

# [The study of cell death](https://assignbuster.com/the-study-of-cell-death/)

Cell death is absolutely the most important event for organisms. Two forms of cell death, necrosis and apoptosis are discovered with distinct morphological features. Morphological characteristics in necrotic cells are frequently implicated to distinguish the two cell death forms. Necrotic cells swell initially with little alteration in chromatin. Subsequently, nuclear structure and compartments in necrotic cells become disorganized including mitochondria swelling and the shrinkage of inner mitochondrial membrane. At late stage in necrosis, with the disruption of cyto-architecture, chromatin looses while nuclease and lysosomal contents are released. Inflammatory response, a striking consequence of necrosis occurs after cytoplasma membrane rupture and cell contents release. Necrotic cells are eventually disposed of by monocytes and macrophages recruited to the death site. However, necrosis is pathological cell death caused by external death factors which is harmful to surrounding cells. Each individual cell in multicellular organisms must function harmoniously with its neighbouring cells, its specialized tissue and the whole organism. Inappropriate death or proliferation of cells leads to the loss of tissue homeostasis. The physiological in-built suicide under the genetic control, apoptosis, is pivotal to tissue homeostasis and prerequisite to organism survival.

A novel form of cell death exhibiting morphological difference from necrosis was firstly discovered in vertebrate ontogeny and named ‘ cell degeneration’ (Glucksmann, 1951). Subsequently, the definition of ‘ programmed cell death’ was prompted from descriptive work to experimental verification by Saunders & Fallon (1966). With a conclusion of its morphological features, a nomenclative phrase ‘ programmed cell death’ was referred to the novel type of cell death discovered in animal embryogenesis (Lockshin & Beaulaton, 1974). The term ‘ apoptosis’ was given by Kerr et al. in 1972 to refer the novel form of cell death in a wide range of physiological progresses after physiological stimuli in addition to development.

When cells undergo apoptosis, sequential morphological changes occur in cell structure distinct from necrosis. Initially, apoptotic cells separate from neighbouring cells and lose some specialized membrane structure, i. e. microvilli and desmosomes. After cell shrinkage, apoptotic cells start blebbing, which is reversible extruding and resorbing formed by cytosol and membrane. Subsequently, rapid and irreversible condensation of chromatin and compaction of cell organelles occur. Endoplasmic reticulum (ER) swells and connects to the cell surface whilst mitochondria appear normal. The final destination of apoptotic cells is phagocytosis carried out by viable neighbouring cells or professional phagocytes. Macro-autophagy is an alternative pathway towards cell elimination, and some scientists even divide programmed cell death into type I (apoptosis ending in phaygocytosis) and type II (autophagic cell death). In autophagic cell death, double membrane vesicles containing portion of cytoplasma and unexpected cell organelle are formed as autophagosome and then fused with lysosome for turnover. Autophagy or phagocytosis is determined by cell type, stress type and maybe some other mechanisms. Compared with necrosis, the most striking feature of apoptosis is that apoptotic cells disappear rapidly without inflammatory response. Changes on apoptotic cell surface enable them to be recognized and eliminated before cytoplasm membrane rupture thus inflammatory response is avoided (Savill, 1997). In apoptosis, DNA cleavage is an evident nuclear feature in contrast to necrosis. Genomic DNA is degraded randomly into 50kbp and 300kbp fragments. Furthermore, DNA fragments at 180bp and 200bp are detected with further cleavage in apoptotic cells. The degradation of DNA in necrosis leads to a smear called ‘ DNA laddering’ on agarose electrophoresis gel.

Apoptosis widely exists in animal developmental, physiological and immune processes throughout the life time. In development, inappropriate differentiated cells and no longer required structures are eliminated by apoptosis. The disappearance of Xenopus tadpole tail is a frequently cited example of apoptosis in embryogenesis. Detrimental cells can be also eliminated through apoptosis. An intrinsic apoptotic strategy to prevent virus proliferation in vivo is evolved in animals. The virus-induced apoptosis in infected and neighbouring cells in host can prevent infection spreading as primitive defence but viral encoded apoptosis inhibitors are produced meanwhile. Conversely, aberrant apoptosis is also disastrous to tissue homeostasis i. e. Alzheimer disease.

Several experimental approaches are implicated in the distinguishing of apoptosis from necrosis based on their morphological and biochemical features. Apoptotic cell blebbing and chromatin condensation can be identified using microscopy and electron microscopy. Terminal deoxynucleotidyl transferase dUTP nick end labelling (TUNEL) is a method to detect DNA fragmentation by labelling the exposed -OH group on the 3′ end of nucleic acids. DNA laddering, cytochrome c release and induction of caspase activity are also widely used indicates of apoptosis.

## 1. 1. 2 Programmed cell death in plants

Programmed cell death in plants refers to the apoptotic-like cell death which is under the strict genetical control. Typical apoptosis in animal cells must exhibit all of the following features: cell shrinkage, chromatin condensation, DNA fragmentation and DNA laddering, caspase activation, apoptotic bodies shedding and phagocytosis or autophagy. Except the shedding of apoptotic bodies and phagocytosis, other apoptotic features can be detected in plant cells undergoing PCD. For instance, plant protoplast retraction and cytoplasm condensation after heat shock are defined as features of PCD (Reape et al., 2008). Based on the genetic and morphological similarity in plant PCD and animal apoptosis, several animal apoptosis hallmarks are applied in plant PCD distinguishing. For example, cell death in monocot aleurone layer and endosperm, senescence of petal, carpel tissue and leaves or during anther development induced by different stimuli are defined as PCD using DNA laddering (Danon et al., 2000). TUNEL, the most widely used apoptotic detection method in animals can be implicated in plant PCD identification. The induction of caspase-like activity in plant PCD is detected. Specific feature in plant PCD which is absent in animal apoptosis such as the crescent nucleus is also mentioned.

Plant PCD occurs in developmental processes as well as response to abiotic or biotic stresses i. e. innate immune against pathogens, heat shock and starvation. Plant cells undergoing PCD in developmental stage show swelling vacuoles initially and sequential elimination of ER and other cell compartments. After the breakdown of mitochondria and nuclei, rupture of vacuolar membrane and cytoplasm membrane are observed. In some cells i. e. suspensor cells, vessels, xylem and phloem fibres, cork cells etc, cell wall is not affected in PCD and specially differentiated tissue with cell wall such as vascular bundle is therefore formed. On the contrary, cell walls in aerenchyma, endosperm and senescent mesophyll disappear completely after PCD (van Doorn & Woltering, 2005). Autophagy, which is a process of intracellular components turnover by lysosome and autophagosome provides an alternative destination of disposal cells distinct from phagocytosis in animal apoptosis. In plant PCD, phagocytosis is absent due to the inhibition of cell wall. The degradation of cell organelles in plant PCD should be owing to the autophagy exerted by vacuole in most of plant cells. However, PCD of endosperms in cereal such as barley, wheat and rice is exception from the vacuole mediated autophagy.

Hypersensitive response (HR) is an innate immune response in plants against pathogen attack. HR includes PCD in infected and neighbouring cells, local anti-pathogen chemical secretion and induction of host resistance (Mur et al., 2008). HR triggered by fungal toxin, bacteria and virus can reduce susceptibility of host plants to pathogen and evade further invading. HR mediated PCD is triggered and processed faster than that in plant development (Pozo & Lam, 1998). Initiation of HR mediated PCD relies on the interaction of “ Resistance gene” (R gene) products and pathogen avirulence gene (avr gene) products (Lorrain et al., 2003). At the same time, basal defence response also occurs to prevent infection propaganda with R-gene mediated PCD. Autophagy has not been reported in PCD triggered by HR and mechanism is still unknown.

## 1. 1. 3 Programmed cell death is ubiquitous throughout the evolution

Apoptosis is ubiquitously discovered in multicellular animals throughout the evolution since it is critical in development and tissue homeatosis. Apoptotic machinery is also remarkably conserved from invertebrate to vertebrate with homologous apoptotic factors and pathway. The most clearly comprehended animal apoptotic pathway is that in nematode Caenorhabditis elegans, in which the predetermined differentiation fate of every individual cell was precisely narrated (Hengartner & Horvitz, 1994). In the 1090 cell births and 131 cell deaths in its ontogenesis from zygotes to adult, several genes are involved in C. elegans apoptotic regulation. Homologues of the thirteen genes with highly functional and structural similarity are identified in vertebrate hence the relative simply apoptotic pathway in nematode is conserved in complicated mammalian apoptosis. For instance, anti-apoptotic gene bcl-2 is mammalian homologue of ced-9 which can suppress apoptosis in C. elegans.

Plant PCD identified in xylem development, pollen self-incompatibility, senescence or hypersensitive reaction exhibits homologous morphological alteration. However, lacking major animal apoptotic pathway such as caspase cascade, the genetic apoptotic pathway in plant PCD is still unknown. Extrapolation and identification of animal apoptotic analogues in plants is coming to the fore and will eventually improve the elucidation of mechanism in plant PCD. Interestingly, monocellular organism yeast can perform apoptotic-like death after chemical stimuli i. e. H2O2, acetic acid or sugar. Such apoptotic-like cell death is also detected in yeast aging and reproducing. A cluster of apoptotic gene orthologues are identified (Madeo et al., 2004; Wissing et al., 2004; He et al., 2007). Apoptosis even occurs in primitive monocellular protists Dictyostelium discoideum. The formation of stalk in nutrient starvation includes a cell death process showing high similarity with animal apoptosis in morphological aspects. It is therefore suggested PCD evolves from primitive single cell organisms and might be a result of conflict between archaebacteria and protomitochondria (Blackstone & Kirkwood, 2003). Phagocytosis is absent in the final stage of PCD in Dictyostelium, yeast and plant while autophagy is demonstrated to contribute in dead cell disposal (Levine & Klionsky, 2004; Hofius et al., 2009).