Apoptosis Signaling and Detection

Early Stage:
1. Translocation of phosphatidylycerine to the outer leaf of plasma membrane
2. Loss of mitochondrial membrane potential (ΔΨm)
3. Cytochrome c is ATP release
4. Activation of caspases 8 and 9

Mid Stage:
5. Activation of caspases (3, 6, 7)
6. Poly-ADP-Ribose Polymerase (PARP) cleavage
7. Cell shrinkage
8. Activation of nucleases

Late Stage:
9. DNA fragmentation
10. Nuclear collapse
11. Formation of apoptotic bodies
12. Phagocytosis by macrophages

MitoLight® Mitochondrial Apoptosis Detection Kit uses the lipophilic, membrane-permeant dye DiOC6(3) to detect the loss of mitochondrial membrane potential, which indicates early apoptosis. The loss of mitochondrial membrane potential, (ΔΨm), is a sign of early apoptosis. The MitoLight® Mitochondrial Apoptosis Detection Kit (Cat. No. APT142) also includes 7-AAD, a cell impermeant dye that helps distinguish live, viable cells from apoptotic and dead cells. Annexin V is conjugated to a red sensitive marker (green) and subsequently with Hoechst™ stain (blue). Cells were labeled with FLICA In Situ Caspase Detection Kits (multiple) and negative controls. Suspension cells were incubated with staurosporine (right) and then stained using FlowCellect® Annexin V Red Kit (Cat. No. FCCH100108) and subsequently with Hoechst™ stain (blue). Cells were labeled with FLICA In Situ Caspase Detection Kits (multiple) and negative controls. For detecting late-stage apoptosis, in addition, BodyFit® labels can detect mid-stage apoptosis, in which chromatin condensation has begun but major changes in the nuclear are not visible.

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