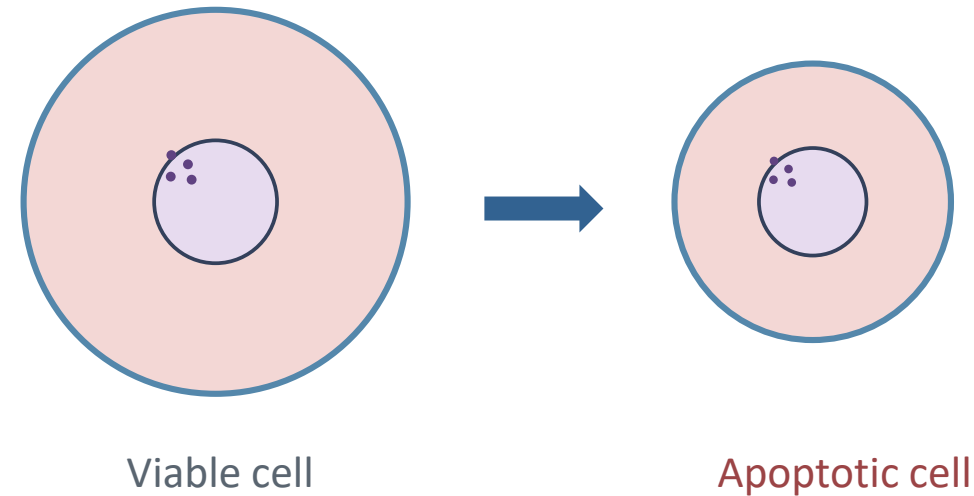
- Organelles as relatively normal during cell shrinkage.
- The cytoplasmic volume contracts around those organelles.
- Tightly packed organelles can become enclosed within apoptotic bodies.



Schematic

Shrinkage versus swelling is diagnostically useful

- Apoptosis: Reduced cell size and dense cytoplasm.
- Necrosis: Early cell and organelle swelling is typical.
- Interpret size together with nuclear changes and membrane fate.

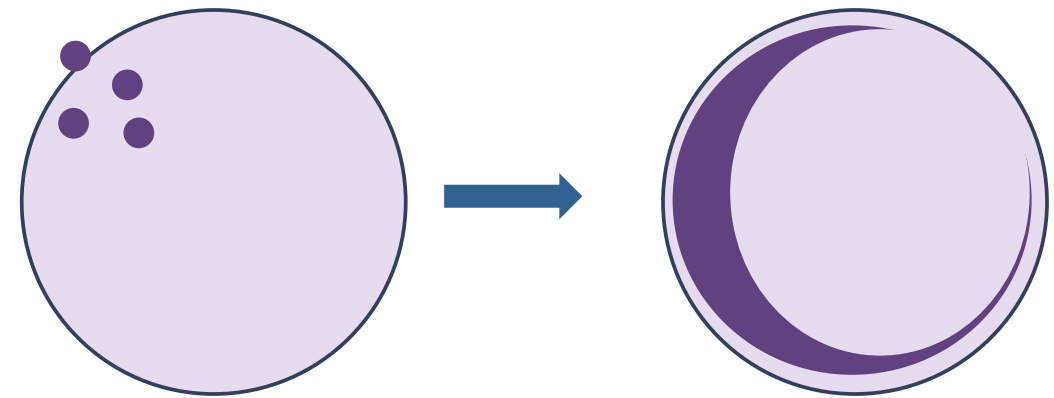


Opposing volume responses: Apoptosis $\neq$ necrosis

Schematic

Chromatin condensation is the most characteristic feature

- Chromatin coalesces into strikingly dense aggregates.
- These aggregates are much more compact than normal chromatin.
- Peripheral location beneath the nuclear envelope is particularly characteristic.



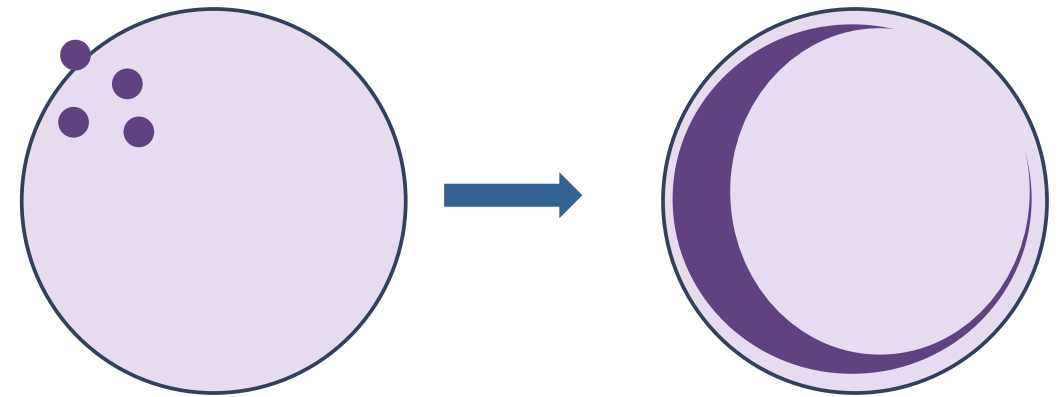
Dispersed chromatin

Peripheral compaction

Schematic

Chromatin shifts toward the nuclear periphery

- Condensed chromatin often accumulates immediately below the nuclear membrane.
- Under electron microscopy this can form a peripheral rim.
- The distribution adds specificity to the observation of a dark nucleus.



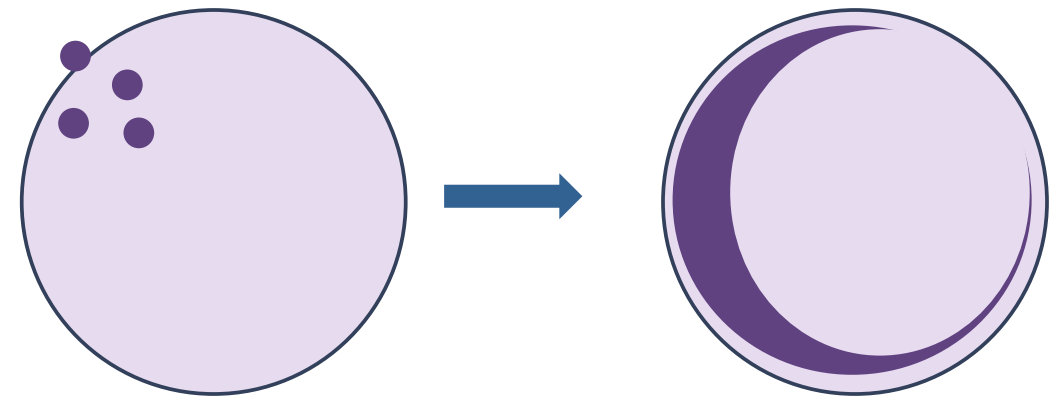
Dispersed chromatin

Peripheral compaction

Schematic

Peripheral crescents are a classic ultrastructural clue

- Compacted chromatin may form dense crescent-shaped masses.
- The masses differ in size and shape among affected cells.
- Robbins Fig. 2.15 describes these nuclear patterns by electron microscopy.



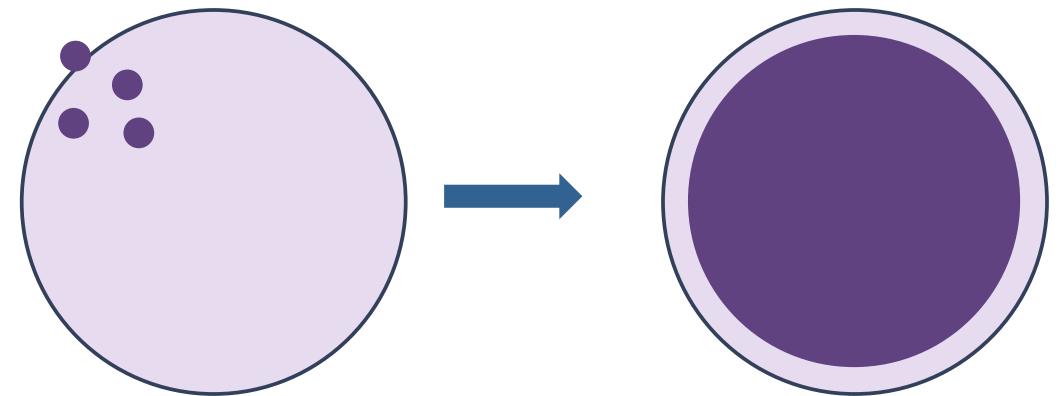
Dispersed chromatin

Peripheral compaction

Schematic

Some apoptotic nuclei become uniformly dense

- Peripheral chromatin crescents are not the only endpoint.
- The nuclear profile can appear almost uniformly electron-dense.
- Nuclear appearance varies with the stage of apoptosis.



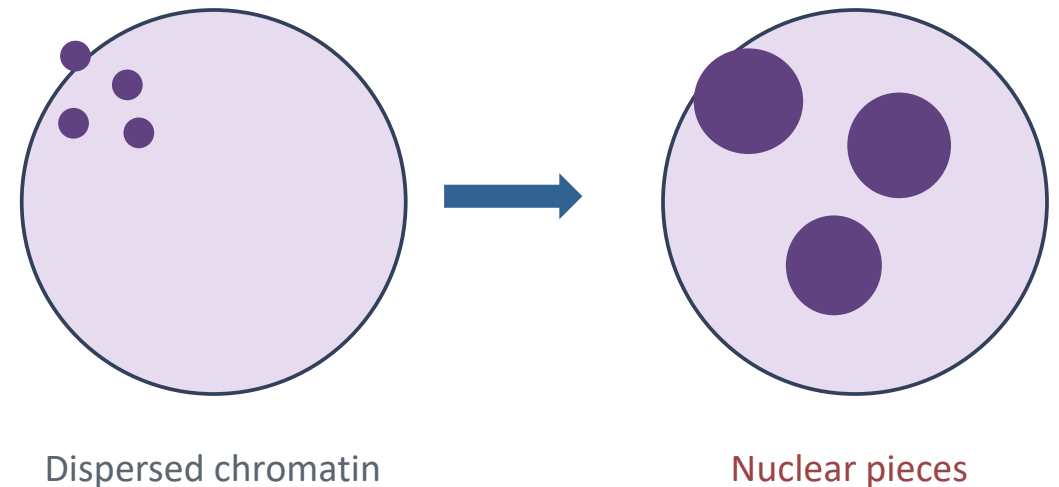
Dispersed chromatin

Uniformly dense

Schematic

The nucleus may fragment into multiple pieces

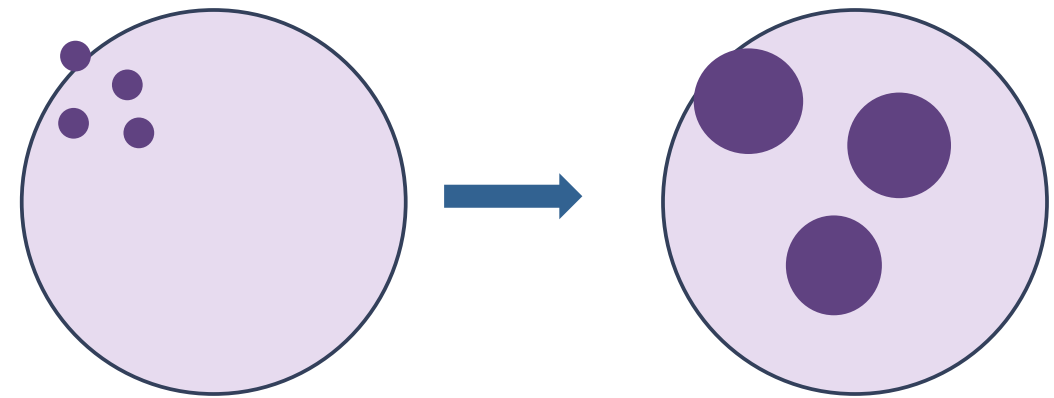
- Apoptosis may divide one condensed nucleus into two or more fragments.
- These pieces can remain within the apoptotic cell or enter apoptotic bodies.
- The fragments retain dense chromatin.



Schematic

Condensation and fragmentation are related but distinct

- Condensation describes how chromatin is packed.
- Fragmentation describes physical separation of the nucleus.
- A cell can show condensed chromatin before fragmentation is visible.



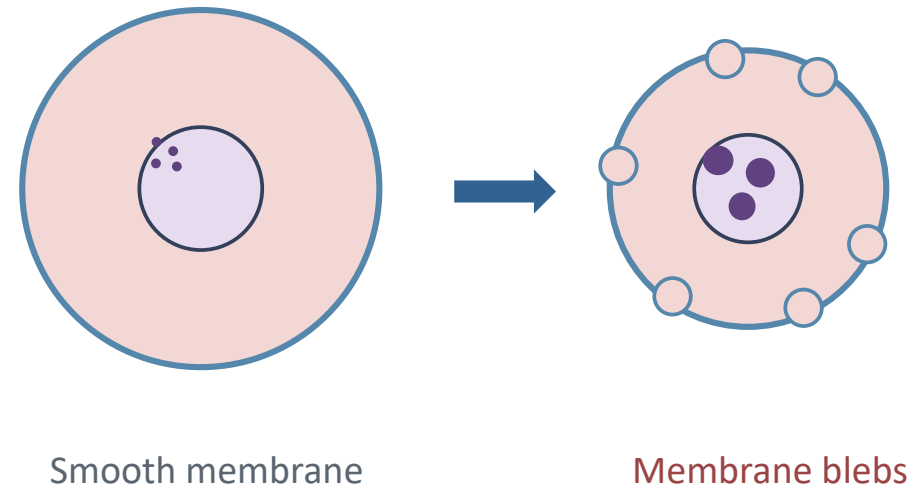
Dispersed chromatin

Fragmented nucleus

Schematic

Plasma membrane blebbing precedes body formation

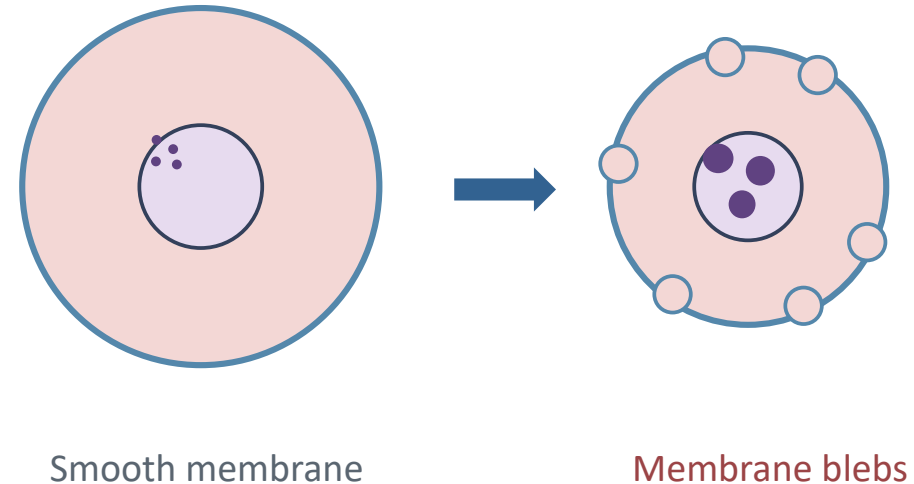
- The cell surface develops multiple outward membrane protrusions.
- Extensive blebbing precedes fragmentation into apoptotic bodies.
- Blebbing does not by itself imply lysis of the cell membrane.



Schematic

Blebs are membrane-enclosed protrusions

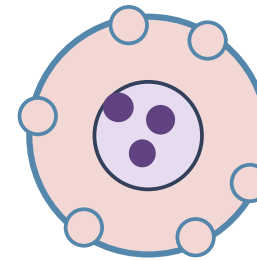
- A bleb is continuous with the plasma membrane of the parent cell.
- It is not the same as membrane rupture with release of contents.
- Apoptotic blebbing can be recognized with phase-contrast imaging.



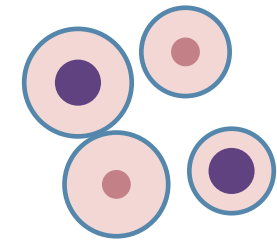
Schematic

Apoptotic bodies are discrete cellular fragments

- The parent cell separates into small membrane-bound pieces.
- Each fragment can be readily handled by a phagocyte.
- This packaging minimizes extracellular release of cellular contents.



Blebbing cell

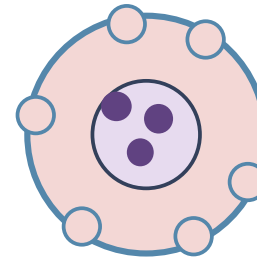


Membrane-bound
bodies

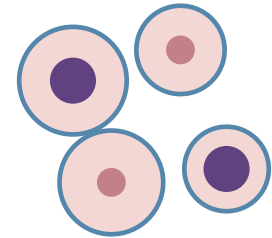
Schematic

Apoptotic bodies contain packed cell material

- Bodies may contain cytoplasm and closely packed organelles.
- Some also contain fragments of nuclear chromatin.
- A nuclear fragment is not required in every apoptotic body.



Blebbing cell

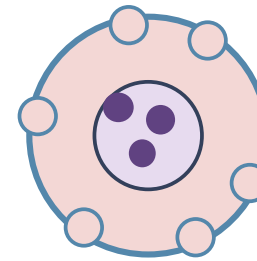


Membrane-bound
bodies

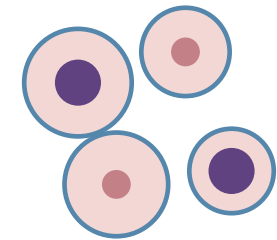
Schematic

Nuclear material varies among apoptotic bodies

- Some fragments carry prominent condensed DNA-containing nuclear pieces.
- Others contain cytoplasm and organelles without visible nuclear fragments.
- Avoid equating every pink globule with a nuclear fragment.



Blebbing cell

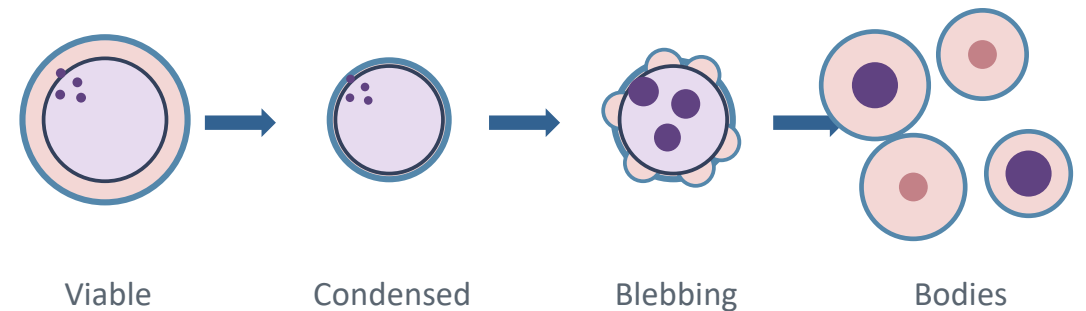


Membrane-bound
bodies

Schematic

A structural sequence links all four findings

- Cell contraction → chromatin compaction → membrane blebs → apoptotic bodies.
- Phagocytes remove the fragments before extensive membrane breakdown.
- The progression is a teaching model, not a rigid time scale.

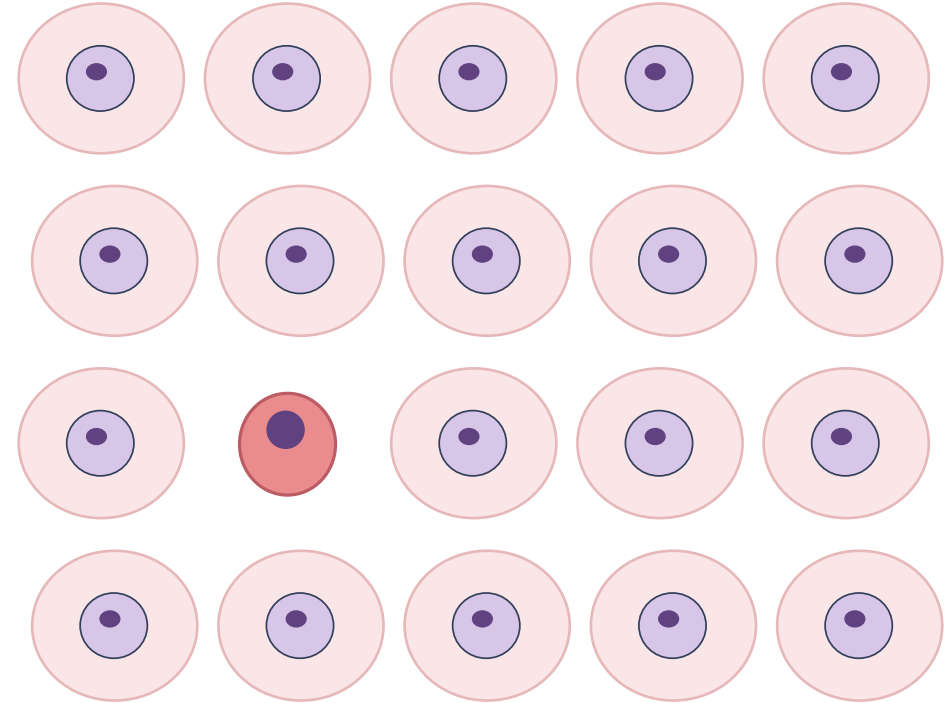


Schematic sequence; Stages may overlap

Schematic

H&E microscopy: The typical apoptotic profile

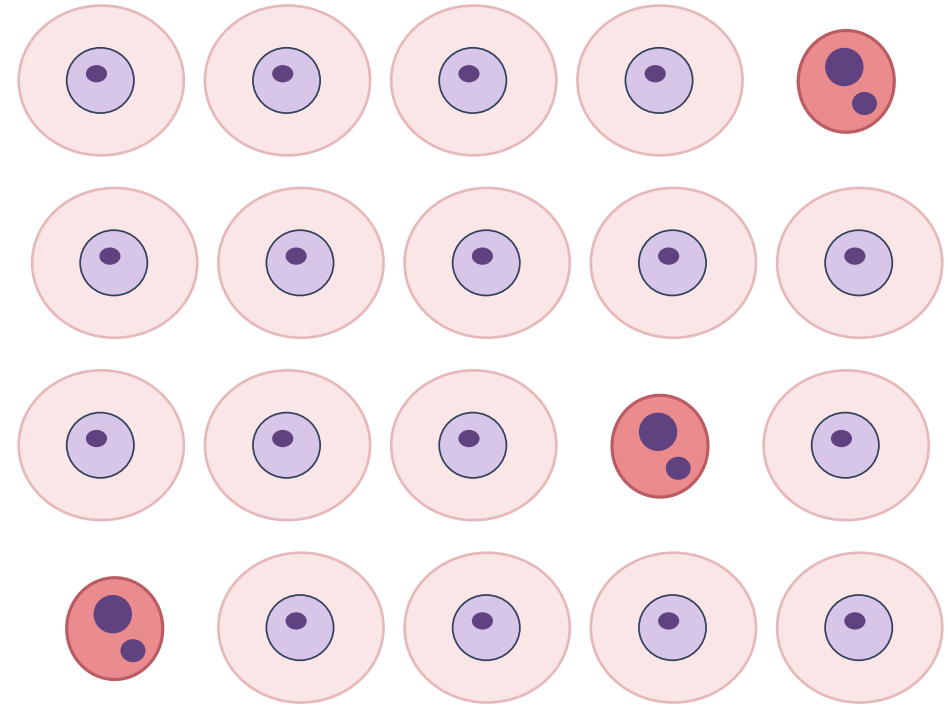
- Round or oval intensely eosinophilic cytoplasmic mass.
- Small condensed nuclear material, sometimes fragmented.
- Frequently surrounded by viable-appearing adjacent cells.



H&E-like tissue schematic — not a photomicrograph

H&E microscopy: Looking for a single dying cell

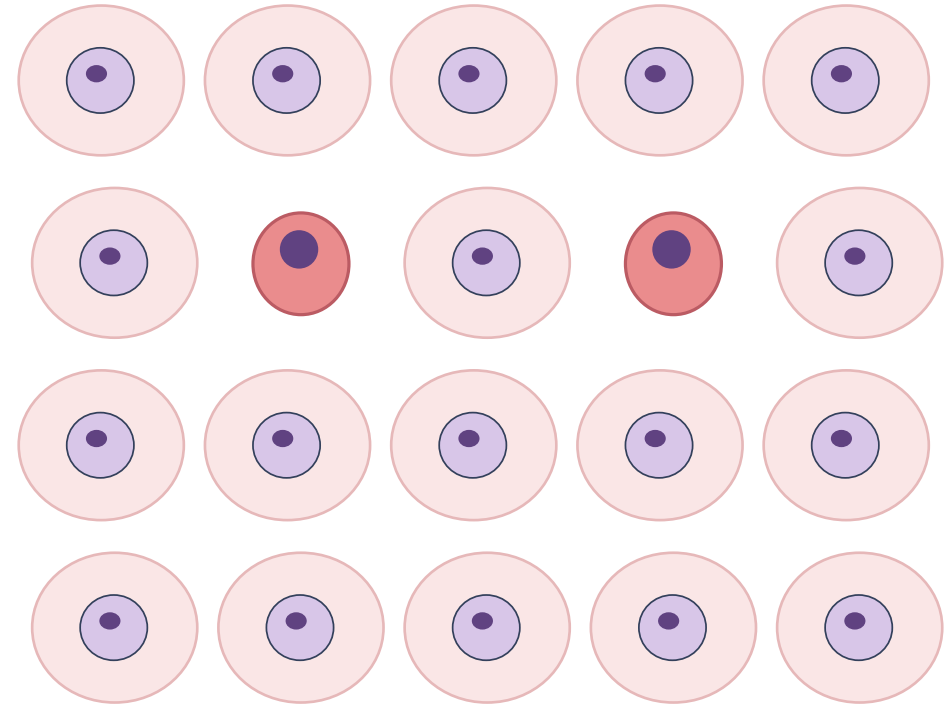
- Scan for isolated small bright-pink bodies among larger cells.
- Compare nuclear staining intensity and fragmentation.
- Examine the immediate surroundings for an inflammatory reaction.



H&E-like tissue schematic — not a photomicrograph

Cytoplasmic and nuclear clues work together

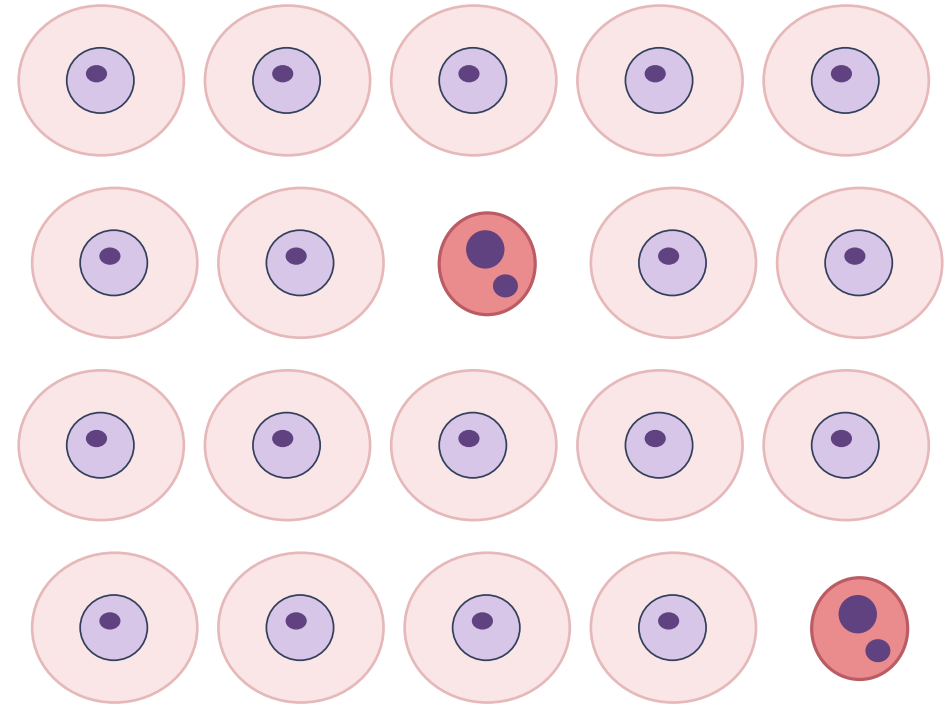
- Intense eosinophilia alone is nonspecific.
- Highly condensed chromatin alone is also insufficient.
- A reduced cellular profile with both changes strongly supports apoptosis.



H&E-like tissue schematic — not a photomicrograph

Nuclear fragments may be inconspicuous in H&E

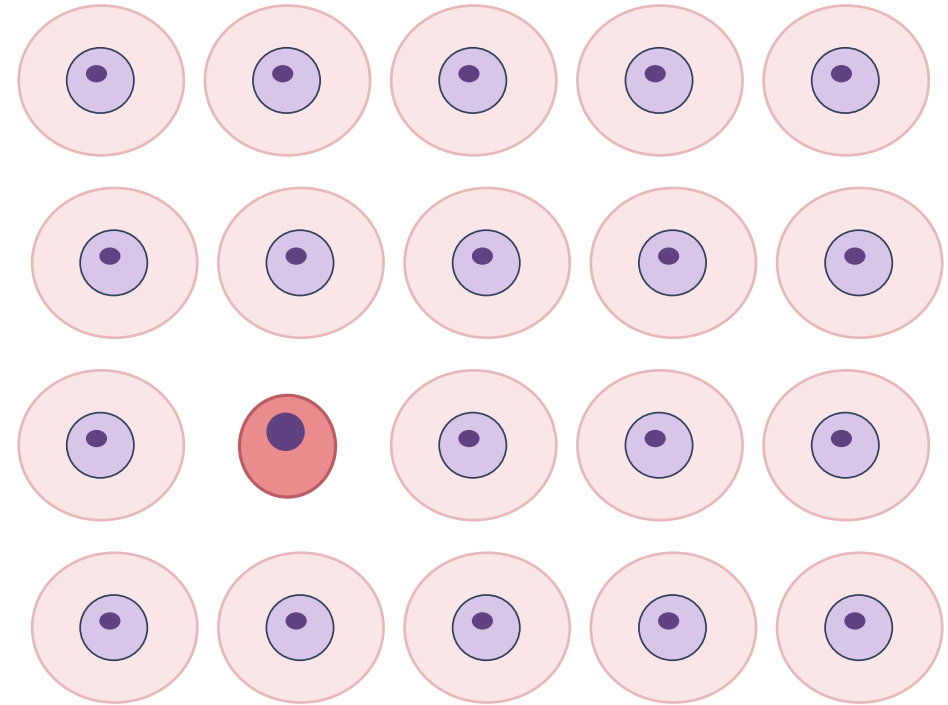
- Some apoptotic profiles show several small dense basophilic pieces.
- Other profiles lack a recognizable nucleus in the plane of section.
- Sectioning geometry limits what can be observed in any one cell.



H&E-like tissue schematic — not a photomicrograph

Apoptosis often occurs in scattered cells

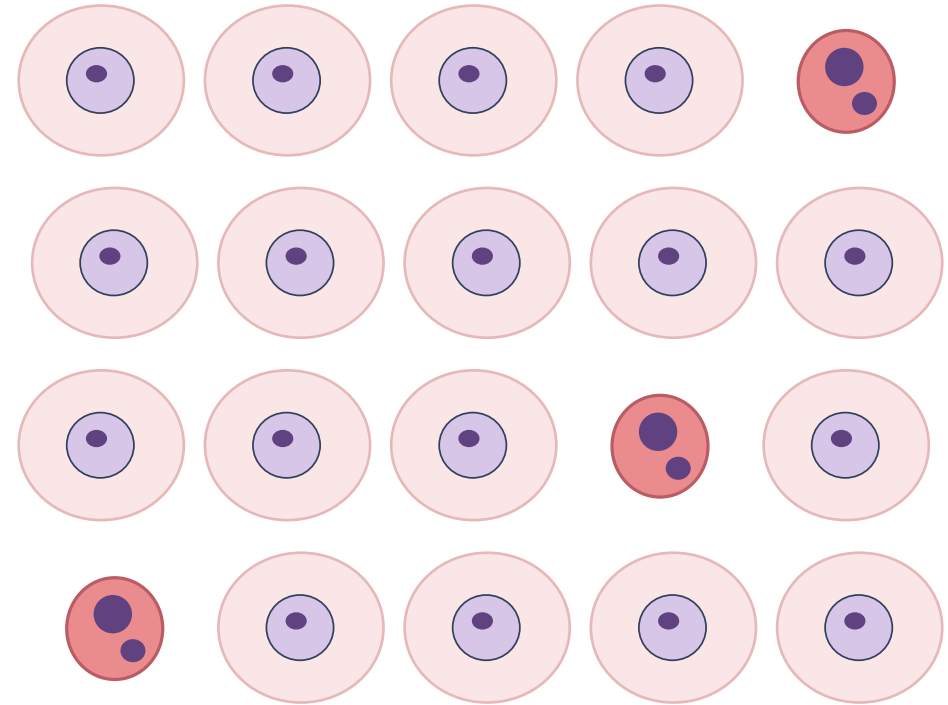
- Affected cells may be separated by morphologically preserved neighbors.
- The pattern contrasts with confluent destruction common in many necrotic lesions.
- Spatial pattern supports—but does not establish—the mechanism.



H&E-like tissue schematic — not a photomicrograph

Tissue can appear relatively preserved

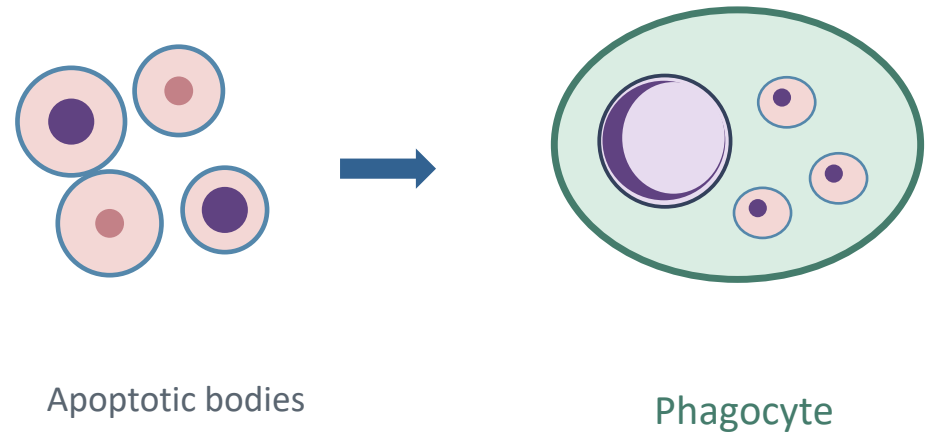
- Rapid removal of single apoptotic cells may leave little visible residue.
- The surrounding tissue need not show broad areas of cytoplasmic leakage.
- A focal snapshot can underestimate cumulative cellular loss.



H&E-like tissue schematic — not a photomicrograph

Phagocytic uptake is part of the morphologic phenotype

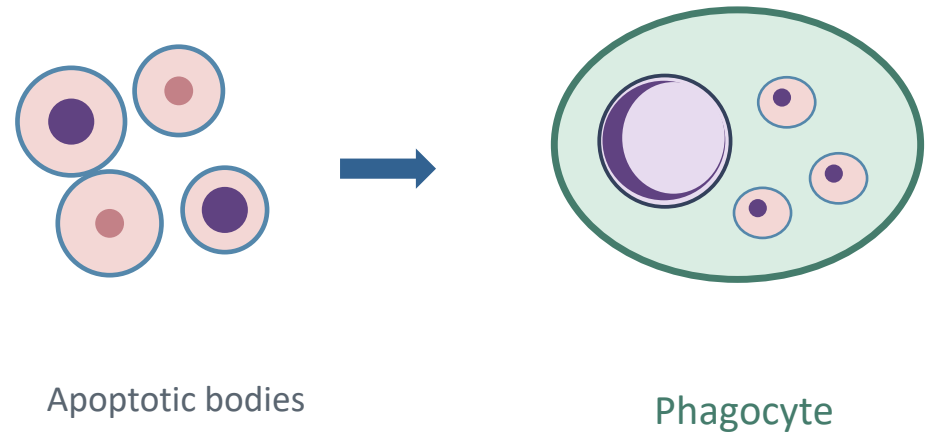
- Apoptotic cells and bodies are typically engulfed by phagocytes.
- Macrophages as frequent phagocytic cells.
- Removal follows fragmentation and may occur very quickly.



Schematic

Apoptotic material is degraded inside phagocytes

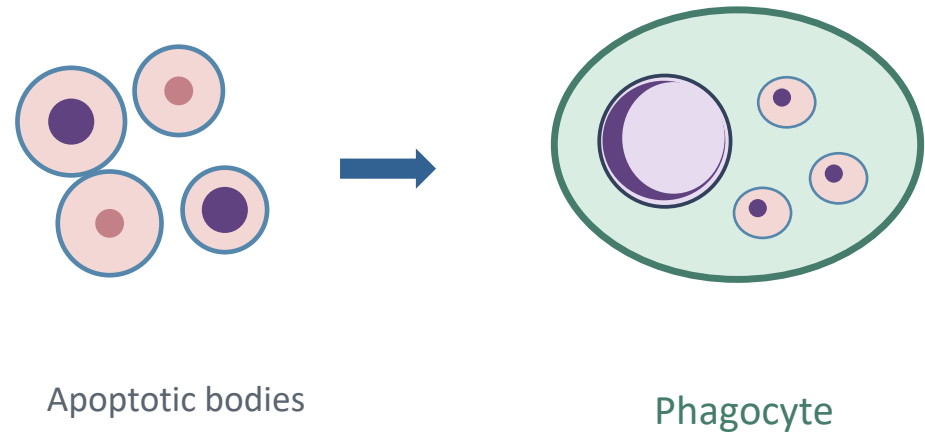
- Engulfed bodies enter the phagocytic compartment.
- Lysosomal enzymes break down the ingested cellular material.
- Intracellular disposal limits exposure of neighboring tissue to cell contents.



Schematic

Clearance changes what the pathologist sees

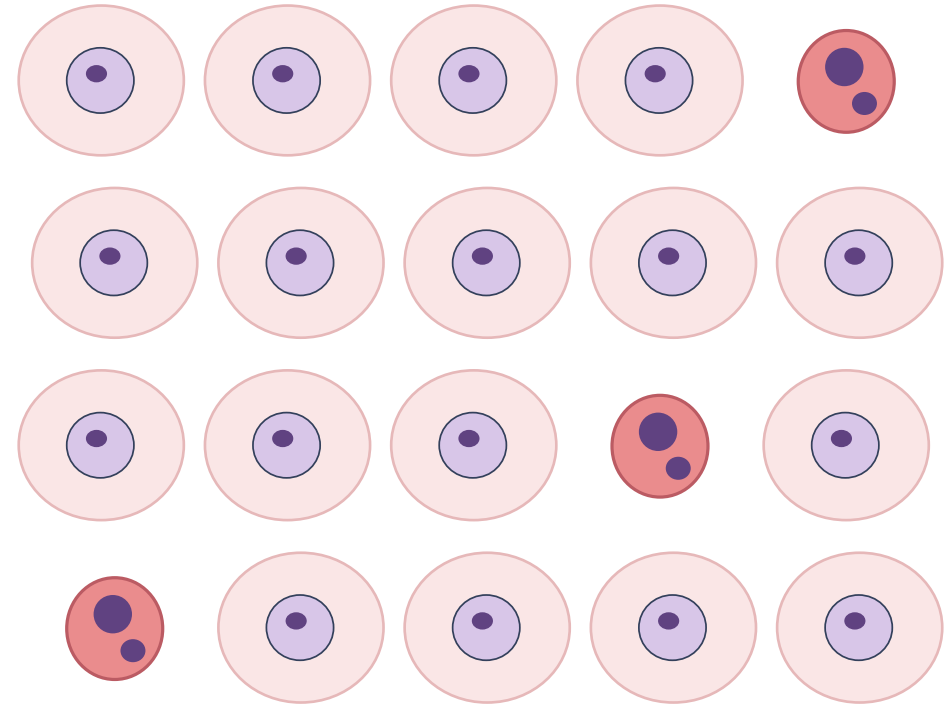
- An apoptotic cell may be visible only transiently.
- Extensive apoptosis need not produce many visible apoptotic figures.
- Morphologic counts represent a dynamic balance of formation and removal.



Schematic

Why a routine biopsy may miss active apoptosis

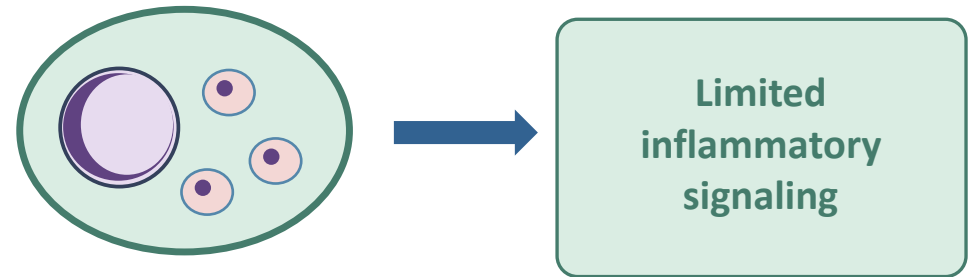
- Shrinkage, fragmentation, and clearance occur rapidly.
- Apoptotic bodies are easily overlooked in crowded tissue.
- Lack of accompanying inflammation removes a conspicuous contextual clue.



H&E-like tissue schematic — not a photomicrograph

Minimal inflammation is a distinguishing feature

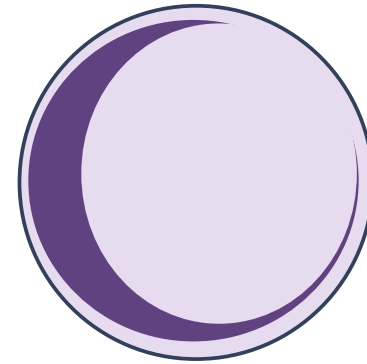
- Apoptotic cells generally do not provoke substantial inflammation.
- Membrane-bounded fragments are removed before their contents spill.
- Phagocytes that ingest apoptotic cells also reduce proinflammatory signaling.



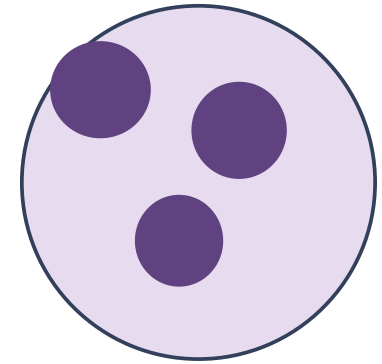
Schematic

Electron microscopy reveals chromatin topography

- Peripheral crescents of dense chromatin are best appreciated ultrastructurally.
- Other cells may show fully dense or fragmented nuclei.
- Electron microscopy clarifies structure but is not usually needed for routine diagnosis.



Peripheral crescent



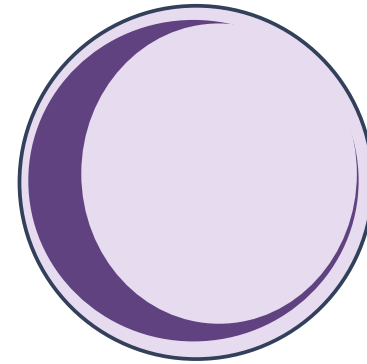
Fragmented nucleus

Ultrastructural nuclear patterns — schematic

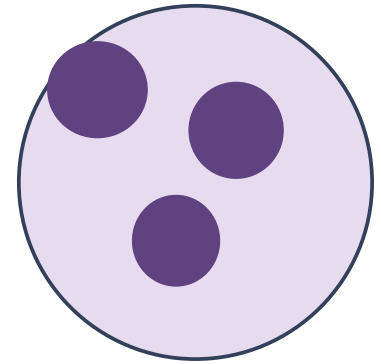
Schematic

Electron microscopy: Preserved packaging

- Shrinkage compacts relatively preserved organelles.
- Cell surfaces can form blebs and discrete bounded pieces.
- Ultrastructure can separate bounded fragmentation from uncontained breakdown.



Peripheral crescent



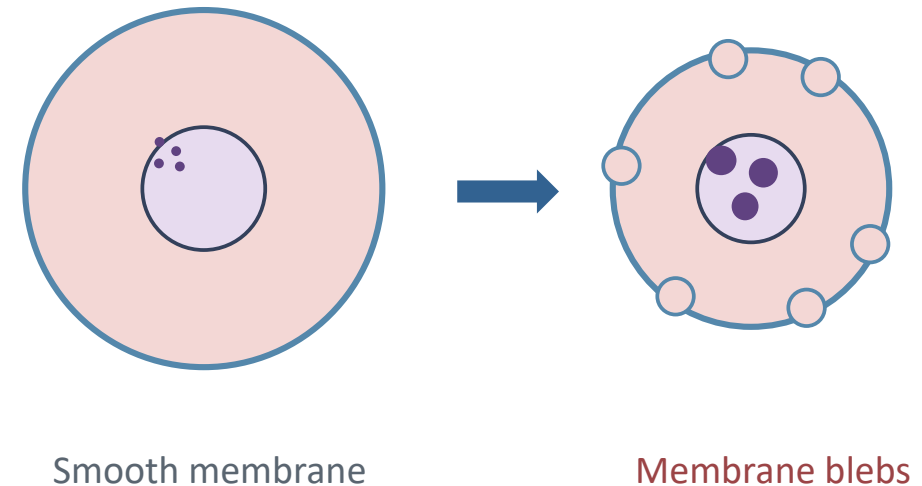
Fragmented nucleus

Ultrastructural nuclear patterns — schematic

Schematic

Phase-contrast microscopy demonstrates blebbing

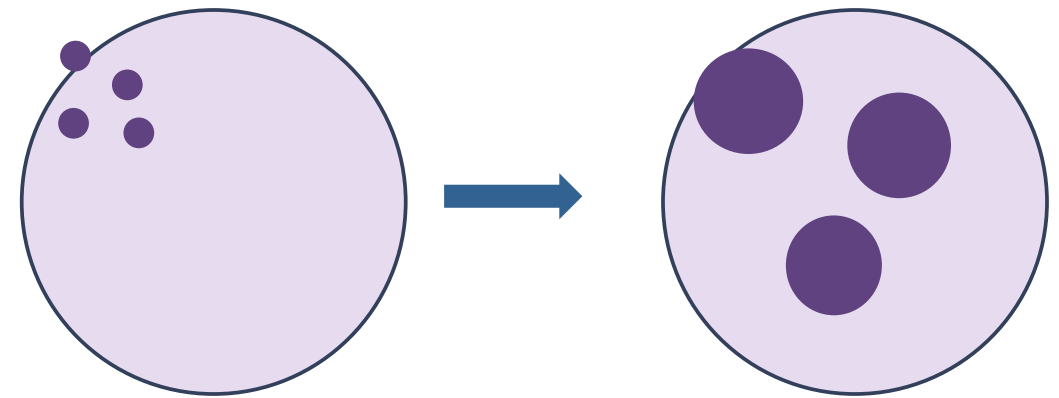
- Live-cell imaging can show irregular surface protrusions.
- Blebs can progress to detached apoptotic bodies.
- Time-resolved observation captures events missed by fixed tissue sections.



Schematic

DNA stains demonstrate nuclear fragmentation

- DNA-directed stains can reveal separated nuclear masses.
- Interpret nuclear fragmentation alongside the cell outline and membrane changes.
- A nuclear stain alone does not establish that the cell underwent apoptosis.



Dispersed chromatin

Separated DNA signals

Schematic

Active caspase-3 marks biochemical execution

- Robbins Fig. 2.15 illustrates immunofluorescence for active caspase-3.
- Enzyme activation is a molecular readout complementary to morphology.
- Positive staining should be interpreted in assay and tissue context.



Conceptual immunofluorescence — not acquired microscopy

Microscopy modalities answer different questions

- H&E: Location, cell shrinkage, cytoplasmic eosinophilia.
- EM: Peripheral condensed chromatin and ultrastructural packaging.
- Phase contrast / DNA stain / active caspase-3: Dynamic, nuclear, and enzyme-level evidence.

H&E

Cell & tissue

EM

Nuclear
topology

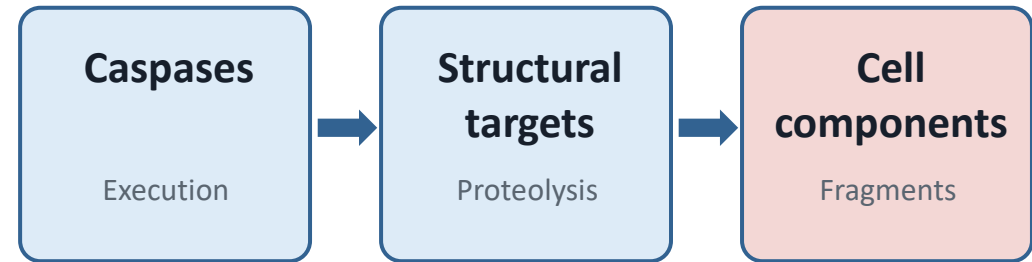
Biomarkers

Enzyme/DNA

Schematic

Caspases are the core apoptotic proteases

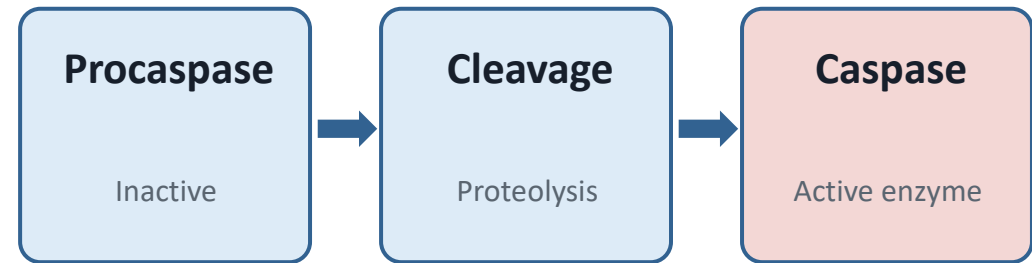
- Caspases are cysteine proteases that cleave proteins after aspartate residues.
- They exist as inactive precursors that require activation.
- Activated caspases drive the biochemical execution of apoptosis.



Schematic

Activation is controlled by proteolytic processing

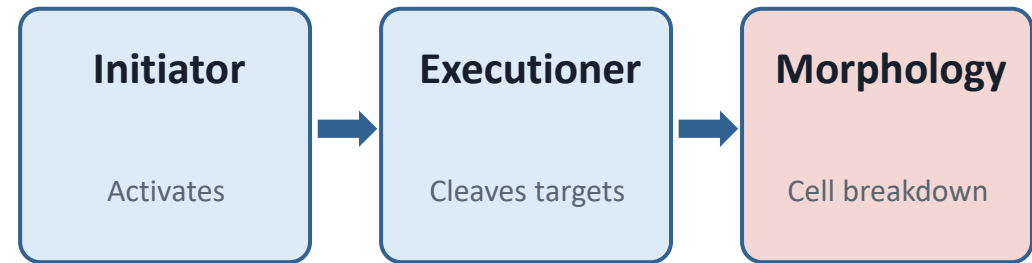
- Inactive procaspases are converted into catalytically active enzymes.
- Sequential caspase activation amplifies the death program.
- Detection of active enzymes is more informative than total protein abundance.



Schematic

Initiation and execution are separable biochemical phases

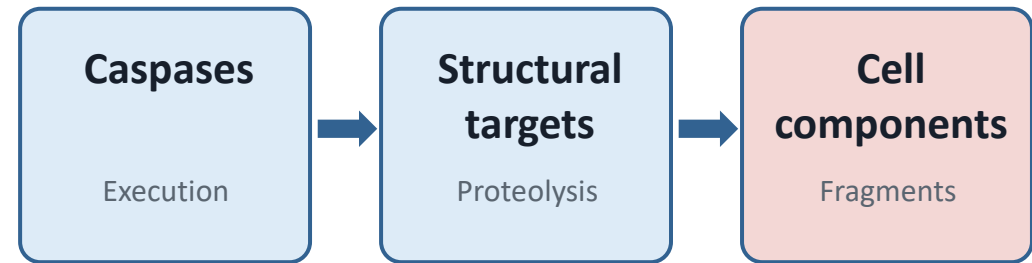
- Initiator caspases activate downstream proteases.
- Executioner caspases modify nuclear and cytoplasmic substrates.
- The present topic focuses on those downstream structural effects.



Schematic

Executioner caspases produce the final phenotype

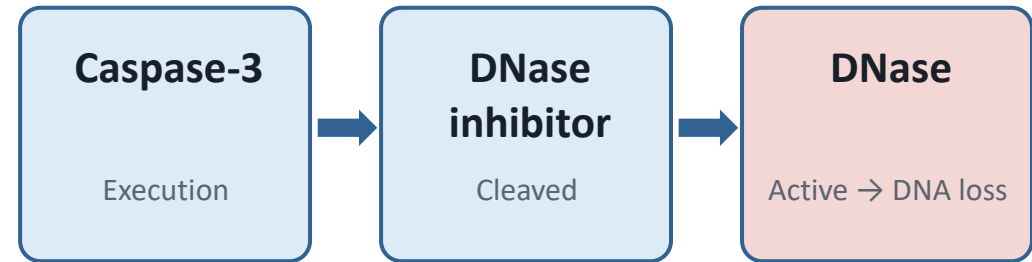
- Caspase-3 and caspase-6 are examples given by Robbins.
- They act on multiple cellular structural proteins and regulatory substrates.
- The coordinated biochemical changes culminate in fragmentation.



Schematic

Caspases activate a DNA-degrading nuclease

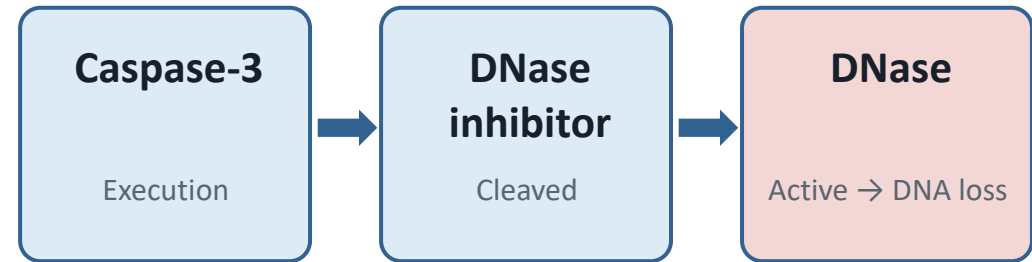
- An executioner caspase cleaves an inhibitory protein bound to a DNase.
- Removing the inhibitor allows the DNase to degrade DNA.
- This explains an important biochemical component of nuclear destruction.



Schematic

DNA degradation contributes to nuclear dismantling

- Active DNase cleaves cellular DNA.
- Structural nuclear changes are also needed for visible nuclear fragmentation.
- Fragmented DNA and fragmented nuclei are related but different endpoints.



Schematic

Nuclear matrix proteins are caspase substrates

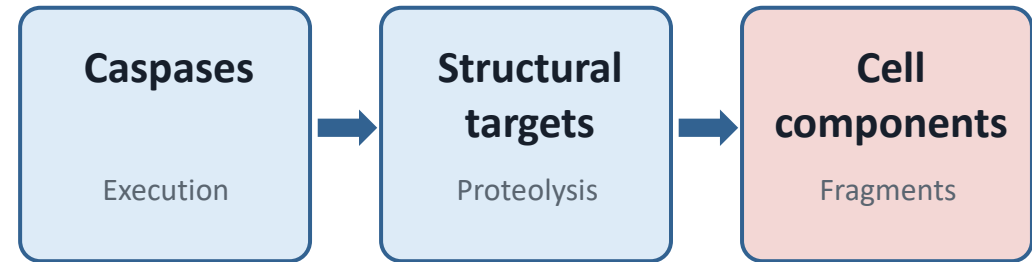
- Executioner caspases proteolyze structural components of the nuclear matrix.
- Loss of scaffold integrity favors disassembly of nuclear architecture.
- This contributes to physical nuclear fragmentation.



Schematic

Proteolysis alters cell architecture

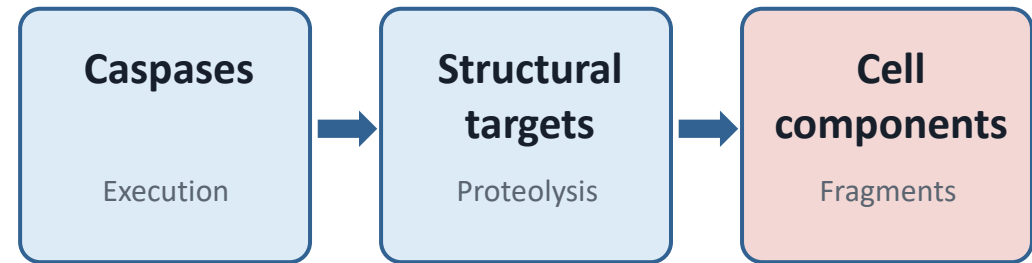
- Caspases also act on components of the cytoskeleton.
- Structural breakdown is associated with the reorganization of dying cells.
- Some details of bleb and body formation remain unresolved.



Schematic

Cell shape reflects coordinated biochemical events

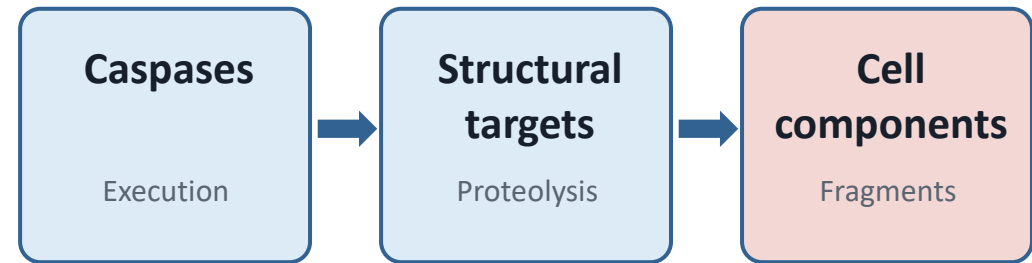
- Nuclear substrate cleavage contributes to chromatin/nuclear breakdown.
- Cytoplasmic substrate cleavage contributes to structural remodeling.
- The observable phenotype is produced by many targets, not a single reaction.



Schematic

Mechanism and morphology should not be conflated

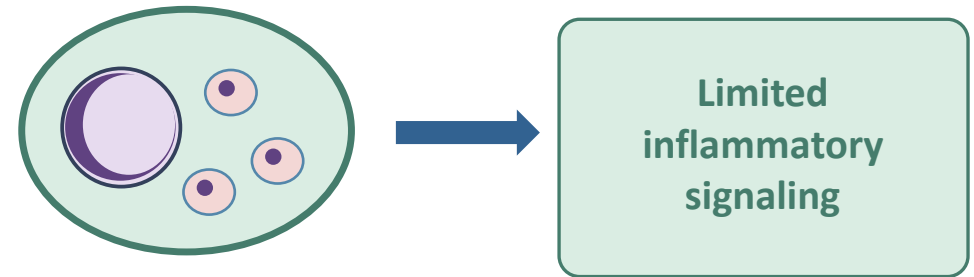
- Condensation and blebbing are observational features.
- Caspase activation and DNase activation are biochemical processes.
- An assay for one feature does not automatically prove all others occurred.



Schematic

Membrane integrity supports noninflammatory removal

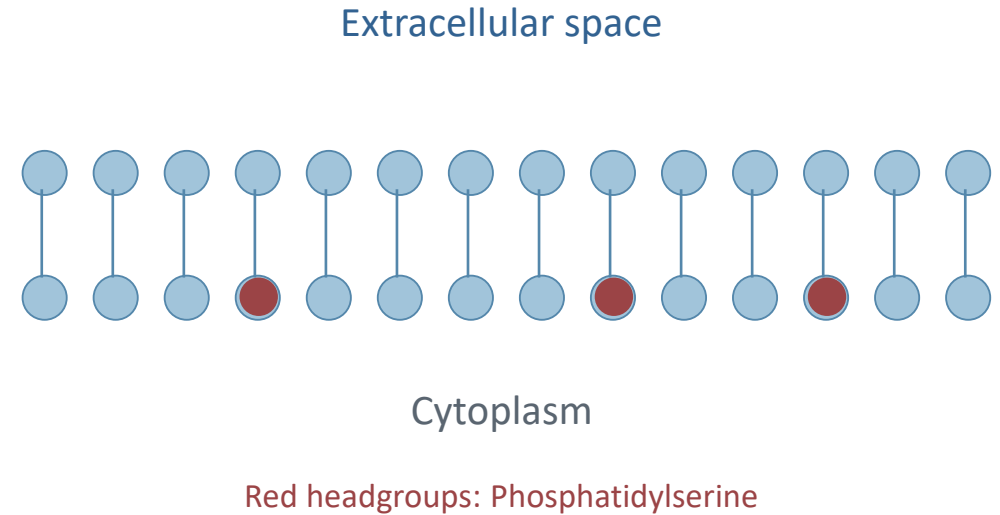
- Apoptotic bodies retain delimiting membranes during early clearance.
- Phagocytes engulf material before widespread release of contents.
- Membrane integrity is time-dependent; Late uncleared apoptotic cells may deteriorate.



Schematic

Phosphatidylserine is normally on the inner leaflet

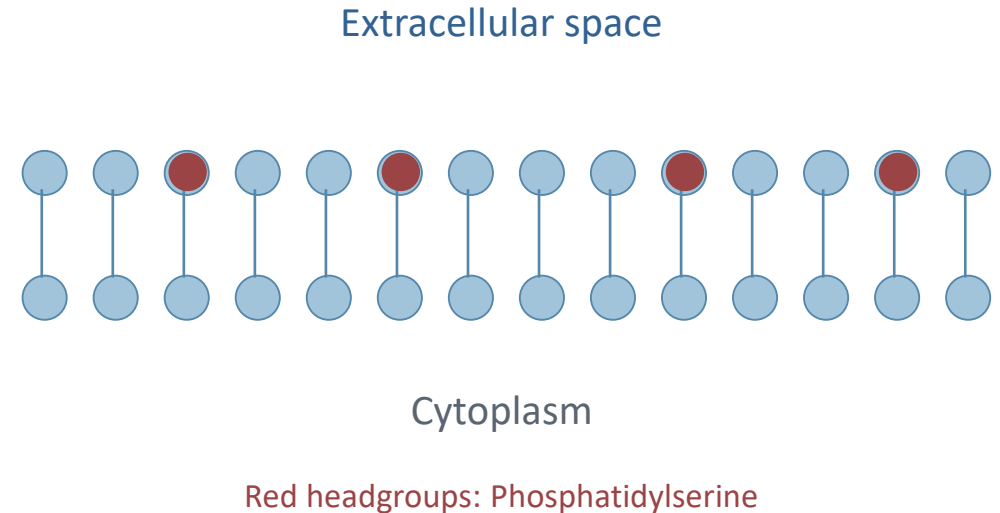
- In viable cells, phosphatidylserine is concentrated on the cytoplasmic face.
- This asymmetric distribution changes in dying apoptotic cells.
- The topologic switch supplies a recognition signal.



Schematic

Apoptosis exposes phosphatidylserine at the surface

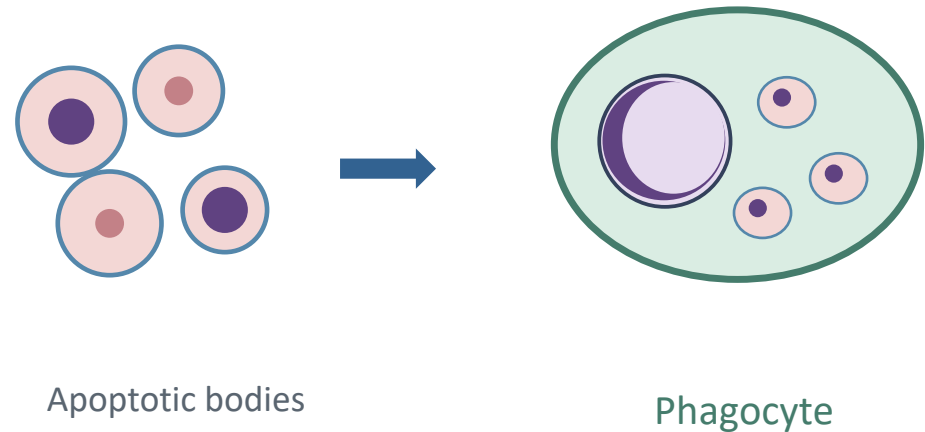
- Phosphatidylserine becomes displayed on the outer membrane leaflet.
- External phosphatidylserine is an “eat-me” cue for phagocytes.
- The lipid signal is a biochemical change with a direct disposal function.



Schematic

Phagocytes recognize exposed surface cues

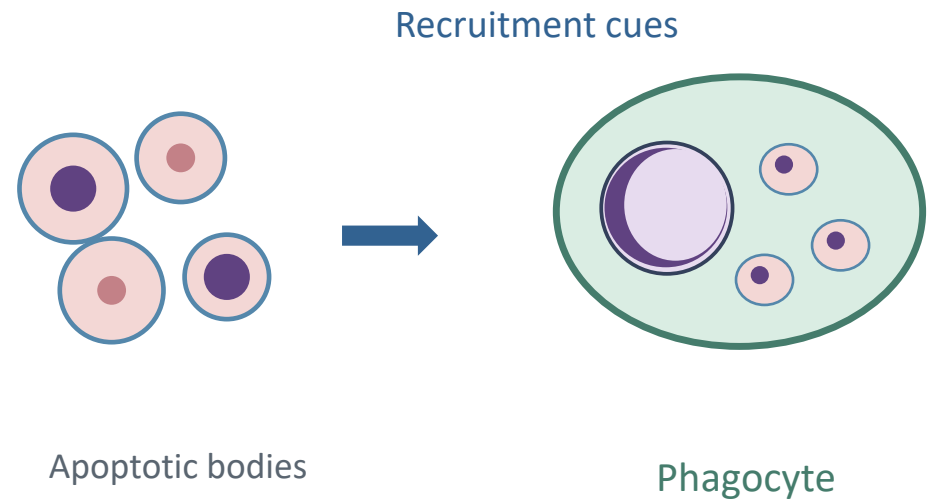
- Macrophage receptors bind apoptotic surface ligands directly or indirectly.
- Surface changes distinguish dying cells from healthy neighbors.
- Recognition initiates engulfment rather than extracellular disintegration.



Schematic

Soluble factors help recruit phagocytes

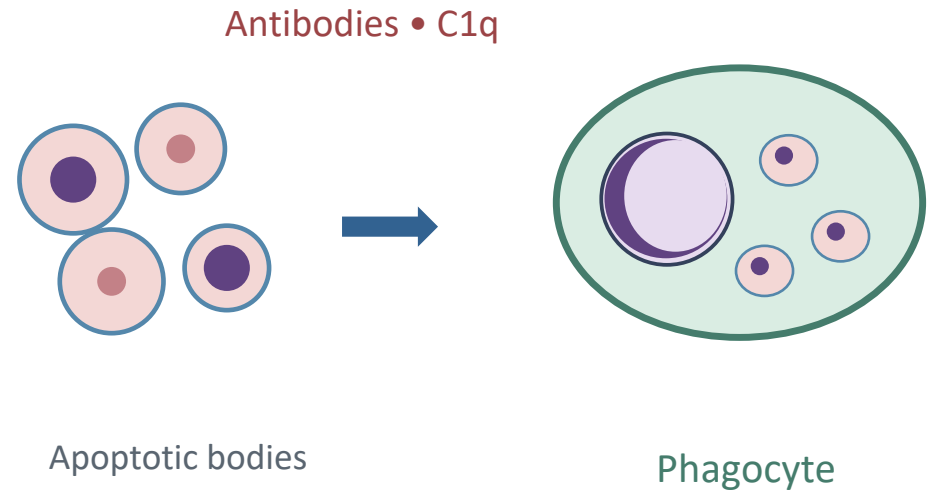
- Dying cells may release signals that attract phagocytes.
- These cues bring scavenger cells near apoptotic targets.
- The exact molecules are not specified in the assigned Robbins passage.



Schematic

Opsonins can further promote engulfment

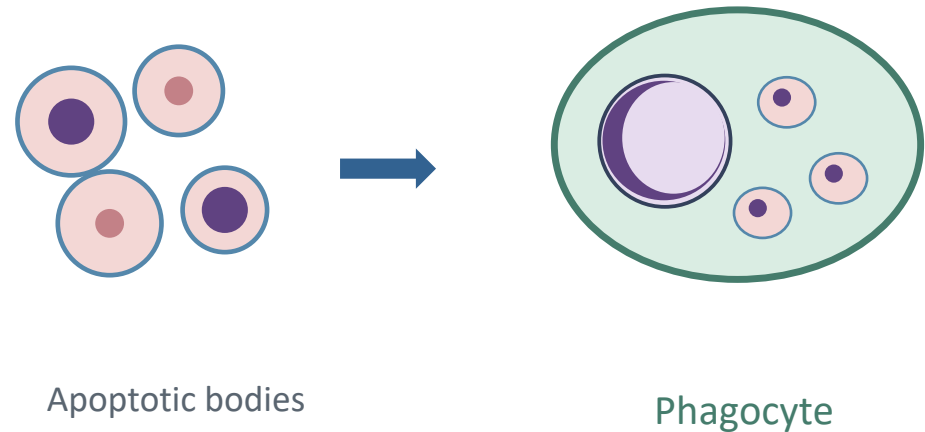
- Natural antibodies and complement proteins can coat apoptotic fragments.
- The complement component C1q.
- Coated targets become recognizable to appropriate phagocytic receptors.



Schematic

Efferocytosis is removal of apoptotic cells

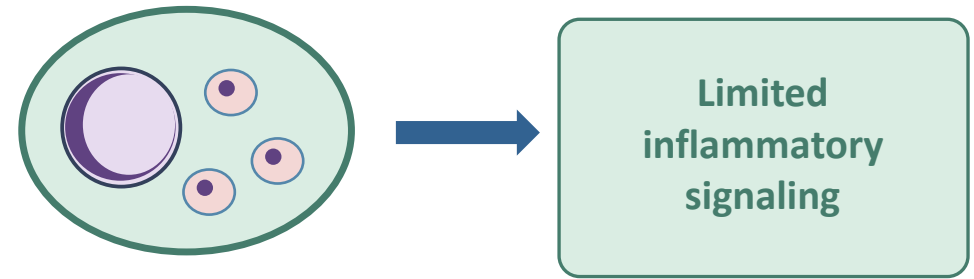
- Efferocytosis denotes phagocytic clearance of apoptotic cells and fragments.
- Membrane changes, recruitment cues, and opsonic signals work together.
- The process is efficient enough for dead cells to vanish without a trace.



Schematic

Efferocytosis suppresses inflammatory signaling

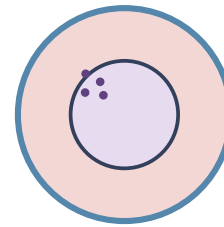
- Macrophages that ingest apoptotic material reduce proinflammatory cytokine production.
- Rapid containment and clearance limit local inflammatory injury.
- This biochemical behavior helps explain the quiet histologic picture.



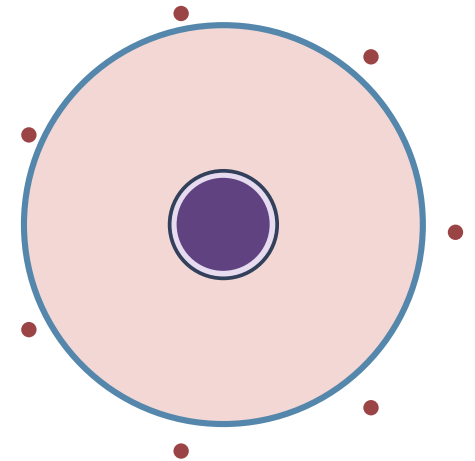
Schematic

Apoptosis and necrosis differ at the cell surface

- Apoptosis: Shrinkage, bounded blebs/bodies, prompt removal.
- Necrosis: Swelling followed by membrane damage and leakage.
- These are typical patterns; Tissue context and timing still matter.



Apoptosis

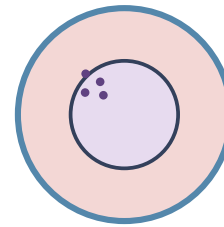


Necrosis

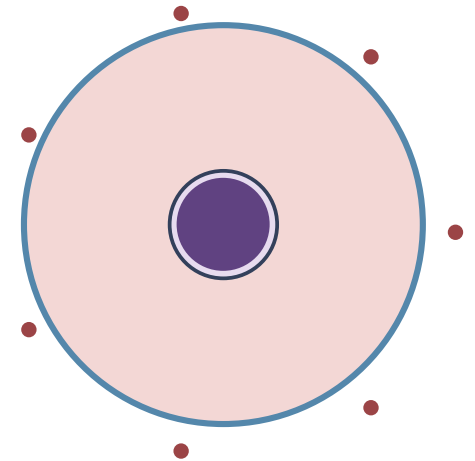
Schematic

Nuclear and inflammatory distinctions aid interpretation

- Apoptosis favors chromatin compaction and controlled fragmentation.
- Necrosis can show nuclear loss during cellular disintegration.
- Prominent inflammation favors necrotic injury but is not absolute.



Apoptosis



Necrosis

Schematic

Diagnostic synthesis: Use convergent findings

- Seek small eosinophilic cells with dense, often fragmented chromatin.
- Look for bounded apoptotic bodies and phagocytic clearance.
- Use biochemical markers selectively as supporting evidence.

H&E

Cell & tissue

EM

Nuclear
topology

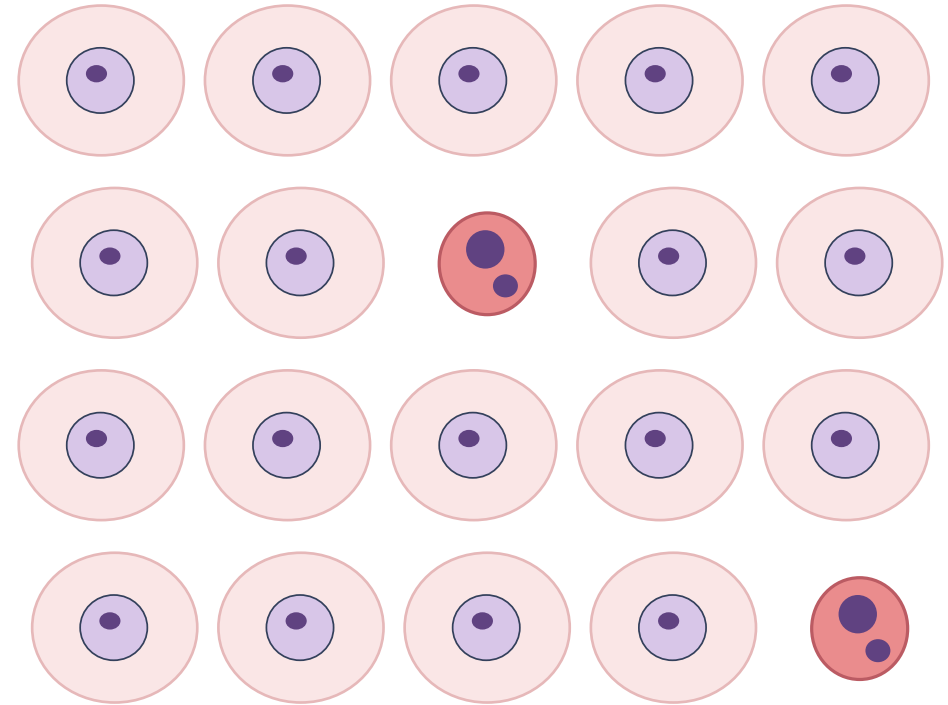
Biomarkers

Enzyme/DNA

Schematic

Application: Identify the mechanism

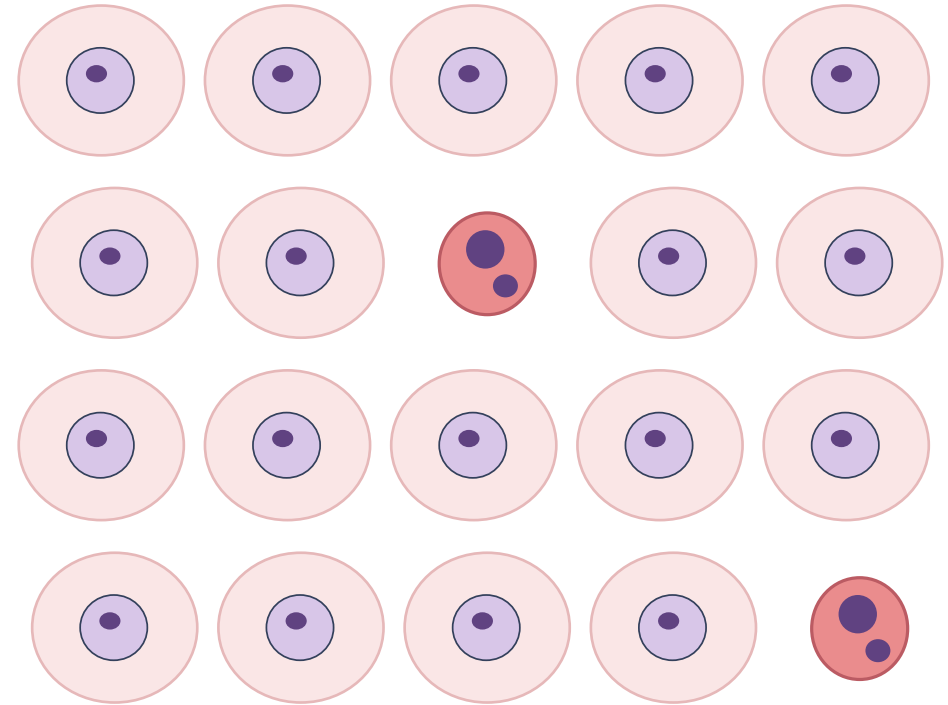
- A biopsy contains scattered contracted intensely pink cells with multiple dark nuclear fragments. Adjacent tissue is preserved.
- Question: Which form of cell death best explains this combined phenotype?



H&E-like tissue schematic — not a photomicrograph

Application: Interpretation

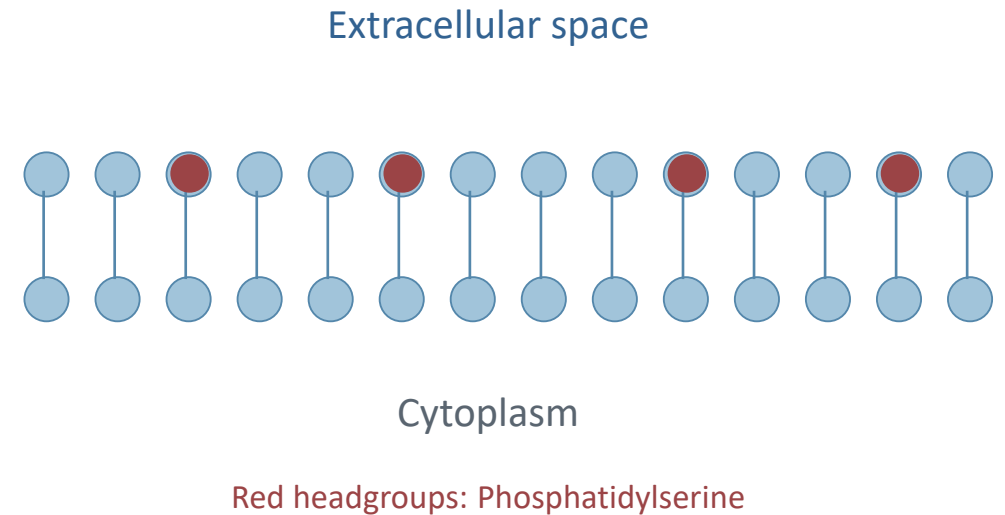
- Apoptosis is the best morphologic interpretation.
- Supporting findings: Shrinkage, eosinophilia, fragmented condensed chromatin, isolated distribution.
- Confirm that there is not prominent membrane rupture with widespread inflammation.



H&E-like tissue schematic — not a photomicrograph

Application: Connect biochemistry to disposal

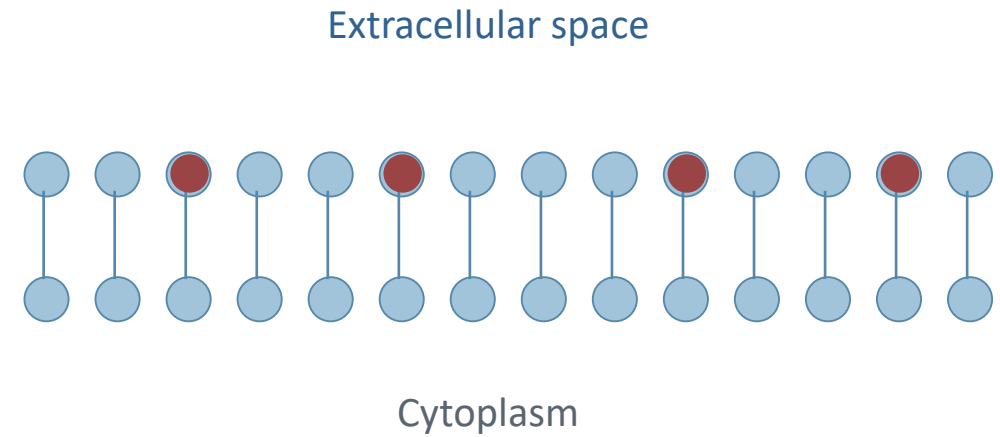
- A dying cell displays phosphatidylserine on its external membrane and is rapidly engulfed by a macrophage.
- Question: Which clearance process is occurring, and why is inflammation limited?



Schematic

Application: Mechanism and consequence

- The process is efferocytosis: Phagocytes recognize eat-me signals and engulf cell bodies.
- Rapid clearance prevents leakage; Engulfing macrophages reduce proinflammatory cytokine output.
- Thus a large amount of apoptosis can remain histologically subtle.

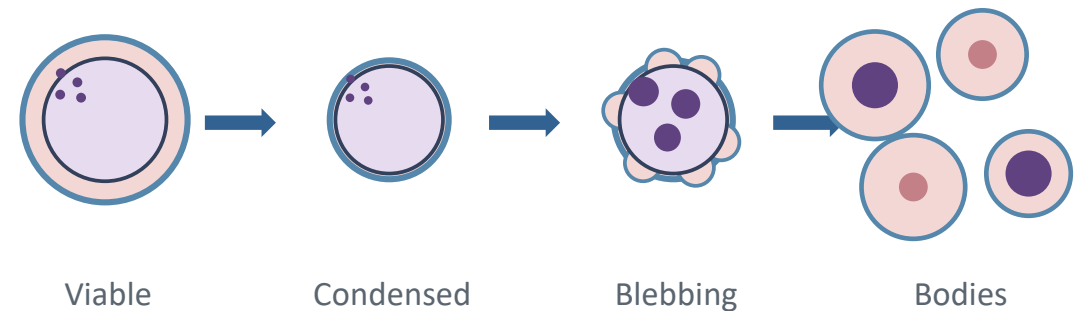


Red headgroups: Phosphatidylserine

Schematic

Integrated picture of apoptotic cell death

- Morphology: Shrinkage → chromatin condensation → blebs/bodies.
- Biochemistry: Executioner-caspase effects on DNase and structural targets.
- Resolution: Phosphatidylserine recognition → efferocytosis → limited inflammation.



Schematic sequence; Stages may overlap

Schematic

Sources and illustration status

- Robbins, Cotran & Kumar, Pathologic Basis of Disease, 11th ed. (2026), Chapter 2, pp. 41–44.
- Assigned section: “Morphologic and Biochemical Changes in Apoptosis,” p. 41 onward.
- Biochemical execution and removal details are drawn from adjacent “Mechanisms of Apoptosis,” pp. 42–44.
- All cell/tissue drawings in this deck are original schematic PowerPoint shapes, not photomicrographs.

Illustrations are labeled as schematic; genuine diagnostic micrographs were not available for embedding.