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NEGATIVE CONTROL OF HAEMOPOIESIS

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Several molecules which exert effects on the growth of normal haemopoietic cells have been described , which fulfill the definition of negative regulators. Some, including a non-aggregative variant of MIP-I∝, BB10010, are at present undergoing clinical trials. However, recent experimental protocols suggest that MIP1-∝, originally described as a stem cell inhibitor, has a complex role in the regulation of haemopoiesis. In addition to its classic property of blocking the progression of primitive cells to the S phase of the cell cycle, stimulatory effects on defined subpopulations of progenitor cells and of their ancestor cells (long-term culture initiating cells) have been documented: CD34+ cells which have MIP1-∝ receptors have a higher capacity to generate clonogenic progenitors than CD34+ cells which do not have detectable MIP1-∝ receptors when cultured for several weeks on marrow stroma.

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THE ROLE OF T CELLS, INDUCTION OF ANTINEOVASCULAR CHEMOKINES, AND APOPTOTIC FACTORS IN THE ANTITUMOR ACTIVITY OF IL-12/PULSE IL-2.

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We have reported that a regimen consisting of IL-12/pulse IL-2 can induce complete regression of established primary and/or metastatic murine renal carcinoma (Renca) in most treated mice. Most mice cured of their original tumor are resistant to tumor rechallenge, and depletion of $CD8^+$, but not $CD4^+$ or NK cells abrogates the antitumor activity of IL-12/IL-2. Administration of IL-12/IL-2 to tumor-bearing mice potently enhances IFN-γ production and antitumor activity is markedly reduced in IFN-γ -/- mice. Despite the clear role of T cells, limited local leukocyte infiltration is observed in the tumor bed of mice treated with IL-12/IL-2. Utilizing a novel latex infusion technique, we have demonstrated a grossly visible reduction in tumor vascularity, and histologic sections reveal a clear inhibition of tumor neovascularization and pronounced central coagulative necrosis in mice treated with IL-12/IL-2 as compared to vehicle treated controls. We have used RT-PCR to demonstrate that while IL-12/IL-2 administration does not appear to downregulate the expression of various proangiogenic genes (i.e. VEGF, TGF-β, angiogenin) in the tumor bed, it potently enhances expression of the genes encoding the IFN-γ-inducible, antiangiogenic chemokines IP-10 and MIG in the spleen, draining lymph nodes and tumor bed of mice treated with IL-12/IL-2. Similarly, production of IP-10 by activated splenocytes from tumor-bearing mice is increased by IL-12/IL-2 ex vivo. Concurrent administration of IP-10 serum attenuates the antitumor activity of IL-12/IL-2, and we are currently investigating the influence of combined neutralization of IP-10 and MIG. IL-12/ IL-2 also potently enhances the expression of genes capable of mediating tumor apoptosis/ necrosis. IL-12/IL-2 administration synergistically enhances circulating serum levels of tumor necrosis factor-α, and increases FAS/FAS-L gene expression in the tumor bed and FAS-L gene expression in the draining lymph nodes of tumor-bearing mice. The relative role of these factors and their dependence on IL-12/IL-2-induced IFN-γ production is currently under active investigation utilizing neutralizing antisera as well as IFN-γ and FAS-L knockout mice. These studies demonstrate a role for T cells and the modulation of tumor neovascularization in the antitumor activity of this regimen, and suggest a potential role for IP-10/MIG and FAS/FAS-L interaction in the overall therapeutic activity of IL-12/IL-2.