Proceedings in Cases Relating to Hospital Medical and Dental Staff (HM(61)112). Originally circulated to health authorities in 1961 by the Ministry of Health, this document remains unchanged since that time. The fact that the health service has moved apace, as has society, seems to have gone unnoticed. The document establishes, though by no means clearly, a series of steps starting from preliminary investigation for the establishment of a *prima facie* case and progressing through to a formal inquiry. These inquiries are formal and legalistic, requiring a high standard of proof, with many rights enshrined for the consultant under investigation, including the ability to comment on the proceedings, to make a plea of mitigation, and to appeal to the Secretary of State. In practice this results in years of delay and excessive cost to the various health authorities. There are currently approximately 40 NHS consultants suspended on full pay pending such inquiries, and this could add up to a cost of £4 000 000 to the health service. There is certainly a case to be made for reviewing this disciplinary procedure which benefits neither the doctor nor the service. Until we have a clearer system which has the support of consultants and health authorities, difficult and often painful cases will continue to arise in which problems of competence may be confused with difficult relationships and interpersonal problems.

Teaching districts and university based medical schools have different disciplinary procedures. The teaching districts' consultant

contracts are district based and disciplinary procedures receive the attention of the district medical officer rather than being directed to the region, while teaching hospitals may direct problems through the university disciplinary procedure. Cases concerning professional conduct and competence in general practice will be directed to the family practitioner committee.

Conclusion

It seems that the procedure for dealing with a sick doctor has improved in recent years, although the reasons for doctors' greater vulnerability to particular health problems have received little attention, especially in medical schools. Where illness is not an issue, however, the position is clearly unsatisfactory and in need of change.

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Basic Molecular and Cell Biology

Stem cells in normal growth and disease

T M DEXTER

Mature blood cells have a short life span. Erythrocytes, for example, are functionally active for only a few months before being sequestered and destroyed, and granulocytes persist for only a few hours. In practical terms this means that about 3.7×10^{11} —that is, 370 000 million—mature blood cells are lost each day.¹ Cell loss of a similar magnitude undoubtedly occurs in other regenerating systems such as the skin and intestine.² Obviously, these cells need to be replaced to maintain the integrity of the various body tissues, and some mechanism is required to ensure that the means of cell production is maintained throughout a normal lifetime. The mature cells of the regenerating tissues are highly specialised cells which are themselves incapable of further growth. Their numbers must therefore be replaced through proliferation and development of more primitive cells, known as stem cells.

Origin of stem cells

Two models have been proposed to explain the persistence of stem cells throughout life.^{3,5} The first model suggests that a fixed number of primitive cells is laid down during embryogenesis to supply the body's needs throughout its lifetime. These cells are randomly recruited into proliferation, differentiation, and

development as required—like the recruitment of oocytes. As these stem cells proliferate there is a progressive loss in their ability to produce more stem cells or to act as founder cells of the various mature cell lineages—in other words, the stem cell pool declines with age. The second model also suggests that a small population of stem cells arises during embryonic development but that these cells can reproduce themselves (undergo self renewal) to produce daughter cells which retain the same proliferative and developmental potential as the original parental cells. In this model, therefore, the recruitment of stem cells into proliferation and development does not necessarily lead to a reduction in the number of stem cells. The weight of evidence strongly supports this second model of a self renewing stem cell population.^{6,7} The critical point here is that birth and death processes must be balanced: the input of cells into the stem cell compartment must be balanced by exit from the stem cell compartment as a consequence of differentiation and death.⁸ It follows that a prolonged, albeit minor, perturbation in this balance would have dramatic consequences. If too many stem cells underwent self renewal then the production of mature cells would be reduced to life threatening levels; if too many stem cells underwent differentiation and development then the small pool of stem cells would rapidly become exhausted. It is clear, however, that some degree of flexibility must be built into the system to allow for the extra proliferative demands imposed by stress conditions such as hypoxia or wound healing. How, then, are the regenerating tissues organised? Many lessons have been learnt from the haemopoietic system, and this will be used as an example, although a similar cellular organisation is seen in the skin epidermis and the intestinal epithelium.

Paterson Institute for Cancer Research, Christie Hospital and Holt Radium Institute, Manchester M20 9BX

T M DEXTER, DSC, MRCPATH, professor of haematology

Haemopoietic system

Haemopoietic stem cells arise in the yolk sac and then migrate to the fetal liver and subsequently to the developing bones.⁹ Within the bones the stem cells associate with a complex non-haemopoietic stromal cell matrix, which provides the milieu for further proliferation and development. The mature cells produced then traverse the endothelium lining the marrow sinuses and are released into the circulation.¹⁰ Extensive work has shown that the different kinds of blood cells—the neutrophils, monocytes and macrophages, eosinophils, basophils, erythrocytes, megakaryocytes (platelets), B lymphocytes, and T lymphocytes—are all derived from a common stem cell which is resident in the marrow.¹¹⁻¹³ In other words, the stem cell is multipotent. The self renewal ability of haemopoietic stem cells is readily seen during marrow transplantation. For example, about 50 000 marrow cells taken from a mouse and transferred into a potentially lethally irradiated recipient mouse will fully reconstitute the haemopoietic system. The reconstituted animals then have a normal life span, producing mature blood cells of donor origin. Since 50 000 marrow cells represent only 1/5000 of the total marrow reserve it follows that a normal mouse must have at least 5000 stem cells. In fact, the number is probably much higher, and a figure of 500 000 has been proposed.¹⁴ It is difficult to extrapolate these figures to man, but it is likely that stem cells are present in a similar concentration. It is clear that in order to produce the numbers of mature cells necessary to maintain normal steady state haemopoiesis an extensive cellular amplification must occur during development. Also, to produce the variety of mature cell types the stem cells must undergo differentiation into the various lineage restricted cell types. This may be readily detected using *in vitro* assay systems.¹⁵⁻¹⁷

When marrow cells are removed from the body and plated on to nutrient growth media they can be induced to proliferate and form colonies containing various cell types. Some of these colonies contain multiple lineages—for example, erythroid cells, neutrophils, and megakaryocytes—and are obviously derived from multipotent cells. Other colonies contain mature cells representative of only one or two cell lineages and are derived from bipotent or unipotent progenitor cells. By means of these *in vitro* systems it has been possible to build up a cell hierarchy and define the nature of the various differentiated cells (figure). Clearly, the cellular amplification occurs as a consequence of proliferation and development of the differentiated “committed” progeny of the stem cells.

Growth promoting agents

These *in vitro* systems have also been used to isolate and characterise a range of growth promoting agents and other factors which modulate proliferation and development of the blood

forming cells. All of the growth promoting agents have one thing in common: their continued presence is essential for the survival of haemopoietic cells *in vitro*. Marrow cells that are removed from the body and cultured in nutrient media in the absence of growth factors die. In the presence of growth factors they survive, proliferate, and mature into functional blood cells.¹⁵⁻¹⁹

Differentiation and development of stem cells. CFU-S=mouse spleen colony forming cell; CFC-Mix=a multipotential cell producing mixed myeloid cell colonies *in vitro*; GM CFC=granulocyte/macrophage colony forming cell; BFU E=a primitive erythroid progenitor cell; CFU E=a more mature erythroid progenitor cell; Eos CFC=eosinophil colony forming cell; Bas CFC=basophil/mast cell progenitor; Meg CFC=megakaryocyte colony forming cell.

The type of blood cells produced depends on the growth promoter, used (see table). Interleukin 3, for example, can stimulate proliferation and development of multipotent stem cells as well as more differentiated progenitor cells, which are restricted to the neutrophil, macrophage, erythroid, eosinophil, megakaryocyte, and mast cell lineages. Some growth promoters are restricted to a certain lineage—for example, macrophage colony stimulating factor will stimulate only the development of monocytes and macrophages from the appropriate progenitor cells. Other growth promoters, such as granulocyte and macrophage colony stimulating factor, are intermediate in their target cells. In other words, haemopoiesis *in vitro* can be induced by a variety of growth promoting agents, which show various degrees of restriction, but also extensive overlap, in their target cells.

Growth factors active on haemopoietic stem cells and committed cells

Growth factor	Species	Purified		Molecular weight (kilodalton)	Stem cells*	Target cells							Activity <i>in vivo</i>
		Native	Cloned			GM CFC	BFU E	Eos CFC	Meg CFC	Mast cells	CFU E	Mature cells	
Interleukin 3	Mouse	+	+	26	+	+	+	+	+	+	±	±	Yes
	Human	-	+	20	+	+	+	ND	ND	+	ND	+	ND
Granulocyte and macrophage colony stimulating factor	Mouse	+	+	14	±	+	±	+	+	-	-	+	Yes
	Human	+	+	19	+	+	+	+	+	-	-	+	Phase I/II trials under way
Granulocyte colony stimulating factor	Mouse	+	+	24	-	(G)	-	-	-	-	-	+	Yes
	Human	+	+	19	-	(G)	-	-	-	-	-	+	Phase I/II trials under way
Macrophage colony stimulating factor	Mouse	+	+	45	-	(M)	-	-	-	-	-	+	Yes
	Human	+	+	26	-	(M)	-	-	-	-	-	+	Phase I/II trials under way
Interleukin 4	Mouse	+	+	24	-	-	-	+	-	±	-	ND	ND
Erythropoietin	Mouse	-	+	Not known	-	-	-	-	-	-	+	-	ND
	Human	-	+	Not known	-	-	-	-	-	-	+	-	Yes, phase I/II trials now complete
Interleukin 1	Human	+	+	17	Studied in murine system; synergises with growth factors							-	Yes (radioprotective in mice)

For abbreviations see footnote to figure. ND=not done; G=granulocyte; M=macrophage.

*Stem cells refers to effects on CFU-S or CFC-Mix.

In some respects the overlap in biological activities could be considered to be redundant. This may not be the case, however, since in the haemopoietic system in particular (and probably other systems in general) we are dealing with interactive cellular networks, where the outcome of a response depends on the growth promoting agents to which the cells are exposed. Furthermore, it is important to bear in mind that these different factors not only facilitate proliferation and development of immature cells but also activate the function of mature cells.^{16,20} Granulocyte and macrophage colony stimulating factor and macrophage colony stimulating factor, for example, enhance the ability of macrophages to engulf and destroy bacteria and protozoa; granulocyte and macrophage colony stimulating factor and granulocyte colony stimulating factor induce increased protein synthesis and superoxide generation in mature neutrophils; and eosinophil colony stimulating factor facilitates maturation and mobilisation of eosinophils in response to nematode infections. Thus after local infection it is likely that the first line of defence entails the production of these growth factors by the surrounding tissue cells to recruit and activate circulating leucocytes. In more aggressive infections circulating endotoxins derived from bacteria act as a powerful stimulus for the production and release of various growth factors which, as their concentrations rise, begin to influence haemopoiesis in the marrow. The actual growth promoter produced probably reflects the inducing stimulus—that is, viral or bacterial infection, physical damage, and so on.

Apart from growth promoters, which act directly, other agents have been described which act in synergy with growth factors.^{21,22} Interleukin 1 is such an agent. Multipotent stem cells, for example, do not respond to macrophage colony stimulating factor; nor do they survive or develop in the presence of interleukin 1. But when stem cells are cultured in the presence of both macrophage colony stimulating factor and interleukin 1 they are stimulated to produce colonies containing mature macrophages. Similarly, interleukin 1 can synergise with granulocyte and macrophage colony stimulating factor and with interleukin 3 to "recruit" more stem cells to a state in which they can respond to the respective growth promoter²² (T M Dexter, unpublished observations). Thus interleukin 1 may have a major role in differentiation and commitment of stem cells. Furthermore, since most cells in the body produce interleukin 1 (and many cells in the body have receptors for interleukin 1) this molecule may be important not only in haemopoiesis but also in modulating growth and development in other tissues.

Effects of haemopoietic growth promoters on animals

Of fundamental importance is to determine the biochemical and molecular events which are driven by these various growth modifiers. Molecular cloning of the genes for many of these factors^{24,29} has facilitated the production of large amounts of material from bacteria, which has been used to study the effects of haemopoietic growth promoters on animals. The effects are dramatic. Interleukin 3 stimulates stem cell proliferation *in vivo*^{30,31}; granulocyte and macrophage colony stimulating factor stimulates marrow haemopoiesis and induces a fivefold to tenfold increase in circulating blood leucocytes³²; granulocyte colony stimulating factor induces a pronounced increase in the numbers of stem cells, committed myeloid progenitor cells, and circulating blood neutrophils (tenfold to twentyfold)^{33,34}; and interleukin 1 can protect mice from potentially lethal radiation without the use of a marrow graft.

From a clinical point of view several other observations are noteworthy. Firstly, infusion of granulocyte and macrophage colony stimulating factor in a monkey with aplasia caused by a viral infection led to a rapid increase in circulating lymphoid and myeloid cells and to a resolution of the aplasia during treatment with growth factor.³² Secondly, injections of granulocyte colony stimulating factor in mice completely prevented the leucocyte nadir seen after chemotherapy with cyclophosphamide.³³ Thirdly, administration of granulocyte and macrophage colony stimulating factor to monkeys after marrow transplantation led to a substantially earlier recovery

of neutrophil concentrations in the blood (D Nathan, personal communication). In other words, the administration of growth factors was of major benefit in these three examples of life threatening conditions.

The potential benefits of growth factors are obvious, and much work is being done to investigate the various diseases in which growth factors can be of clinical benefit. Indeed, the first phase I and II clinical trials are now in progress using granulocyte and macrophage colony stimulating factor and granulocyte colony stimulating factor in patients with solid tumours undergoing treatment with aggressive chemotherapy. As damage to the haemopoietic system induced by chemotherapy is dose limiting in many therapeutic regimens the objective in treating with growth factors is to maintain circulating leucocyte concentrations above a critical baseline. The early results are encouraging. Moreover, the work emphasises how an understanding of the structure of regenerating systems has led to the discovery of the various regulatory influences controlling its development and the use of these agents to alleviate damage and to promote function.

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