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Stem cell biology: Molecular control of normal hematopoiesis, leukemia and apoptosis

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Hematopoiesis gives rise to cells of different lineages during embryogenesis and throughout adult life and is an excellent model to study stem cell biology. Our establishment of a cell culture system for the clonal development of hematopoietic cells made it possible to discover the cytokines that regulate cell viability, multiplication and differentiation of different hematopoietic cell lineages and the molecular basis of normal and abnormal blood cell development.

The first cytokines discovered in this way are now called colony stimulating factors (CSFs). They also now include various other cytokines such as interleukins (ILs). There is a network of cytokine interactions and a cytokine cascade that couples growth and differentiation.

A network allows considerable flexibility and ready amplification of response to a particular stimulus. A network may also be necessary to stabilize the whole system. Malignancy can be suppressed in certain types of leukemic cells by inducing differentiation with cytokines that control normal hematopoiesis or with other compounds that use alternative differentiation pathways. The suppression of malignancy by inducing differentiation can bypass genetic abnormalities that give rise to malignancy. The results showed that leukemic cells can be epigenetically reprogrammed to regain normal behavior and created the basis for the clinical use of differentiation therapy. There is considerable plasticity in the developmental programs of normal and malignant hematopoietic cells.

Different CSFs and ILs induce cell viability by suppressing programmed cell death (apoptosis) and induce cell multiplication and these processes can be separately regulated. The same cytokines suppress apoptosis in normal and leukemic cells, including apoptosis induced by irradiation and cytotoxic cancer chemotheapeutic compounds, and apoptosis induced by wild-type p53. Apoptosis is controlled by a network of apoptosis-inducing and apoptosis-suppressing genes. Cytokines suppress apoptosis by changing the balance between apoptosis-inducing and apoptosis- suppressing gene products (Table 1).

Table 1.
Cytokine regulation of genes of the apoptotic machinery

Cytokine	Genes	
	Upreulation	Downregulation
IL-3	*bcl-2, bcl-x_L , mcl-1*	*bim, hrk,* Bad^{phos}, *Bax^{mit}*
IL-6	*bcl-x_L , mcl-1, FLIP*	*bcl-2, Bax*
IL-7	*bcl-x_L , mcl-1*	*bak,* Bax^{mit}
G-CSF	*Al, survivin*	
GM-CSF	*bcl-x_L , mcl-1, Al, survivin*	

Bad^{phos}, phosphorylation of Bad; Bax^{mit}, prevents Bax translocation to mitochondria.

Accumulation of wild-type p53 protein in cells following DNA damage is mainly due to post-translational events that inhibit the mdm-2 mediated p53 ubiquitination and proteasomal degradation. We have shown, together with Yosef Shaul and Gad Asher, that p53 protein stability is also regulated by the enzyme NADH quinone oxidoreductase 1 (NQO1). Inhibition of NQO1 activity induced p53 degradation and protected cells from wild-type p53- mediated apoptosis. Inhibition of NQO1 also caused degradation of a mutant p53 and this can be clinically useful for the therapy of tumors with mutant p53.

There are different pathways that regulate apoptosis induced by different compounds. Apoptosis induced by wild-type p53 can be suppressed by cytokines, various protease inhibitors, antioxidants and calcium mobilizing compounds. Suppression of apoptosis by cytokines and these other compounds is upstream of activation of caspases. Apoptosis induced by doxorubicin, vincristine, cycloheximide or cytokine withdrawal can be suppressed by cytokines, but not by some of these other inhibitors. This showed that cytokines can suppress pathways

of apoptosis that are not suppressed by these other agents. We have also shown that suppression or induction of apoptosis can be induced in the same cells by calcium-mobilizing compounds by opposing pathways that diverge downstream from calcium-activated calcineurin. The p38 mitogen-activated protein kinase (p38 MAPK) is involved in induction of apoptosis but not in its suppression by calcium-mobilizing compounds (Fig. 1).

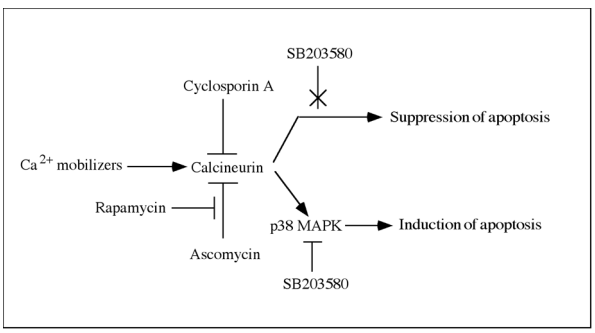

Fig. 1 *Model of calcineurin dependent pathways for suppression or induction of apoptosis. Arrow indicates pathway; T, suppression of pathway; and X, pathway not suppressed.*

Hematopoietic cytokines such as granulocyte CSF (G-CSF) are now used clinically to correct defects in hematopoiesis, including repair of chemotherapy-associated suppression of normal hematopoiesis in cancer patients and to correct defects in the production of granulocytes in children with congenital agranulocytosis. Injection of G-CSF also induces the migration of hematopoietic stem cells from the bone marrow to the peripheral blood, so that peripheral blood rather than bone marrow can now be used for hematopoietic stem cell transplantation. Cytotoxic cancer therapy could be improved by treatments that decrease the level of apoptosis-suppressing cytokines, and by decreasing expression of apoptosis-suppressing genes or inducing expression of apoptosis-inducing genes in cancer cells. The basic studies have thus provided new approaches to therapy.

Selected Publications

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Lotem, J. and Sachs, L. (1998) Different mechanisms for suppression of apoptosis by cytokines and calcium mobilizing compounds. Proc. Natl. Acad. Sci. USA 95, 4601-4606.

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Lotem, J., Kama, R. and Sachs, L. (1999) Suppression or induction of apoptosis by opposing pathways downstream from calcium-activated calcineurin. Proc. Natl. Acad. Sci. USA 96, 12016-12020.

Asher, G., Lotem, J., Cohen, B., Sachs, L. and Shaul, Y. (2001) Regulation of p53 stability and p53-dependent apoptosis by NADH Quinone Oxidoreductase 1. Proc. Natl. Acad. Sci. USA 98, 1188-1193.

Lotem, J. and Sachs, L. (2001) Cytokine control of developmental programs in normal hematopoiesis and leukemia. Oncogene, (in press).

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