Prospects for Interleukin-2 Therapy in Hematologic Malignant Neoplasms

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ABSTRACT—Interleukin-2 (IL-2) is a regulator of diverse functions of the immune system that can induce regressions in some experimental and human tumors. These early findings suggest a potential role for IL-2 in the treatment of certain malignant neoplasms including lymphomas and leukemias. Advanced, rapidly growing tumors are generally not amenable to immunotherapy. Therefore, it is more likely that protocols with IL-2 will be used to prolong remission and prevent relapse in leukemia patients with minimal tumor load. Several approaches are currently being tested in animal experiments and clinical trials. Activation of tumor-reactive lymphocytes (specific or nonspecific) by IL-2 in vivo may eradicate residual leukemia in patients with occult disease. In vitro-propagated autologous or allogeneic leukemia-reactive T cells may be infused with IL-2 to facilitate the tumor destruction. The IL-2 enhances monoclonal antibody-dependent effector systems, such as antibody-dependent cell-mediated cytotoxicity in vivo. Monoclonal antibodies recognizing epitopes on leukemia/lymphoma cells could therefore be used with IL-2 to target nonspecific effectors to destroy tumor cells. Other cytokines appear to potentiate various antitumor activities of IL-2, including cytokicity of antigen-specific T lymphocytes or lymphokine-activated killer cells in vitro, and these combined effects may be exploited in clinical trials in which more than one cytokine is used simultaneously or in sequence. Finally, a stepwise completion of clinical protocols testing this immunologic approach in combination with other treatment modalities is necessary. Currently, it is not clear which, if any, of the many hematopoietic tumors are most likely to respond to IL-2 treatment. More information is needed, not only on IL-2 stimulation of host antitumor responses but also on the direct effects of IL-2 and IL-2-induced cytokines on hematopoietic neoplasms.—J Natl Cancer Inst Monogr 10:69–72, 1990.

Despite the advances in chemoradiotherapy and bone marrow transplantation, a considerable number of patients with all types of lymphoma and leukemia die of their disease. Recently, the availability of highly purified recombinant cytokines, such as the interferons, interleukins, TNF, and hematopoietic growth factors, opened conceptually new approaches to treatment of these diseases. Recombinant IFN-α has already become an important tool in the management of hairy cell leukemia, whereas cytokine therapy of other hematologic tumors is still in an experimental stage.

One of the molecules that shows promise is IL-2. This potent regulator of diverse functions of the immune system can induce antitumor responses in many experimental neoplasms including lymphomas. The mechanism of this antitumor activity is currently the subject of intensive investigations. Clinical testing has documented responses in IL-2-treated patients, but these occur only in a minority of cases and most are temporary. Crucial questions regarding the therapeutic use of IL-2 still need to be answered. Researchers conducting laboratory and clinical studies are currently investigating which malignant neoplasms to treat, at what stage, and with what therapeutic schedules.

In this review, we will discuss advances in IL-2 research relevant to its potential use in the therapy of leukemia and lymphoma and summarize future investigations that may be needed to define the role of IL-2, if any, in the treatment of hematologic tumors.

INTERLEUKIN-2 AND ITS RECEPTORS

As a 15,000-molecular-weight glycoprotein of 133 amino acids, IL-2 is synthesized mainly by activated T cells (1–3). Originally described as a T-cell growth factor, IL-2 can cause functional changes in T and B lymphocytes, natural killer cells, monocytes, and oligodendrocytes (4–7). Incubation of normal lymphocytes with high doses of IL-2 leads to generation of powerful cytotoxic cells (designated LAK cells) that can lyse most autologous and allogeneic tumor cells and cell lines as well as virally transformed cells and certain normal tissues (8, 9). The LAK activity is mediated by a heterogeneous population of cells, with most LAK cell precursors being CD8–, CD4–, CD16+, and NKH-1+ (10). Unlike the cell lysis by specific CTL, which recognize nominal antigen in association with major histocompatibility complex determinants, the cytotoxic action of LAK cells is not restricted by the major histocompatibility complex (11). Also, IL-2 allows the differentiation and proliferation of CTL, including tumor-specific killer T cells in animal models. Playing a central role in the regulation of the immune response, IL-2 can induce the synthesis and release of many other cytokines. In vitro activation of peripheral blood mononuclear cells by IL-2 induces mRNA expression and release of TNF-α, TNF-β, lymphotoxin, IL-6, and IFN-γ (12, 13).

The effects of IL-2 are mediated through specific receptors on the cell surface (4, 14). High-affinity IL-2 receptors (Kd ~ 10 pM) are formed by the association of two subunits, the Tac-subunit (mol wt = 55,000) recognized by the CD25 (anti-Tac)

ABBREVIATIONS: IL-2 = interleukin-2; LAK = lymphokine-activated killer; CTL = cytotoxic T lymphocytes; TNF = tumor necrosis factor; TIL = tumor infiltrating lymphocytes; IFN = interferon; Kd = dissociation constant.

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monoclonal antibody and the p75 subunit (mol wt = 75,000). The p55 or p75 subunit alone forms IL-2 receptors with low ($K_d \sim 10\ nM$) or intermediate affinity ($K_d \sim 100\ pM$), respectively (15). Both high- and intermediate-affinity receptors are involved in IL-2 signaling. The high-affinity receptor is expressed on activated T and B cells mediating a number of functional changes. The p75 molecule alone is expressed on large granular lymphocytes and mediates the induction of LAK activity (16). The expression of IL-2 receptors is not limited to normal lymphocytes. Cells from most acute leukemias (including T-, common, and non-lymphoid leukemias) as well as T-lymphoblastic lymphomas and peripheral T-cell lymphomas express the p75 usually without the p55 IL-2 receptor molecules (17). Chronic lymphocytic leukemia, hairy cell leukemia, and other differentiated B-cell neoplasms often express both p55 and p75 subunits (17). The precise functional significance of these IL-2 receptors is unclear. In some cases, IL-2 induces proliferation and immunoglobulin secretion in leukemia B cells and LAK cell-like cytotoxicity in neoplastic T cells (18–20). It remains speculative whether this differentiating activity of IL-2 could be useful in the treatment of leukemia.

INTERLEUKIN-2 AND LYMPHOKINE-ACTIVATED KILLER CELL THERAPY

Early preclinical studies with IL-2, with or without infusions of LAK cells activated in vitro with IL-2, have demonstrated striking antitumor effects in mice bearing immunogenic and nonimmunogenic tumors. With the immunogenic tumors, T cells presumably recognizing tumor-associated antigens were primarily responsible for this in vivo response; whereas non-T cells (presumably mediating the major histocompatibility complex-unrestricted LAK response) were principally involved in the destruction of the nonimmunogenic tumors (21, 22). Interestingly, a biphasic (low dose and high dose) therapeutic optimum was observed in treating mice bearing an experimental transplantable lymphoma with intraperitoneal IL-2 (23). The low-dose therapeutic activity was not observed in nude mice, which suggests that it was associated with a T-cell-mediated effect.

The initial clinical trials documenting antitumor effects in humans used protocols consisting of high-dose IL-2 and in vitro-activated autologous LAK cells (24). Since then, several groups (25, 26) have confirmed that different regimens in which IL-2 was used induced measurable regressions in approximately 10–30% of the patients with melanoma and renal cell carcinoma. Similar response rates have also been observed in smaller groups of patients with non-Hodgkin’s lymphoma and colorectal carcinoma (24, 27). Only occasional responses have been noted in patients with other malignant neoplasms. Because the first trials demonstrated a significant dose-related toxicity, several researchers attempted to modify the IL-2 dose and schedule (25, 26).

The search for regimens with enhanced antitumor activity and tolerable toxicity has been hampered by the lack of understanding of the IL-2-induced antitumor effects in humans. Preclinical and in vitro data suggest several possible mechanisms. It is conceivable that their relative importance may be different in different tumors. The direct tumor lysis by LAK cells can be demonstrated in vitro and in some animal models. However, LAK activity induced in the peripheral blood lymphocytes of IL-2-treated patients does not necessarily correlate with tumor response. This may be due to other variables involved, such as tumor size and susceptibility to LAK cells, heterogeneity of tumor cell and LAK cell populations, LAK cell mobility, vascular effects, etc. Therefore, it is not surprising that laboratory–clinical correlates are difficult for one to identify (27).

A second postulated mechanism of IL-2 therapy is the activation and expansion of tumor-specific T lymphocytes that are detectable in some human tumors (28). This is supported by data indicating that IL-2-activated TIL from some patients with melanoma can show a selective cytotoxic activity against the tumor from which they were isolated (28). Yet another important component of the antitumor effects of IL-2 may be mediated by the secondary cytokines released from stimulated cells. These molecules can exert immunomodulatory and anti-tumor activities that may be clinically important. The expression of mRNA for TNF-α and IL-6 can be induced by IL-2 therapy in patients’ peripheral blood lymphocytes, and increased levels of TNF-α and IFN-γ have been found in their plasma (12, 13). Many of the observed side effects that cannot be explained by direct activities of IL-2 or direct cytolysis of normal tissue by LAK cells have been ascribed to toxicity of secondary cytokines (12, 13). The in vivo interactions between these molecules and IL-2 are probably complex. In vitro IFN-γ regulates growth and differentiation of CTL and LAK cells (29, 30). It can also change the ability of leukemia and lymphoma cell lines to be lysed by LAK cells (31) and TIL (32). The diverse effects of TNF-α include direct cytotoxic and cytostatic activities on certain tumor cells and the synergy with IL-2 in the generation of LAK cells (33). Many other synergistic and antagonistic activities within the cytokine network may be important for the final outcome of IL-2 therapy.

Although IL-2 does not appear to exert any direct cytotoxic or cytostatic effect on melanoma, renal cell carcinoma, or other carcinomas, many hematopoietic tumors carry IL-2 receptor molecules on their surface (see above). In some of the more “mature” lymphoid tumors, IL-2 induces differentiation and/or proliferation in vitro, whereas cells from acute leukemias appear to be unresponsive to IL-2 (17, 18). Tumor cell growth due to IL-2 therapy has not been documented, but we (34) have observed persistent changes in the phenotype of circulating lymphoma cells from some patients after treatment with this cytokine. Currently, it is difficult for anyone to predict if malignant lymphoid cells might be induced to differentiate or proliferate upon stimulation by IL-2 treatment. Given the variability of these tumors and the lack of satisfactory animal models, some kind of in vitro screening may be useful prior to immunotherapy.

INTERLEUKIN-2 THERAPY IN HEMATOPOIETIC TUMORS

The iv administration of relatively toxic doses of IL-2, with or without LAK cells, induced regressions of advanced non-Hodgkin’s lymphoma in a minority of treated patients (24, 25). As was true in other tumors, these responses were not durable. Therefore, it seems unlikely that IL-2 as a single agent would play a major role in the therapy of advanced progressive lymphoma. Similarly, patients with rapidly proliferating acute leukemia would be unlikely to benefit from IL-2 alone. Nevertheless, its usefulness in minimal tumor load has been contem-
plated by several investigators (26, 36, 37). We know LAK cells that lyse autologous and allogeneic leukemia cells can be generated from patients in remission (36). Treatment protocols are being investigated for use in this patient group. Low doses of IL-2 administered to outpatients by constant iv infusion, or sc over many weeks can cause several-fold enhancement of LAK activity in peripheral blood lymphocytes and are generally well tolerated (Sosman JA, Hank JA, Moore KH, et al; Stein RC, Malkovska V, Gordon S, et al: Submitted for publication). The antitumor activity of such a regimen could be tested in an outpatient setting.

The idea of integrating IL-2 therapy into autologous bone marrow transplantation protocols has also been suggested (38). In such a setting, the tumor load is minimized, whereas the immunocompetent cells are preserved and infused with the marrow autograft following chemoradiotherapy. Gottlieb et al. (39) have demonstrated that IL-2 infusions can be safely administered to patients recovering from chemotherapy or autologous bone marrow transplantation for acute myeloid leukemia and multiple myeloma. In these patients, IL-2 did not significantly suppress the recovering hematopoiesis but induced LAK cells cytotoxic to clonogenic leukemia cells in vitro. Interestingly, Reittie and co-workers (40) suggested that IFN-γ-secreting activated killer cells with the capacity to lyse natural killer-resistant targets are normally generated 4-6 weeks after autologous bone marrow transplantation but do not appear after treatment with chemotherapy. The authors speculated that these cytokine-producing cytotoxic cells may be responsible for the lower risk of relapse after autologous bone marrow transplantation compared with chemotherapy alone. Investigators will find it interesting if the potentiation of this autologous “graft-versus-leukemia” activity with IL-2 leads to prolonged remissions.

Following allogeneic bone marrow transplantation, a well-described graft-versus-leukemia (41, 42) effect appears to prevent some leukemia recurrences. This effect may be mediated by IL-2 released endogenously as part of the allorecognition response of donor T cells against host tissues. The released IL-2 may activate LAK effector mechanisms and induce the secretion of other cytokines. Alternatively, donor T cells may be recognizing alloantigens (43) or leukemia-associated antigens (44) on the host leukemia cells and induce specific T-cell-mediated destruction. If the latter is the case, activation, selection, and propagation of such T cells in vitro may enable their infusion (with IL-2 infusion to prolong their in vivo effects) into patients with minimal leukemia burdens, as has been successful in mice (22). Propagation of such human clones, with apparent selective recognition of allogeneic leukemia cells, has now been demonstrated (44). Some autologous leukemia-reactive lines have also been propagated (45).

It has been well-established that other cytokines can potentiate various IL-2-dependent functions, including CTL and LAK-cell cytotoxicity. Many investigators are currently studying whether a better therapeutic index could be achieved by combining IL-2 with other immunomodulators, as has been shown in animal models (29).

The therapeutic effect of monoclonal antibodies in lymphoid tumors may depend on the recruitment of host effector systems inducing antibody-dependent cell-mediated cytotoxicity. Using a model in which lymphoma cells are transplanted into mice, researchers (46, 47) have demonstrated that IL-2 enhances monoclonal antibody-dependent effector systems in vivo. We have similarly documented a twofold-to-tenfold increase in in vitro antibody-dependent cell-mediated cytotoxicity when the effector cells tested are obtained from patients after 1 month of IL-2 treatment (Hank JA, Robinson RR, Surjus J, et al: Submitted for publication). Such results provide a rationale for using antileukemic monoclonal antibodies with IL-2 in clinical trials.

More information is needed on the direct functional effects of IL-2 and IL-2-induced cytokines on various leukemia and lymphoma cell populations. Specific receptors for these molecules have already been demonstrated on hematopoietic tumors. Studies of the signaling through these pathways may indicate new ways of influencing tumor cell growth and differentiation with cytokines or their analogues.

It is not clear which, if any, of the many hematopoietic neoplasms are most likely to respond to IL-2 treatment. The final therapeutic outcome may depend on both the stimulation of host antitumor responses and direct effects on hematopoietic tumor cells.

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