Dying Cells in Living Tissues
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Abstract
“For every cell, there is time to live and a time to die”. As important as cell division and cell migration, regulated or programmed cell death allows the organism to tightly control cell numbers and tissue size, and to protect itself from rogue cells that threaten homeostasis. When the interest of the cell demands the elimination of unwanted cells, then the cells commit suicide by the process called “Apoptosis”. However, interest in the subject was modest, and the concept of physiological cell death did not receive widespread recognition until the publication of a seminal paper on apoptosis by Kerr and colleagues some 30 years ago. Apoptosis can be initiated through various signals via the extrinsic pathway which involves death receptors, or via the intrinsic pathway which is initiated by intracellular damage and involves the mitochondria and release of cytochrome c from it to further activate caspases. The pathogenesis of several diseases such as cancer, neurodegenerative disorders, autoimmune disorders, heart disease, and infectious diseases including AIDS is closely related to aberrant apoptosis. There are number of publications on apoptosis in different diseases, this article is an attempt to synoptically describe the apoptosis in health and different diseases.

Keywords: Apoptosis; Organogenesis; Synaptogenesis; Neurodegenerative; Premalignancy; Cytotoxic; Therapeutic.

Introduction
The human body is composed of nearly \(10^{14}\) cells.\(^1\) Every day billions of cells are produced by mitosis and a similar number die by different types of cell death to maintain tissue homeostasis. Necrosis and apoptosis are the two modes of cell death processes in living organisms.\(^2\) Necrosis is pathological cell death and occurs during the extreme cases of cell injury and repair.\(^3\) Apoptosis is a normal physiological cell death process which occurs in all living organisms in their embryonic or adult development thus eliminating unwanted, functionally abnormal or harmful cells.\(^4\) Physiologically, all multicellular organisms use apoptosis during development, homeostasis, defence, metamorphosis, terminal differentiation, immune response, cellular response to growth factors and hormones. As important as cell division and cell migration, apoptosis facilitates the organism to tightly control cell numbers and tissue size, and to protect itself from rogue cells that threaten homeostasis.\(^5\) There are a wide variety of stimuli, both physiological and pathological, that can trigger apoptosis, but not all cells will necessarily die in response to the same stimulus.\(^6,7\)

A violation of cellular homeostasis may play a role in various human diseases. Apoptosis dysregulation contributes to half of all human diseases.\(^8\) Thus, too much or little cell death can have catastrophic consequences. As mentioned in the article of Peter ME, et al, Carl Vogt in 1842 discovered apoptosis then in 1972, Kerr, Wyllie, and Currie coined the term apoptosis and evoked an unparalleled interest in this field of science.\(^9\) During development, the role of apoptosis is to sculpt structures, remove excess cells and delete tissues that have outlived their usefulness. Apoptosis also contribute in cellular proofreading and eliminating the cells that are inappropriate locations during development and infected or damaged cells during the lifetime.\(^10\) Not surprisingly, deregulation of cell death has severe consequences for the developing organism and adult. Inappropriate cell death is associated with degenerative neurological diseases, stroke, cardiac ischemia and immune suppression associated with AIDS, whereas suppression of naturally occurring cell death contributes to autoimmune disease and cancer.\(^11,12\)

The study of programmed cell death, or apoptosis, have become a major focus of
research interest in many areas of medicine in the last decade, due to recognition of the fact that apoptosis plays a major role in both physiological and pathological conditions. With the intention of researching the role of apoptosis in healthy and pathological conditions, the literature within this article is reviewed.

Apoptosis in Healthy Tissues

Apoptosis plays an important role in morphogenetic processes in early developmental stages of intrauterine life. It is associated with normal differentiation and formation of organs during organogenesis. Apoptosis is seen first, in the organs which undergo complicated development in early fetal period. In the process of reproduction, after the fertilization of the two gametes, cell proliferation takes place by mitosis to increase the numbers of the cells in the developing organism.  

Apoptosis in different adult cells

1. Germ cells
2. Haematopoietic system
3. Neurons
4. Muscle cells
   a. Cardiomyocytes
   b. Smooth muscle
   c. Skeletal muscle
5. Fibroblasts
6. Tooth

1. **Germ Cells:** Apoptosis play an important role in the three discrete processes related to ovarian development and cyclicity,
   (1) Perinatal oogonium and oocyte attrition,
   (2) Follicular atresia and
   (3) Luteolysis

The largest peak of germ cell occurs within the dividing oogonial population during later stages of fetal development, although cellular degeneration continues into the first few days after birth and is observed in both mitotic and post mitotic germ cells. There are three discrete waves of germ cell loss; firstly, attrition of dividing oogonia, secondly, degeneration of pachytene-stage oocytes and thirdly, loss of diplotene stage oocytes. All these results in less than one third of the total number of potential germ cells being endowed in the ovary within primordial follicles shortly after birth.

2. **Hematopoietic System:** Both hematopoietic cell production and elimination are regulated by apoptosis. The maintenance of erythropoietic stem cells (BFU-Es and CFU-Es) is dependent upon the presence of erythropoietin. Withdrawal of Erythropoietin (EPO) results in apoptosis of these red cells.  

It has been exposed that thyroid hormones induces apoptotic cell death of differentiating erythropoietic progenitor cells. These myeloid hematopoietic cells require viability factors, which suppress apoptosis, which are required throughout the differentiation process from immature cells to mature granulocytes, eosinophils and macrophages. However IL-6 and IL-1 can induce viability in normal immature myeloid cells without inducing growth.

Apoptosis is a fundamental part of T-lymphocytes maturation and selection while T-lymphocytes are maturing in thymus, any T-cells that bind to self antigens or make nonfunctional receptors undergo apoptosis. In several studies aging was found to increase CD8 T-cell apoptosis by hyperstimulation through the T-cell receptor, thus leading to decreased levels of CD8 T-Cells.

Macrophages also undergo apoptosis, although little is known about the mechanism of the process. Tidball and St. Pierre reported that macrophages predominate in the inflammatory response and rapidly disappear through apoptosis.

3. **Neurons:** Neuronal cell death by apoptosis occurs during brain development. Like all cells, neuronal survival requires trophic support. It has been estimated that about 50% of the neurons present at birth die after synapses are formed in vertebrates. This includes cells with the most receptors and cells with the most efficient signal transduction system. It turns to be more effective to eliminate the less competitive cells than to prevent the proliferation of new cells.

4. **Muscle Fibers:** It was first observed in insects that developing muscles undergo apoptosis. A related type of fragmentation occurs in embryonic skeletal muscles of birds and mammals,
including humans. Apoptosis in embryonic muscles is a well-known phenomenon. Apoptosis in muscle appears at different stages of development in human embryos and enhanced apoptosis was observed in early postnatal life in many pathological conditions.

5. **Fibroblasts**: In vivo there is good evidence that primary fibroblasts can undergo apoptosis, in wound healing of skin and is thought to occur in other organs also. In vitro studies suggest that C-myc is involved in the fibroblast apoptosis in a process that involves the interactions of Fas and Fas-L on the cell membrane. Apoptosis signals are also involved in the regulation of collagen degradation by inducing collagenase activity. It is seen that p53 binds to the promoter of the human type IV collagenase (MMP-2) gene.

6. **Apoptosis in the Tooth Development**: Tooth development is a complex process that involves epithelial mesenchymal interactions, resulting in the various stages which exhibit different morphologic patterns. Several studies have proved that apoptosis in epithelial and mesenchymal cells during embryonic tooth development exists in rodent animals models. During tooth development, it was reported that complementary cellular events, such as cell division, migration and cell death, occurred continuously and the difference in cell division ability played an important role in tooth development.

In the initiation stage of tooth development, no apoptotic cells are observed in the epithelial thickening or in the surrounding mesenchyme. In the early bud stage, apoptotic cells are found in the budding epithelium, particularly cells facing the oral cavity. During further development, at the late bud stage, when the tooth germ prolongs its central axis, apoptotic cells become accumulated at the tip of the tooth bud. At the bud stage, no apoptotic cells are observed in the mesenchyme surrounding the budding epithelium. When the tooth germ reaches the cap stage, this cluster of apoptotic cells is still apparent and becomes localized within the epithelial enamel knot. During further development the number of apoptotic cells increases but, interestingly, without any evident loss of the cell mass, suggesting a rapid replacement by high-proliferating cells surrounding the enamel knot. With disappearance of the primary enamel knot, apoptosis is no longer observed in this area but is detected in the gubernaculum. At the cap stage, a few apoptotic cells are detectable in the condensed dental mesenchyme. Tooth crown morphogenesis and cytodifferentiation occurs during the bell stage. At this time, apoptosis is evident in secondary enamel knots and in stratum intermedium cells adjacent to the enamel knots and some scattered apoptotic cells are also found in the mesenchyme. All teeth, regardless of shape or identity, pass through the same developmental stages and consist of the same tissues.

**Apoptosis and Human Disease**: Derailedment of apoptosis in the regulation of immune system leads to several diseases with either too much or too little apoptosis. Lack of apoptosis plays role in generation of tumors such as follicular lymphomas. Immune homeostasis and maintenance of immune tolerance are dependent on apoptosis induction and rapid clearance of apoptotic cells in peripheral and central lymphoid organs. Autoimmune disease arise from either effective elimination of auto-reactive T or B cells, resulting in tissue destruction, or from defective clearance of apoptotic cells displaying auto-antigen on their cell surface as in the case of systemic lupus erythematosus. The disease can be divided into two groups; one associated with the inhibition and other with excess of apoptosis.

**A. Diseases caused due to hindering of apoptosis**:

1. **Autoimmune diseases**
   - Systemic lupus erythematosus (SLE)
   - Scleroderma
   - Rheumatoid arthritis
   - Crohns disease
   - Sjogrens syndrome
   - Recurrent apthous stomatitis
   - Lichen planus
   - Erythema multiforme

2. **Viral infection**
   - Human Herpes Virus (HHV)
   - Human Papilloma Virus (HPV)

3. **Premalignancy**
Oral submucous fibrosis.
Oral leukoplakia.

4. Malignancies
B-cell lymphoma
Squamous cell carcinoma
Liposarcoma
Osteosarcoma
Leiomyosarcoma
Mucoepidermoid carcinoma
Adenoid cystic carcinoma.

B. Diseases occurring due to excess of apoptosis:
1. AIDS
2. Haematological diseases
Leukaemias
Myelodysplastic syndrome
Aplastic anaemia
Cyclic neutropenia
3. Neurogenerative diseases
Alzheimer’s disease
Parkinson’s disease
Epilepsy
4. Tissue damage
Myocardial infarction.

Autoimmune Diseases: During development after migration to the thymus majority (90 ± 95%) fail to produce T-cell receptor (TCR) and die via the apoptotic pathway.37 Thymocyte apoptosis can be induced by endogenously produced glucocorticoids and a lack of TCR.38 Dysfunction of the apoptotic pathway causes autoimmune disease, immunodeficiencies and lymphoid malignancies.

Viral Infection: Many viruses (RNA and DNA) inhibit apoptosis in their target cells to increase host cell life and permit viral replication. Viruses may encode anti-apoptotic proteins, like the baculovirus IAPs, the baculovirus p35 and cowpox serpin protein crmA, to induce apoptosis in infected host cells by production of an array of viral products and promote the development of certain cancers.39

Oral Premalignancy: Studies have shown that Bcl-2, anti-apoptotic marker is dysregulated during progression from oral epithelial dysplasia to squamous cell carcinoma. Expression of this oncoprotein was directly related to the degree of dysplasia, suggesting its role of this protein in the early stages of tumor progression.40

Neurodegenerative Diseases: In contrast to the rapid turnover of cells in proliferative tissues, neurons commonly survive for the entire lifetime of the organism; this enduring nature of neurons is necessary for maintaining the function of those cells within neuronal circuits. During development of the central and peripheral nervous systems, many neurons undergo apoptosis during a time window that coincides with the process of synaptogenesis.41 Initial overproduction of neurons, followed by death of some, is probably an adaptive process that provides enough neurons to form nerve cell circuits that are precisely matched to their functional specifications.42 Accordingly, the decision as to which neurons die is made by cellular signal transduction pathways that are ‘tuned’ to the functionality of neuronal circuits. Many people experience excessive death of one or more populations of neurons as the result of disease or injury. The number of people with such neurodegenerative disorders is rapidly increasing as the average lifespan is getting longer.43

Cancer: The notion that apoptosis might influence the malignant phenotypes goes back to the early 1970s. There is evidence that a failure to initiate apoptosis following DNA damage may cause cancer. Cancer cells develop the ability to escape the activation of the apoptotic genes.44 In multicellular organisms, cells have many apoptotic mechanisms for the removal of damaged cells. It is therefore essential for cells to overcome cell death induced by these stimuli for cancer to develop.45 Tumor cells may evade apoptosis by inactivation of apoptosis-inducing genes or by enhancement of the activity of anti-apoptosis genes.46 Levels of Bcl-2 are elevated in various other human cancers.47

Conclusion
The term apoptosis was first used in 1972 to describe a form of cell death with markedly different morphology to necrosis and was first postulated 130 years earlier in 1842 by Carl Vogt.44 Apoptosis continued to drift into and out of fashion throughout the first half of the 20th century until an influential review on cell death was published in 1951, implicating that cell death plays an important role in embryogenesis and development of vertebrates.45 The number of researches related to apoptosis is increasing exponentially every year, since it has now become clear that many diseases are characterized by dysregulation of apoptotic programs. Although apoptosis has been recognized for a considerable length of time, its importance is just beginning to be truly
appreciated, especially with respect to neoplastic growth. Not surprisingly this process has inherent weaknesses that, when compromised, can result in inappropriate cell death (either too little or too much) and disease pathogenesis. The potential for therapeutic intervention in any of these scenarios is theoretically possible as apoptosis plays an important role in several areas of interest. Although we still have much to learn, as we already know that this system is extremely complex and there are several areas where the modulation of apoptotic death could be of benefit in the treatment of many diseases in the future.

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