Abstract

Hematopoietic stem cells (HSCs) have the ability of both self-renewal and differentiation. After bone marrow damage, surviving host HSCs or transplanted donor HSCs are able to restore hematopoiesis and maintain it for a long time due to the self-renewal potential. HSCs reside in a specific microenvironment in the bone marrow, in stem cell niche, which supports their survival and controls their functioning.

In this study, we investigated the impact of bone marrow damage induced by increasing doses of irradiation on engraftment efficiency of transplanted donor repopulating cells. Using the CD45.1/CD45.2 congenic mouse model, we developed a new approach enabling estimation of surviving HSCs in damaged hematopoietic tissue. Its principle is in measuring of the donor chimerism resulting from transplantation of a defined dose of normal congenic bone marrow cells. The transplanted donor cells contain repopulating cells, progenitors (STRCs) and HSCs (LTRCs) that give rise to blood cell production which proceeds in parallel with that present in the host hematopoietic tissue. We applied this approach to monitor spontaneous regeneration of repopulating cells, including HSCs, in mice irradiated with a sublethal dose of 6 Gy or by a lethal dose of 9 Gy and rescued by syngenic bone marrow cells. This was accompanied by functional assays testing the transplantability of regenerating bone marrow cells, recovery of productive hematopoiesis, and analysis of the Lin^{low}c-kit^{+}Sca-1^{+} (LSK) population of the bone marrow which is highly enriched in progenitors and HSCs. LSK cells were further analyzed according to CD150 and CD48 markers.

Our results demonstrate that the damage caused by sublethal irradiation does not interfere with the engraftment of intravenously administered progenitors and HSCs; what is more, they are engrafted with very high efficiency. We experimentally demonstrated existence of different types of niches for progenitors and HSCs in the bone marrow. While niches for HSCs were available for transplanted repopulating cells for a relatively long time after both sublethal and lethal irradiation, niches for transplanted progenitors “closed” more rapidly. Although the cellularity of regenerating bone marrow normalized in approximately 20 days after irradiation, and it produced high numbers of blood cells, its repopulating capability was very low still after 30 days. Subfractions of the LSK population classified according to expression of CD150 and CD48 markers were significantly altered during the entire 30 day regeneration period following sublethal irradiation.