HAEMOPOISES

Chapter 1

CELL CYCLE

The cell cycle

The cell division cycle, simply the *cell cycle*. Dysregulation of cell proliferation is also the key to the development of malignant disease.

The cycle is divided in to the mitotic phase (M-*phase),* during which the cell physically divides, and *interphase* during which the chromosomes are duplicated and cell growth occurs prior to division. The M phase is further partitioned into classical *mitosis* in which nuclear division is accomplished, and *cytokinesis* in which cell fission occurs. Interphase is divided into three main stages:

G1 *phase:* cell begins to commit to replication

S *phase:* DNA content doubles and the chromosomes replicate and the *G2 phase* in which the cell organelles are copied and cytoplasmic volume is increased. If cells rest prior to division they enter a Go state where they can remain for long periods.

The cell cycle is controlled by two *checkpoints* which act as brakes to coordinate the division

 process at the end of the G1 and *G2* phases.

Two major classes of molecules control these checkpoints, *cyclin dependent protein kinases*

(Cdk) which phosophorylate downstream protein targets and *cyclins* which bind to Cdks

and regulate their activity. An example of the importance of these systems is demonstrated b

which results from the constitutive activation of cyclin D1 as a result of a chromosomal translocation.
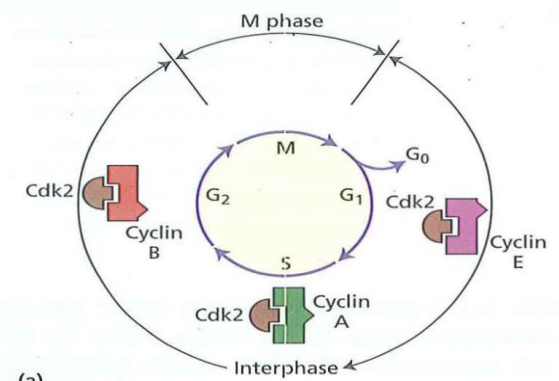
APOPTOSIS

Apoptosis is a regulated process of physiological cell death in which cells are triggered to activate intracellular proteins that lead to the death of the cell. Morphologically it is characterized by **cell shrinkage, condensation of the nuclear chromatin, fragmentation of the nucleus and cleavage of DNA at internucleosomal sites**. It is an important process for maintaining tissue homoeostasis in haemopoiesis and lymphocyte development.

Apoptosis results from the action of intracellular cysteine proteases called *caspases* which are activated following cleavage and lead to endonuclease digestion of DNA and disintegration of the cell skeleton.

There are **two major pathways** by which caspases can be activated.

1. Signalling through membrane proteins such as Fas or TNF receptor via their intracellular death domain. An example of this mechanism is shown by activated cytotoxic T cells expressing Fas ligand which induce apoptosis in target cells.

2. Via the release of cytochrome c from mitochondria. Cytochrome c binds to Apaf-1 which then activates caspases. The protein p53 has an important role in sensing DNA damage. It activates apoptosis by raising the cell level of BAX which then increases cytochrome c release. P53 also shuts down the cell cycle to stop the damaged cell from dividing. The cellular level of p53 is rigidly controlled by a second protein MDM2. Following death, apoptotic cells display molecules that lead to their ingestion by macrophages.

As well as molecules that mediate apoptosis there are several intracellular proteins that protect cells from apoptosis: BCL-2. BCL-2 is the prototype of a family of related proteins, some of which are anti-apoptotic and some, like BAX, pro-apoptotic. The **intracellular ratio of BAX and BCL-2 determines the relative susceptibility of cells to apoptosis** and may act through regulation of cytochrome c release from mitochondria. Many of the genetic changes associated with malignant disease lead to a reduced rate of apoptosis and hence prolonged cell survival.

APOPTOSIS
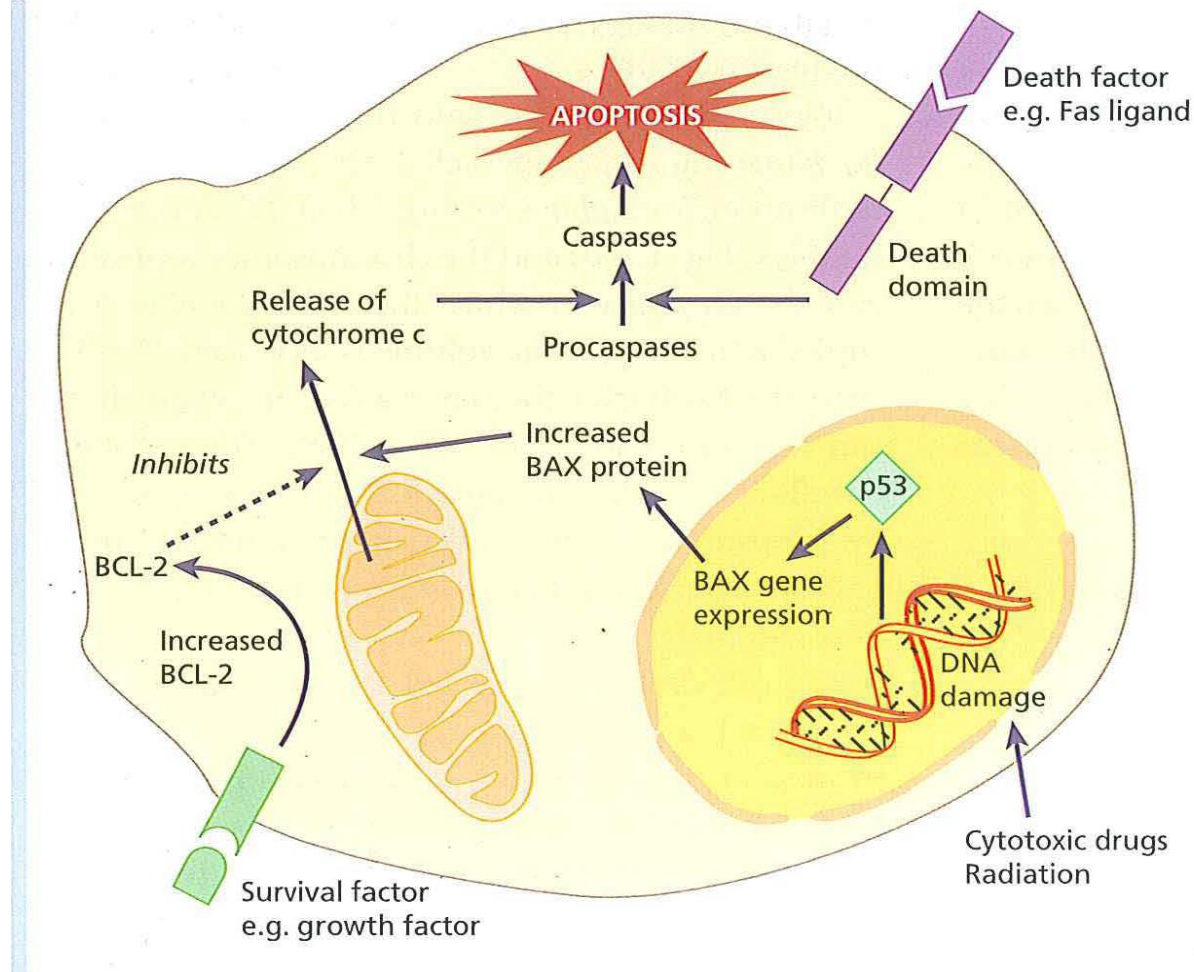
Fig. 1.11 Representation of apoptosis. Apoptosis is initiated via two main stimuli: (i) signalling through cell membrane receptors such as FAS or tumour necrosis factor (TNF) receptor; or (ii) release of cytochrome c from mitochondria. Membrane receptors signal apoptosis through an intracellular death domain leading to activation of caspases which digest DNA. Cytochrome c binds to the cytoplasmic protein Apaf-1 leading to activation of caspases. The intracellular ratio of pro- (e.g. BAX) or anti-apoptotic (e.g. BCL-2) members of the BCL-2 family may influence mitochondrial cytochrome c release. Growth factors raise the level of BCL-2 inhibiting cytochrome c release whereas DNA damage, by activating p53, raises the level of BAX which enhances cytochrome c release.

Transcription Factors and Adhesion Molecules

Transcription factors

Transcription factors regulate gene expression by controlling the transcription of specific genes or gene families. Typically, they contain at least two domains: a *DNA-binding domain* such as a leucine zipper or helix-loop-helix motif which binds to a

specific DNA sequence, and an *activation domain* which contributes to assembly of the transcription complex at a gene promoter.

Adhesion molecules

A large family of glycoprotein molecules termed adhesion molecules mediate the attachment of marrow precursors, leucocytes and platelets to various components of the extracellular matrix, to endothelium, to other surfaces and to each other. The adhesion molecules on the surface of leucocytes are termed receptors and these interact with molecules

(termed ligands) on the surface of potential target cells, Three main families exist:

1 Immunoglobulin **superfamily:**This includes receptors that react with antigens (the T-cell receptors and the immunoglobulins) and antigen-independent surface adhesion molecules.

2 Selectins: These are mainly involved in leucocyte and platelet adhesion to endothelium during inflammation and coagulation.

3 Integrins: These are involved in cell adhesion to extracellular matrix (e.g. to collagen in wound healing and in leucocyte and platelet adhesion).

The adhesion molecules are thus important in the development and maintenance of inflammatory and immune responses, and in platelet-vessel wall and leucocyte-vessel wall interactions. Expression of adhesion molecules can be modified by extracellular and intracellular factors and this alteration of expression may be quantitative or functional. IL-l, TNF, IFN-y, T-cell activation, adhesion to extracellular proteins and viral infection may all up-regulate expression of these molecules. The pattern of expression of adhesion molecules on tumour cells may determine their mode of spread and tissue localization (e,g. the pattern of metastasis of carcinoma cells or non-Hodgkin's lymphoma cells). The adhesion molecules may also determine whether or not cells circulate in the blood stream or remain fixed in tissues, They may also partly determine whether or not tumour cells are susceptible to the body's immune defences.