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Apoptosis - A Review Article

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ABSTRACT

The survival of living beings depends on the correct and co-*ordinate* functioning of various cell types. For each cell line, control of the number of cells is the result of a dynamic balance between cell proliferation and cell death. Apoptosis is a programmed cell death prosing that cell death during development is not of accidental nature but follows a sequence of controlled steps lending to locally and temporally define self destruction. Cells can die by necrosis but apoptosis is a form of death that the cell itself initiates regulates and executes using molecular machinery. The cell brings about its own death and lyses signaled from outside or programmed in its genes by systematically degrading its own macromolecules. Gain- and loss-of function models of genes in the core apoptotic pathway suggest that perturbation of cellular homeostasis can be a primary pathological event that results in disease. There is now compelling evidence that insufficient apoptosis can result in cancer or autoimmunity whereas accelerated cell death is evident in degenerative diseases, immunodeficiency and infertility.

Keywords: Cell, Life, Death, Decision

INTRODUCTION

For every cell there is a time to live and a time to die. While philosophers have spent centuries searching for the meaning of life, cell biologists are fascinated by the meaning of death. Kerr and colleagues observed that death is an important and predestined fate of individual cells of organisms [1]. Apoptosis is of Greek origin, meaning "falling off or dropping off", in analogy to leaves falling off trees or petals dropping off flowers. The analogy emphasizes that the death of living matter is an integral and necessary part of life cycle of organisms. The apoptotic mode of cell death is an active and defined process which plays an important role in the development of multicellular organisms and in the

regulation and maintenance of the cell populations in tissues upon physiological and pathological conditions [2].

During the initial stages of development, the viability of the organism depends on the selection and differentiation of the adequate cells in the various tissues. At later stages, the maintenance of the organism requires a specific cellular adaptability. For instance, blood cells are instantly renewed from hematologic precursors while on the other hand, neural cells evidence a limited capability for renewal and many neurons survive and persist throughout the life of the individual. Cell death can occur through two different processes termed necrosis and apoptosis. For these two modalities of cell death, there are differentiating functional and morphologic characteristics [3].

Diseases associated with apoptosis include stroke, hepatitis, Parkinson's disease and Alzheimer's disease. Many existing treatment (such as non-steroidal anti-inflammatory and anticancer treatments) act through apoptosis. New treatments aimed at modifying apoptosis are being developed and are likely to be used to manage common diseases in the next decade [4].

PHYSIOLOGICAL ROLE OF APOPTOSIS

Apoptosis is a well-organized process of cell suicide, which is mandatory to normal development and homeostasis of the adult organism. Apoptosis counterbalances the increase of cell number by proliferation and removes unwanted and surplus cells. The first role of apoptosis is during intrauterine development. During development many cells are produced in excess which eventually undergo programmed cell death and thereby contribute to sculpturing many organs and tissues carving out the interdigital webs of the fingers and toes [5]. Cells of an adult organism constantly undergo physiological cell death which must be balanced with proliferation in order to maintain homeostasis in terms of constant cell numbers. The majority of developing lymphocytes die

either during genetic rearrangement events in the formation of the antigen receptor, during negative selection or in the periphery, thereby tightly controlling the pool of functional but not self-reactive immune cells and at the same time keeping lymphocyte numbers relatively constant [6].

Normal homeostasis is not just a passive process but regulated through apoptosis. The same mechanisms serve to "mop up" damaged cells. Apoptotic processes are of widespread biological significance involved in development, differentiation, proliferation/homeostasis, regulation and function of the immune system and in the removal of defect and harmful cells. Thus, dysfunction or deregulation of the apoptotic programmme is implicated in a variety of pathological conditions. Defects in apoptosis can result in cancer, autoimmune disease and spreading of viral infections while neurodegenerative disorders, AIDS and ischemic diseases are caused or enhanced by excessive apoptosis [7].

Programmed cell death occurs in all metazoans such as mammals, nematodes and insects [8]. Programmed cell death also plays a role in plant biology and apoptosis- like cell death mechanism has been observed in yeast [9, 10]. Programmed cell death is also an integral part of the life cycle of other unicellular eukaryotes such as the kinetoplastid parasite *Trypanosoma brucei brucei,* the ciliate *Tetrahymena thermophilia* and the slime mould *Dictyostellium discoideun*. Prokaryotes such as *Bacillus subtilis, streptomyces* and mycobacteria sometimes undergo regulated cell death [11].

CAUSES OF APOPTOSIS

Initiators of apoptosis include anticancer drugs, gamma and ultraviolet irradiation and deprivation of survival factors such as interleukin-1 and various other cytokines that activate "death receptors" such as fas and tumour necrosis factor receptors. Cells can be killed by injurious agents or induced to commit suicide [12, 13]. Injury to cells includes mechanical

damages and exposure to toxic chemicals. The affected cells and their organelles like mitochondria swell because the ability of the plasma membrane to control the passage of ions and water is disrupted, the cell contents leak out and this leads to inflammation of the surrounding tissues.

Cells commit suicide for two major reasons; first, apoptosis is one means by which a developing organism shapes its tissue and organs. For instance, a human foetus has webbed hands and feet early on its development, later apoptosis removes skin cells, revealing individual fingers and toes. A foetus's eyelids form an opening by the process of apoptosis. During metamorphosis, tadpoles lose their tails through apoptosis. In young children, apoptosis is involved in the process that literally shape the connections between brain cells and in mature females, apoptosis of cells in the uterus causes the uterine lining to slough off at each menstrual cycle.

Cells may also commit suicide in times of distress for the good of the organism. An example is in the case of a viral infection; certain cells of the immune system called cytotoxic T lymphocytes bind to infected cells and trigger them to undergo apoptosis. Also cells that have suffered damages to their DNA which can make them prone to becoming cancerous are induced to commit suicide [14]. Cells that are induced to commit suicide shrink, develop bubble-like bleds or budding on their surface, have the chromatin (DNA and protein) in their nucleus degraded, have their mitochondria breakdown with the release of cytochrome c, the cells break into small, membrane wrapped fragments and the phospholipid, phosphotidylserine which is normally hidden within the plasma membrane is exposed on the surface. This "eat me" signal is bound by receptors on phagocytic cells like macrophages and dendritic cells which then engulf the cell fragments. The phagocytic cells secrete cytokines that inhibit inflammation like interleukin- 10 [15].

A cell can decide to commit suicide as a result of the balance between the withdrawal of positive signals, that is, signals needed for continued survival and the receipt of negative signals. The continued survival of most cells requires that they receive continuous stimulation from other cells and for many, a continued adhesion on which they are growing. Examples of positive signals are growth factors for neurons and interleukin-2 (IL-2), an essential factor for the mitosis of lymphocytes. Receipt of negative signals is due to increased level of oxidants within the cell, damages to DNA by these oxidants or other agents like *x*-rays, ultraviolet light and chemotherapeutic drugs. Negative signals are also as a result of accumulation of proteins that failed to fold properly into their proper tertiary structure, molecules that bind to specific receptors on the cell surface and signal the cell to begin the apoptosis programme. These death activators include tumour necrosis factor-alpha (TNF –a) that binds to the TNF receptor, lymphotoxin (known as TNF-B) that also binds to the TNF receptor and Fas ligand (Fasl),a molecule that binds to a cell-surface receptor named fas or cell death 95 (CD 95). [16]

MOLECULAR MECHANISM OF APOPTOSIS

Apoptosis describes the orchestrated collapse of a cell characterized by membrane blebbing, cell shrinkage, condensation of chromatin and fragmentation of DNA followed by rapid engulfment of the 'corpse' by neighbouring cells. It is distinguished from death by necrosis by the absence of an associated inflammatory response [17]. The stages of apoptosis may be considered as initiation, genetic regulation and effector mechanism.

Apoptosis can be triggered by various stimuli from outside or inside the cell. Examples include the ligation of cell surface receptors by DNA damage as a cause of defects in DNA repair mechanisms, treatment with cytotoxic drugs or irradiation, by a lack of survival signals, contradictory cell cycle signaling or by developmental death signals. The first protein to have been described as an inducer of apoptosis was Fas

[18]. This is a 48-KD surface protein. The binding of fas to its ligand (FasI) induces apoptosis in a wide variety of cells including activated lymphocytes.

Caspases are central initiators and executioners of apoptosis. In the cell, caspases are synthesized as inactive zymogenes called procaspases which at their N-terminus carry a prodomain followed by a large and small subunit separated by a linker peptide. Upon maturation, the procaspases are proteolytically processed between the large and small subunit. The prodomain is frequently but not necessarily removed during the activation process. A heterotetramer consisting of each two small and two large subunits then forms an active caspase.

The term "caspase" is derived from cysteine-dependent aspartate specific proteases. Their catalytic activity depends on a critical cysteine-residue within a highly conserved active-site pentapeptide and these caspases specifically cleave their substrates after Asp residues. Seven different caspases have been identified in Drosophilia and fourteen in mammals with caspase - 11 and caspase -12 only identified in mouse [19, 8]. According to unified nomenclature, caspase-1 is interleukin – 1β-converting enzyme (ICE), the first mammalian caspase described to be a homologue of ced-3 (cell death 3). [20, 21]. Caspases – 1, -4, -5, -11 and -12 are mainly involved in the proteolytic maturation of pro-inflammatory cytokines such as pro-IL-1β and Pro-IL-18. Mice deficient for caspase-1 or caspase-11 develop normally and cells from these knockout mice remain sensitive to various death stimuli [22]. In contrast, gene knockout experiments targeting caspase-3 and caspase-9 resulted in perinatal mortality due to severe defects in brain development [23] whereas caspase-8 deficient embryos died after day 12 [24]. This underlines the importance of caspases as proapoptotic mediators. Caspases 3, -9, -8 and additionally caspses -2, -6, -7 and -10 have been recognized to play an important role in the apoptotic signally machinery [25].

There are two major apoptotic pathways. One activated via death receptor activation (extrinsic) and the other by stress-inducing stimuli (intrinsic). Triggering of cell surface death receptors of the tumour necrosis factor (TNF) receptor super family including CD 95 and TNF-related apoptosis-inducing ligand (TRAIL) –R1/R2, results in rapid activation of the initiator caspase-8 after its recruitment to a trimerized receptor-ligand complex through the adaptor molecule fas-associated death domain protein (FADD). [26].

In the intrinsic pathway, stress induced apoptosis results in perturbation of mitochondria and consequent release of proteins such as cytochrome c, from the inter-mitochondrial membrane space. The release of cytochrome c is regulated in part by BCL-2 family members with anti-apoptotic (Bcl-2/Bcl-x_L/Mcl 1) and pro-apoptotic (Bax, Bak and tBid) members inhibiting or promoting the release respectively. Once released, cytochrome c binds to apoptotic protease-activating factor 1 (APaf 1) which results in the formation of APaf 1-CASPASE -9 apoptosome complex and activation of the initiator caspase -9. The activated initiator caspses-8 and -9 then activate the effector caspases-3, -6 and -7 which are responsible for the cleavages of important cellular substrates resulting in the classical biochemical and morphological changes associated with the apoptotic phenotype [27, 13]. In addition to their role in cellular energy metabolism, mitochondria are now recognized as central players in cell death through the Bax/Bak channel and the non-specific pore in the inner mitochondrial permeability transition pore (MPTP). The opening of these pores uncouples mitochondria which prevents them from providing energy for the cell and leads to necrotic cell death. MPTP opening is important in the injury to the heart and brain that follows an ischaemic episode such as heart attack or stroke.

Agents that inhibit pore opening can protect hearts and brains from ischaemia/reperfusion injury [28]. Other pro-apoptotic proteins released from the inter membrane space in response to an apoptotic stimulus

through the opening of MPTP include apoptosis inducing factor (AIF), Smac/DIABLO and omi/HtrA2. Apaf l, caspase-9, caspase -3 and the x-linked inhibitor of apoptosis (XIAP) are the main constituents of the native apoptosome.

REGULATORY MECHANISM OF APOPTOSIS

Apoptosis or programmed cell death is the method by which tissue remodeling takes place during normal growth and development and the physiologic mechanism by which labile cell populations such as gastro intestinal epithelial cells, lymphocytes, dendritic cells and neutrophils are regulated. Apoptosis is of particular importance for the immune system as the means by which self-recognizing lymphocytes are deleted and expanded lymphocyte populations are reduced at the conclusion of an acute immune response. This closely related energy-requiring process can be initiated through two different mechanisms, each based on the successive activation of pre existing but dominant cysteine-aspartate proteases or caspases.

The intrinsic apoptotic pathway begins within the cell when toxic alterations bring about a decrease in the mitochondrial transmembrane potential leading to the opening of cytochrome c and other substances into the cytoplasm. The extrinsic pathway by contrast, is triggered by extracellular events through the binding to the cell surface receptors of the tumour necrosis factor superfamily death ligand including TNF – and fas ligand. Although the intrinsic pathway involves early activation of caspase -9 and the extrinsic pathway is mediated through caspase-8, both lead to activation of the "executioner" caspase -3 and a variety of proteases with endonucleases. Once begun, apoptosis may be described as an orderly disassembly of the cell from within. Chromosomal DNA is cleaved into the olingonucleosomaL segments, the nucleus is divided into discrete subunits and the cell itself is partitioned into multiple membrane-bound fragments whose outer surfaces are marked by large number of phosphotidylserine molecules, leading to rapid uptake by

phagocytes. Because all multicellular organisms use programmed cell death to maintain and modify their tissues, this process does not evoke an inflammatory response and its end products actually serve as anti-inflammatory stimuli [29].

It is interesting to note that all cells of a multicellular animal might be intrinsically programmed to self-destruct and would die instantaneously unless cell death is continuously repressed by survival signals such as provided by other cells of the organism, for example growth factors, hormones and nutrients. These survival signals enhance the expression and/or activity of antiapoptotic regulatory molecules thereby keeping in check the activation of proapoptotic factors (30).

Actually, a set of various antiapoptotic molecules and mechanisms has been identified as well as proapoptotic factors that counteract those inhibiting molecules when apoptotic demise of a cell is timely and imperative.

Tumour suppressor genes have been implicated in the regulation of apoptosis. In response to various cellular stresses, p53 expression is increased and the protein is post-translationally modified resulting in either arrest of the cell at gene 1 (G1) or commitment to death through apoptosis. Co-expression of two members of the apoptosis-stimulating protein (ASPP) for p53 family, ASPP1 and APP2, specifically stimulate the transactivation function of P53 on promoters of pro-apoptotic genes such as Bax and p53-inducible gene 3 (PIG 3) but not on the promoters of the cell-cycle arrest- associated gene p21 [30]. Some tumours retain the wild type P53 especially in human breast cancers through the loss of ASPP activity. An inhibitory form of ASPP (iASPP) allows cells to bypass the tumour suppressor functions of P53 and ASPP [31]. Increased expression of iASPP confers resistance to ultraviolet radiation and to cisplatin-induced apoptosis. Whereas ASPP expression is frequently downregulated in human tumours, iASPP expression is upregulated in

human breast carcinomas that express wild-type p53 and normal levels of ASPP.

AGEING AND APOPTOSIS

Ageing is not considered a disease but it is associated with increased cell fragility that causes lymphocytes to undergo apoptosis when activated (32). This cell fragility is the underlying basis of the immune deficiency associated with ageing. In organic ageing, increased cell fragility has been reported and linked to a number of diseases characterized by chronic stimulation of cell lines. The term "replicative exhaustion" has been coined to define this cell process because a cell clone may become activated and proliferate a couple of times. If the stimulus persists, the cells are no longer able to respond and undergo apoptosis. Replicative exhaustion of the responding cell clones has been demonstrated in some diseases including AIDS in which the responding clone is the $CD4^+$ one. In these diseases, the responding clone after an initial phase of intense response to the stimulus becomes exhausted and undergoes apoptosis.

Apoptosis and Organ Transplants

Certain parts of the body such as the interior chamber of the eye and the testis are immunologically privilege sites since antigens within these sites do not elicit immune response. Also cells in these sites differ from other cells in the body as they frequently express high levels of FasL and therefore antigen-reactive T cells that express fas will be killed if they enter these sites. This is a reverse mechanism which is exploited to prevent graft rejection. The graft is protected from attack by the T cells of the host's cell-mediated immune system if some of the cells on a transplanted kidney, liver, heart are engineered to express high levels of FasL. In this way, the present need for treatment with immunosuppressive drugs for the rest of the recipient's life would be reduced or eliminated.

APOTOSIS AND DISEASE

Diseases in which apoptosis has been implicated fall into two groups, those in which there is an increase in cell survival (or diseases associated to inhibition of apoptosis) and those in which there is an increase in cell death and hence hyperactive apoptosis (33,34).

Increased apoptosis is characteristic of AIDS, neurogenerative diseases such as Alzheimer's disease, Parkinson's disease, amyotrophic lateral sclerosis, ischaemic injury after myocardial infarction, stroke and reperfusion and also in autoimmune diseases such as hepatitis and graft versus host disease. Decreased or inhibited apoptosis is a feature of many malignancies, autoimmune diseases such as systemic lupus erythematosus and some viral infections [35].

APOPTOSIS AND ALLERGY

Eosinophils play a significant role in the pathogenesis of allergy and asthma. Apoptotic death of eosinophils actively collaborates in the resolution of the inflammation which is characteristic of bronchial asthma [36]. Eosinophil apoptosis is also involved in the resolution of allergic rhinitis and allergic dermatitis [37].

APOPTOSIS AND CANCER

When functioning properly the body can induce programmed cell death or apoptosis to rid itself of cancer cells but some cancer cells find ways to escape apoptosis. Mutations lead to a loss of programmed cell death [38]. Some β-cell leukemias and lymphomas express high levels of proto-oncogene called Bcl- 2, thus blocking apoptotic signals they receive. The high levels result from a translocation of the BCL-2 gene into an enhancer region for antibody production. Melanoma (the most dangerous type of skin cancer) cells avoid apoptosis by inhibiting the expression of the gene encoding Apaf 1. Cancer cells of the lung and colon secrete elevated levels of a soluble decoy molecule that binds to FasL, plugging it up to prevent binding to Fas. This is why cytotoxic T

cells cannot kill the cancer cells (by the usual mechanism) but other cancer cells express high levels of FasL and can kill any cytotoxic T cells that try to kill them because cytotoxic T cells also express Fas (but are protected by their own FasL) [39].

Some viruses associated with cancers use tricks to prevent apoptosis of cells they have transformed. Some human papilloma viruses are involved in cervical cancer and one of them produces a protein (E6) that binds and inactivates the apoptosis promoter p53. Epstein –Barr virus (EBV) is the cause of mononucleosis and associated with some lymphomas. It produces a protein similar to Bcl -2, and another protein that causes the cell to increase its own production of Bcl-2. These actions position the cell to be more resistant to apoptosis thus enabling a cancer cell to continue to proliferate [40].

APOPTOSIS IN THE IMMUNE SYSTEM

The immune response to a foreign invader involves the proliferation of lymphocytes, the thymus (T) and/or the Bursa (B) cells. When their job is done they are removed by apoptosis leaving only a small population of memory cells [41]. Humans sometimes encounter genetic defect in apoptosis. The most common is the mutation of the gene for Fas. Mutations in the gene for fasl and caspases occur occasionally. In all these, the genetic problem produces autoimmune lymphoproliferative syndrome (ALPS) with characteristic features of accumulation of lymphocytes in the lymph nodes and spleen with enlargement, appearance of clones that are auto reactive, that is, attack self components giving rise to autoimmune diseases such as haemolytic anaemia and thrombocytopena. There is also the appearance of lymphoma, a cancerous clone of lymphocytes.

In some patients with ALP, the mutation is present in the germ line while in others, the mutation is somatic (occurred in a precursor in the bone marrow). These later patients are genetic mosaics, with some

lymphocytes that undergo apoptosis normally and others that do not [42].

APOPTOSIS AND ACQUIRED IMMUNE DEFICIENCY SYNDROME (AIDS)

The hallmark of AIDS is the decline in the number of $CD4^+$ T cells (normally about 1000 per micro liter (uL) of blood). $CD4^+$ T cells are directly or indirectly responsible (as helper cells) for all immune responses. When the number decreases below 200 per microliter, the patient is no longer able to mount effective immune responses and begins to suffer a series of dangerous infections. The dying-off of the infected $CD4^+$ cells is not mainly caused by the human immunodeficiency virus (HIV) as many uninfected CD4+ cells are also killed.

All T cells, both infected and uninfected express fas and the expression of a HIV gene called Nef in a HIV- infected cell causes the cell to express high levels of FasI at its surface while preventing an interaction with its own fas from causing it to self-destruct. However, when the infected T cell encounters an uninfected cell in the lymph node or elsewhere, the interaction of fasl with fas on the uninfected cell kills it by apoptosis, [43].

STUDY OF APOPTOSIS

Morphologically the apoptotic cell shrinks, shows deformation and looses contact to its neighbouring cells. It's chromatin condenses and marginates at the nuclear membrane, the plasma membrane is blebbing or budding and finally the cell is fragmented into compact membrane-enclosed structures called "apoptotic bodies" which contain cytosol, the condensed chromatin and organelles [44] . The cell change most consistently associated to apoptosis is the breakage of DNA into fragments containing 180-200 base pairs or multiples of the size which appear as a result of the digestion of the DNA by endonucleases. When

analyzed by electrophoresis these broken-up DNA fragments appear as a succession of discrete bands resulting in the classical "DNA laddering" pattern. A number of techniques take advantage of this DNA fragmentation for labeling the fragments and for quantifying the proportion of apoptotic cells. Each DNA fragments has a 3'oH terminal portion which can be labeled in various ways. One approach is with the help of a modified terminal dexoynucleotidyl transferase to make the labeling rate proportional to the degree of DNA fragmentation [45].

Light and electron microscopy are also classical techniques for the study of apoptosis. At present, the most widely standardized and accepted technique is based on the changes in the membrane phospholipids that occur early in apoptotic cells [46]. The negatively charged membrane phospholipids exposed to the external environment by the apoptotic cell are labeled with fluorochrome-congugated molecules and the percentage of fluorescent cells can be easily quantified.

CONCLUSION

This brief review has shown that many diseases may result when cells die that shouldn't or others live that should die. Modulation of apoptotic processes may offer valuable methods of treatment. It is now known that many pharmacologic agents (cytotoxic agents, hormones, anti-inflammatory drugs) are effective through inducing apoptosis of target cells. Virtually, all apoptotic drugs and radiotherapy programmes induce apoptosis in tumour cells and resistance to apoptosis is associated with treatment failure. These therapies also induce apoptosis in normal cells and side effects on bone marrows, gut and oral mucosa limit the dose that can be used. Many more treatment strategies are currently in preclinical trials and if future clinical studies are fruitful, this translation from basic science to clinical practice will be unique as it will affect not just one, but a broad range of disorders and many patients will benefit.

REFERENCES

Kerr, J.F., Wyllie, A. H., Curie, A.R (1992). Apoptosis: a basic biological Phenomenon with wide ranging implications in tissue kinetics. Br. J. Cancer 26:239-257

Leist, M and Jaatela, M. (2007). Four deaths and a funeral: from caspases to alternative mechanism". Nat. Rev. Mol. Cell Biol. 2(8): 589-98

Gerschenson, L.E and Rotello, R.J (2001) apoptosis: a general comment FASEB J. 5:2127-2128

Fadeel, B, Orrenius, S and Thwotovsky, B. (2007). Apoptosis in human disease: a new skin for the old ceremony. Biochem Biophys Rev commun 266 (3): 699-717

Meier, P., Finch, A and Evsan, G. (2008) "Apoptosis in development" Nature 407 (6805): 796-801

Rathmell, J.C and Thompson, C.B (2002) "Pathways of apoptosis in lymphocyte development, homeostasis and disease" Cell 109(suppl): 897-107

Fadeel ,B., Gleiss, B And Hogstraid, K et al (1999). "Phosphatidylserine exposure during apoptosis is a cell-type specific event and does not correlate with plasma membrane phospholipid scramblase expression" Biochem Biophys Res commun 266(2): 504-11

Richardson, H and Kumar, S. (2002) "Death to flies: Drosophila as a model system to study programmed cell death". J. Immunol methods 265(1-2): 21-38

Frohlich, K.U and Madeo, F (2009). "Apoptosis in yeast – a monocellular organism exhibits altruistic behvaiour" FEBS Lett 473(1): 6-9

Skulachev, V.P (2012). "Pragmmed death in yeast as adaptation". FEBS Lett 528(1-3): 23-6

Ameisen, J.C (2006)." On the origin, evolution and nature of programmed cell death: a timeline of four billion years" cell death differ: 9(4): 367-93

Adams, J.M (2003). Ways of dying: multiple pathways to apoptosis genes .Dev 17: 2481-2495

Data, E.R, Brunet, A and Greenberg, M.E (2004). "Cellular survival: a play in three Akts". Genes Dev 13(22): 2905-27

Ginsberg, D. (2004). E2F1 pathways to apoptosis. FEBS Lett 529(1): 122-5

Kroemer, G., Zamzam, N and Susin, S.A (1997) Mitochondrial Control of Apoptosis. Immunol Today 18(10): 44-51

Naismith, J.H and Sprang, S.R (2009). Modularity in the TNF-receptor family. Trends Biochem sci 23(2): 74-9

Sartorius,U., Schmitz, 1 and Krammer, P.H (2001) molecular mechanisms of Death-receptor mediated apoptosis. Chembiochem 2(1):20-9

Yamada, K.M and Araki, M (2007). Tumour suppressor PTEN: modulator of cell synalling, growth, migration and apoptosis.Cell sci 114 (pt 13): 2375-82

Dehault, J.B and sulvesen, G.S (2005) caspases: keys in the ignition of cell death". Chem. Rev 102 (12): 4489-500

Creagh, E.M and Martin, S. (2001) "Caspases: cellular demolition experts: Biochem Soc Trans 29 (pt 6): 696-702

Miura, M, Zhu, H and Rotello et al(1993). Induction of apoptosis in fibroblast by IL-1 beta-converting enzyme, a mammalian homolog of the Caenorhabditis elegans cell death gene ced -3. Cel 75 (4): 653-60

Wangs, S., Miura, M and Lung et al (1998) .Murine caspase -11, an ICE-Interacting protease is essential for the activation of ICE. Cell 92(4):50 1-9

Kuida, K., Haydar, T.F Kuan et al (2008) Reduced apoptosis and cytochrome c-mediated caspase activation in mice lacking caspase- 9. Cell 941(3) 325-37

Varfolomeer, E.E., schuchmann, M. and luna et al (1998). Targeted disruption of the mouse caspase -8 gene ablates cell death induction by the TNF receptor Fas/Apol and DR3 and is lethal prenatally. Immunity 9(2): 267-76

arnshaw, W.C, Martins, L.M and Kaufnam, S.H (1999). Mammalian caspases: structure, activation, substrates and functions during apoptosis. Annu Rev Biochem 68:383-424

Farber, J.L (1992). Biology of disease: Membrane injury and calcium homeostasis in the pathogenesis of coagulative necrosis. Lab invest 47:114-123

Cohen, G.M (1997) Caspaes: the executioners of apoptosis. Biochem J 326:1-16

Hockkenbery, D., Nunez, and Milliam et al (1990). Bcl-2 is an inter Mitochondrial membrane protein that blocks programmed cell death. Nature 348(6299) 334-6

Kaufman, S.H and Earnshaw, W.C (2007) Induction of apoptosis by cancer Chemotherapy. Exp Cell Rev 256(1):4-9

Bergamaschi, D., Samuels, Y and Jin, B (2004). ASPP 1 and APSS 2: Common Activators of p 53 family members .Mol Cell Biol 24:1341-1350

Bergamaschi ,D et al (2003). iASPP oncoprotein is a key inhibitor of p 53 conserved from worm to human. Nat Gene 33:162-167

Herudon, F.S, Hsu, H.C and Mountz, D. (2008). Increased apoptosis of CD45RO-T cells with ageing. Mechanism of ageing and development 94:123-134

Burchu, W., Oberhammer, F and Schulte-Hermann, R (1992). Cell death by apoptosis and its protective role in disease. Trends Phamacol sci 13: 245-251

Thompson, CB. (1995). Apoptosis in the pathogenesis and treatment of disease. Science 267:1456-1462.

Aringer, M and Wintersberger et al (2006). High levels of bcl-2 protein in circulating T lympholytes but not B lymphocyte of patients with systemic lupus erythematosus. Arthritis Rheum 37:1423-1430

Wooley, K.I, Gibson, P.G and Carty, K (2006). Eosinophil apoptosis and the resolution of airway inflammation in asthma. Ann J. Respir crit care Med 154:237-241

Boyce, J.A (1997). The pathbiology of eosinophilic inflammation .Allergy Asthma proc 18:293-300.

Tsujimoto, Y and Shimizu, S. (2000) VDAC regulation of the BCL-2 family of proteins. Cell death Differ 7(12): 1174-81

Clarke, A.R and Sanson, O.J (2003) Analyzing tumour suppressor activities in the murine small intestine Oncol Res 13:333-337

Iwadate, Y., Fujimoto, S and Tagawa et al (1996). Association of P53 gene mutation with decreased chemosensitivity in human glioma. Int. J. Cancer 69:236-240

Cory, S (2005). Regulation of lymphocyte suruvial by the BCI-2 gene family. An Rev immunol 13:573-543

Masunaga, A, Arai, T and Yoshitake, et al (1994) Reduced expression of apoptosis related antigens in thymuses from patients with myasthenia gravis. Immunol lett 39:169-17

Ascher, M.S, Sheppard, H.W and krowka et al (2003). AIDS as immune system activation. Key questions that remain. Adv Exp Med Biol 374:301-310.

Arends, M.J., Morris, R.G and Wyllie (2002). The role of the endonuclueases. AM J. Pathol 136:593-608.

Trump, B.F., and Bulger, R.F (1997) Studies of cellular injury in isolated

flounder tubules. Correlation between morphology and functions of control tubules and observation of autophagocytosis and mechanical cell damage. Lab invest 16:453-482

Vermes, I., Aaanen, C and Steffeus-Nakken et al (2008). A novel assay for apoptosis: flow cytometric detection of phosphatidylserine expression on early apoptotic cell using fluorescein labeled Annexin V. J. Immunol methods 184:339-351.