Suramin Prevents Binding of Interleukin 2 to Its Cell Surface Receptor: A Possible Mechanism for Imunosuppression

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ABSTRACT

Suramin, a polysulfonic naphthalene antihelminthic drug, inhibits proliferation of a variety of T-cell lines in vitro and induces immunosuppression in some human patients and thymic atrophy and splenic depletion in mice. Recent clinical trials indicate that suramin has activity against human tumors, indicating that it will be necessary to understand the mechanism by which suramin induces immunosuppression. The T-cell growth factor, interleukin 2 (IL2), is the major growth factor involved in regulating lymphoid differentiation and proliferation and thus regulates, to a major degree, the magnitude and duration of the immune response. We demonstrate herein that suramin induces a concentration-dependent decrease in binding of 125I-labeled IL2 to its receptor complex on human and murine T-lymphocytes. Binding of 125I-labeled IL2 to both M, 75,000 and M, 55,000 IL2 binding molecules was inhibited by suramin. Similar concentrations of suramin were required to inhibit binding of 125I-labeled IL2, IL2-induced tyrosine phosphorylation, and IL2-induced proliferation, suggesting that these processes may be linked. With murine cells, suramin-induced growth inhibition could be overcome completely by increasing the concentration of IL2, suggesting that suramin inhibited growth by competing for the IL2 receptor. With human cells, growth inhibition by suramin could only be partially overcome by increasing the concentration of IL2, suggesting that an additional growth-inhibiting mechanism is present. The ability of suramin to prevent binding of IL2 to its receptor was used to confirm that prolonged interaction of IL2 with its receptor is required to induce cell proliferation. Since IL2 plays a role in lymphocyte proliferation and differentiation, the ability of suramin to inhibit binding of IL2 to its receptor may explain, in part, the in vivo immunosuppressive activities of suramin.

INTRODUCTION

Suramin sodium, the hexasodium salt of 8,8'-[carbonylbis[iminom-3,1-phenylene]carbonyl]bis-1,3,5-naphthalenetrisulfonic acid, prevents binding of several growth factors, including epidermal growth factor, tumor-derived growth factor β, and platelet-derived growth factor, to their cell surface receptors (1–4). The ability of suramin to bind many growth factors may be due to its similarity to heparin sulfate (1, 5). Suramin inhibits coagulation in vivo, at least in part, through altering levels of heparin and dermatan sulfate (1, 5). In addition, suramin binds tightly to a variety of other proteins and has been demonstrated to inhibit reverse transcriptase, DNA polymerase, terminal deoxynucleotid transferase, urease, hexokinase, succinic dehydrogenase, and acid phosphatase (reviewed in Ref. 5).

Suramin inhibits in vitro growth of a number of cell lines, particularly those of lymphoid origin. However, whether this is due to preventing binding of growth factors or due to inhibition of intracellular enzymes is unclear (1–6). In mice, suramin induces profound depletion of splenic lymphocytes and thymic atrophy with little effect on other organs, suggesting a tropism for lymphoid cells (6). Because of its anti-reverse transcriptase activity, suramin has been used in trials for treatment of AIDS, wherein, although it decreased viral replication, it induced immunosuppression in some patients (7, 8). Its antiproliferative activity in vitro has resulted in Phase I therapy trials in patients with both lymphoid and nonlymphoid tumors where it has been demonstrated to inhibit tumor progression (9).

The T-cell growth factor, IL2, is the primary growth factor which induces proliferation of T-lymphocytes and thus regulates, to a major degree, the magnitude and duration of the immune response (reviewed in Refs. 10 to 12). IL2 also appears to be required for T-cell differentiation and expansion of precursor cells in the thymus (13, 14). IL2Rα (p55) binds IL2 with a low affinity (Kd of approximately 10⁻⁴ mol), whereas IL2Rβ (p75) binds IL2 with an intermediate affinity (Kd of approximately 10⁻⁹ mol) (10, 15–19). Binding of IL2 to both IL2Rα and IL2Rβ forms a high-affinity complex (Kd of approximately 10⁻¹¹ mol) (10, 15–19). IL2Rα, commonly called TAC, is a M, 55,000 glycoprotein which crosses the membrane once and has a short intracellular domain without significant homology to other growth factor receptors or signalling molecules (21).

The mechanism whereby activation of the IL2 receptor leads to cell proliferation is uncertain (reviewed in Refs. 22 to 25). We, and others, have demonstrated that IL2 does not induce phosphatidylinositol hydrolysis with resultant increases in cytosolic calcium (24, 26–28). In addition, we have demonstrated that activation of protein kinase C is not required for IL2-induced phosphorylation (29). Binding of IL2 to its high-affinity receptor or to the p75 IL2 binding molecule induces tyrosine phosphorylation of a series of intracellular substrates including the IL2 receptor complex (22, 23, 30, 31). This tyrosine phosphorylation is likely to play a role in IL2-induced proliferation.

Since suramin prevents binding of several growth factors to their receptors and is a potent inhibitor of lymphoid function and growth in vitro and in vivo, we tested whether suramin would prevent binding of IL2 to its receptor. Suramin inhibited binding of 125I-labeled IL2 to both high- and low-affinity receptors. As demonstrated by cross-linking studies, suramin inhibited binding of 125I-labeled IL2 to either IL2 binding chain independently. Inhibition of 125I-labeled IL2 binding paralleled in vivo IL2-induced proliferation as well as IL2-induced tyrosine phosphorylation. The inhibitory effect of suramin could be overcome completely in murine cells and partially in human cells by increasing the concentration of exogenous IL2. Inhibition of IL2 binding and function thus provides a potential biochemical basis for the in vivo immunosuppressive effect of suramin.

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2 The abbreviations used are: IL2, interleukin 2; IL2Rα, interleukin 2 receptor; IL2Rα, p55 or α chain of the IL2 receptor; IL2Rβ, p75 or β chain of the IL2 receptor; PBL, peripheral blood lymphocytes; ptyr, phosphotyrosine; PBS, phosphate-buffered saline; BSA, bovine serum albumin; SDS, sodium dodecyl sulfate; PAGE, polyacrylamide gel electrophoresis; PBL blasts, peripheral blood lymphoblasts.
MATERIALS AND METHODS

Media and Stock Solutions. Complete culture medium was RPMI 1640 (Gibco, Grand Island, NY) supplemented with 2 mM glutamine (Gibco), 5 x 10^-3 M 2-mercaptoethanol (Sigma Chemical Co., St. Louis, MO), gentamycin (Sigma), and 10% fetal calf serum (Sterile Systems, Inc., Logan, UT). Suramin was a kind gift of Dr. E. Shevach and was purchased from Mobay Corporation (New York, NY). Recombinant IL2 and an excipient buffer control were kind gifts of Cetus Corporation (Emeryville, CA). Phytohemagglutinin-P was obtained from Difco Laboratories (Detroit, MI). Rabbit affinity-purified, polyclonal, ptyr-specific antibodies and a monoclonal ptyr-specific antibody were kind gifts from Drs. Kamps and Sefton (32). In addition, ptyr-specific antibodies were affinity purified from serum [provided by T. Pawson (Toronto, Ontario, Canada)] from rabbits immunized with a 1:1:1 ratio of ptyr, alanine, and glycine polymerized and coupled to keyhole limpet hemocyanin coupled with EDAC (Calbiochem) by passage over a ptyr column and elution with phenyl phosphate essentially as described by Kamps and Sefton (32). These antibodies gave identical results to those provided by Dr. Kamps and Dr. Sefton. 125I-labeled IL2 was obtained from NEN and ICN (Montreal, Quebec, Canada). The nonreducible homobifunctional cross-linking reagent, disuccinimidyl suberate, was from Pierce Chemicals (Rockford, IL). All other reagents were from Sigma unless otherwise indicated.

Cells. MT-1, a human HTLV-1-infected cell line expressing only the M, 55,000 IL2-binding molecule (17) and YT 2C2 which expresses specific antibodies and a monoclonal ptyr-specific antibody were kind gifts of Dr. Kamps and Dr. Sefton (32). These antibodies gave identical results to those provided by Dr. Kamps and Dr. Sefton. 125I-labeled IL2 was obtained from NEN and ICN (Montreal, Quebec, Canada). The nonreducible homobifunctional cross-linking reagent, disuccinimidyl suberate, was from Pierce Chemicals (Rockford, IL). All other reagents were from Sigma unless otherwise indicated.

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Suramin Inhibition of IL2 Binding to IL2Rα and IL2Rβ. As indicated in Table 1, preincubation of activated human PBL blasts or murine CTLL-2 with suramin (1 mg/ml) inhibited binding of 125I-labeled IL2 to the surface of cells by approximately 90%. If cells were incubated with 125I-labeled IL2 prior to the addition of suramin, 125I-labeled IL2 binding was decreased by 50% with PBL blasts and 70% with CTLL-2 (Table 1), which indicated that suramin was less efficient at displacing IL2 than at preventing binding. Washing of cells at low pH (citrate, pH 3.5) (15, 37) resulted in a similar decrease in bound 125I-labeled IL2 (Table 1), demonstrating that the majority of the 125I-labeled IL2 was retained on the cell surface and was not internalized.

To determine if suramin was preventing binding to both the IL2Rα and IL2Rβ receptor chains, 125I-labeled IL2 was incubated with PBL blasts in the presence and absence of suramin. Following incubation, 125I-labeled IL2 was cross-linked to cells, cells were lysed, and proteins were separated on polyacrylamide gel electrophoresis. As indicated in Figure 1, PBL blasts contain SURAMIN BLOCK OF 1L2 BINDING

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<tr>
<th>Medium</th>
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<td>125I-labeled IL2 binding (cpm) of the following cell types</td>
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*p Mean ± SEM of 4 determinations in 2 independent experiments of 4 performed.

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approximately equal amounts of IL2Rα and IL2Rβ as indicated by the radiolabeled bands with approximate molecular weights of 90,000 and 70,000. This represents IL2 (M, 15,000) bound to IL2Rα (M, 55,000) and IL2Rβ (M, 75,000), respectively. As indicated in Fig. 1, preincubation with suramin inhibited binding to both IL2Rα and IL2Rβ as indicated by loss of both the M, 70,000 and 90,000 radiolabeled bands. Once again, maximal inhibition required 1 mg/ml of suramin.

To confirm that suramin was independently preventing binding to IL2Rα and IL2Rβ, 125I-labeled IL2 was incubated with MT1β and YT 2C2 cells which express exclusively IL2Rα and IL2Rβ, respectively (17). As demonstrated in Fig. 1, incubation of cells with suramin decreased labeling of the M, 70,000 band on MT1β and M, 90,000 band on YT2C2 (and thus radiolabeled IL2 binding), indicating that suramin inhibited IL2 binding to both IL2Rα and IL2Rβ independently.

In an attempt to determine if suramin decreased 125I-labeled IL2 binding by binding to IL2 or to the IL2 receptor, cells were incubated with suramin (1 mg/ml) for 1 h. Cells were either not washed (as above) or subsequently washed twice with medium to remove suramin from the medium. 125I-labeled IL2 was added for an additional 30 min. Without washing, suramin (1 mg/ml) decreased 125I-labeled IL2 binding by greater than 90% (7700 ± 270 cpm in the absence of suramin versus 260 ± 40 cpm in the presence of suramin). Following washing, 125I-labeled IL2 binding was only decreased by approximately 30% (6400 ± 270 cpm in the absence of suramin versus 4060 ± 240 cpm in the presence of suramin). This marked decrease in the effect of suramin following washing suggests that its primary effect was to bind IL2.

Suramin Inhibition of IL2-induced Tyrosine Phosphorylation. We and others have demonstrated that IL2 rapidly increases tyrosine phosphorylation of specific substrates in cells and that this correlates with IL2-induced cell proliferation (22, 23, 30, 31). Preincubation of PBL blasts (Fig. 2) and CTLL-2 (not presented) with suramin markedly inhibited IL2-induced tyrosine phosphorylation of M, 78,000 and 70,000–75,000 bands and decreased the IL2-induced tyrosine phosphorylation of a M, 110,000 band to resting levels. Since tyrosine phosphorylation occurs within seconds of binding of IL2 to its receptor (22, 23, 30, 31), suramin was able to prevent functional association of IL2 with its receptor. The concentration of suramin required to inhibit IL2-induced tyrosine phosphorylation was similar to that required to prevent IL2 binding (Table 1; Fig. 1) and IL2-induced proliferation (see below), suggesting that these processes are linked.

Suramin Inhibition of IL2-induced Proliferation. As measured by thymidine incorporation, suramin inhibited IL2-induced proliferation at similar concentrations to those required to prevent binding of IL2 to its receptor (Figs. 3 and 4). In both PBL blasts (Fig. 3) and CTLL-2 (Fig. 4), suramin induced a marked increase in the concentration of IL2 required to give half-maximal proliferation. In the absence of suramin, approximately 2 units/ml of IL2 induced half-maximal proliferation in both PBL blasts and CTLL-2. When concentrations of suramin which did not significantly alter IL2 binding (100 µg/ml) were added, IL2-induced proliferation was indistinguishable from that of cells incubated in the absence of suramin but in the presence of IL2. With CTLL-2, in the presence of 500 µg/ml of suramin, 10 units/ml of IL2 were required to give half-maximal proliferation and, in the presence of 1000 µg/ml of suramin, 100 units/ml of IL2 were required to give half-maximal proliferation. With PBL blasts, in the presence of 300 µg of suramin, at least 100 units/ml of IL2 were required to give half-maximal proliferation and, in the presence of 1000 µg/ml of suramin, even 1000 units/ml of IL2 were not sufficient to induce half-maximal proliferation. This suggests that incubation with 1 mg/ml of suramin resulted in a marked decrease in the amount of IL2 bound to its receptor.

With CTLL-2, suramin-induced growth inhibition was completely overcome by increasing the IL2 concentration as would be expected if growth inhibition induced by suramin were due to competition for the IL2 receptor. In contrast, with human PBL blasts, suramin-induced inhibition was only partially overcome by increasing the IL2 concentration. This suggested that the growth-inhibitory activity of suramin in PBL blasts was due only in part to competition for the IL2 receptor. The remainder of the growth inhibition was probably related to binding of suramin to intracellular enzymes (5, 6). The reason for this discrepancy between CTLL-2 and PBL blasts is not clear.
Fig. 2. Suramin inhibits IL2-induced tyrosine phosphorylation. PBL blasts (3 x 10^6 cells) were incubated with and without 100 units of IL2 in the presence or absence of suramin (either 100 or 1000 μg/ml) as indicated. Cells were then spun, resuspended, lysed with Laemmli buffer, and separated by SDS-PAGE as described in "Materials and Methods." The Western blot was developed with the monoclonal anti-ptyr antibody and exposed overnight at —70°C with intensifying screens. The results are representative of one of 3 similar experiments.

Suramin Inhibition of IL2-induced Proliferation When Added after IL2. Interaction of IL2 with its receptor is rapid, and a series of biochemical changes occur within seconds of addition of IL2 (22–26, 36). However, experiments wherein IL2 was removed from cells by washing with medium indicated that prolonged incubation with IL2 is required to commit cells to enter S phase (10, 12, 39, 40). However, because of the high affinity of the IL2 receptor and the slow-off rate of IL2 (10, 15), it was unlikely that all of the IL2 would be removed by washing and that significant concentrations of IL2 would be carried over into the second incubation. In addition, the washing process has the potential to damage cells and alter their activation state. The ability of suramin to competitively inhibit interaction of IL2 with its receptor on CTLL-2 cells allowed a reassessment of this question. As indicated in Table 1, suramin was able to displace IL2 from its receptors on CTLL-2 cells and would prevent binding of IL2 to newly expressed receptors. In addition, 1 mg/ml of suramin caused approximately a 50-fold decrease in interaction of IL2 with its receptor as indicated by inhibition of [3H]thymidine incorporation (Fig. 4). Therefore, 1 mg/ml of suramin would effectively compete with low levels of IL2 for binding to the IL2 receptor.

CTLL-2 cells were synchronized in G0-G1 by deprivation of IL2 (39, 40). Cells were stimulated with IL2 and 10 h later pulsed with [3H]thymidine for 2 h. This was the earliest time point that significant IL2-induced [3H]thymidine incorporation was detectable (not presented) and would represent first entry into S phase. Under these conditions, suramin inhibited entry into S phase as measured by [3H]thymidine incorporation at all times of addition (Fig. 5). Although 2, 10, and 100 units gave essentially identical proliferative responses in the absence of suramin (12-h time point in Fig. 5), suramin inhibited [3H]thymidine incorporation induced by lower concentrations of IL2 (2 or 10 units) to a greater degree than higher concentrations (100 units/ml), once again indicating the competitive nature of the inhibition.

Addition of suramin 1 h before or 1 h after low concentrations of IL2 (2 or 10 units) completely inhibited [3H]thymidine incorporation, indicating that a 1-h incubation with IL2 was
not sufficient to induce entry into S phase (Fig. 5). However, when suramin was added after a 3-h incubation with IL2, a small but significant increase in [\(^{3}H\)]thymidine incorporation was observed, suggesting that a 3-h incubation with IL2 is sufficient to commit a small population of cells to enter S phase. When suramin was added 7 to 9 h after IL2, a small but significant inhibition of thymidine incorporation occurred, indicating that not all of the cells had become committed to enter S phase. Therefore, although optimal entry into the cell cycle appears to require prolonged interaction of IL2 with its receptor, a 3-h incubation with IL2 appears to be sufficient to allow some cells to enter S phase. This confirms, through an alternative approach to cell washing, that commitment to the cell cycle requires prolonged interaction of IL2 with its receptor. However, it does indicate, in contrast to previous reports (10, 12, 38, 39), that IL2 may be continuously required to facilitate entry into S phase. The difference between the results in the two systems likely reflects the inability of washing with medium to fully remove IL2 bound to its high-affinity receptor and carryover of bound IL2 into the secondary cultures (10, 12, 38, 39).

**DISCUSSION**

Suramin induced a comparable concentration-dependent inhibition of \(^{125}\)I-labeled IL2 binding, IL2-induced tyrosine phosphorylation, and IL2-induced proliferation as measured by [\(^{3}H\)]thymidine incorporation in both human and murine IL2-dependent cell lines. This suggests that the three processes may be causally linked. With murine CTLL-2 cells, the inhibition of proliferation was reversed by increasing the IL2 concentration, indicating that the antiproliferative effect of suramin was due to competitive inhibition of IL2 binding. With human PBL blasts, the antiproliferative effect of suramin was only partially reversed by increasing the IL2 concentration. This suggests that a component of the effect of suramin on PBL proliferation is due to competitive inhibition of IL2 binding, but that at least a part of the effect is due to interference with another cellular process. Whether this is due to the reported ability of suramin to inhibit a variety of intracellular enzymes (5, 6) or as yet undiscovered modes of action is not clear.

A 30-min preincubation with suramin (1 mg/ml) prevented greater than 90% of binding of \(^{125}\)I-labeled IL2 to its receptor molecules on cells which expressed both IL2R\(\alpha\) and IL2R\(\beta\) (PBL blasts, CTLL-2) or which expressed exclusively IL2R\(\alpha\) (MTI1\(\beta\)) or IL2R\(\beta\) (YT 2C2). However, if \(^{125}\)I-labeled IL2 was allowed to bind to cells prior to addition of suramin, the amount of radiolabeled IL2 associated with cells, in particular PBL, was decreased to a lesser degree than if suramin was added prior to \(^{125}\)I-labeled IL2. Since the \(t_0\) for dissociation of IL2 from its high-affinity receptor is approximately 20 min (10, 15), the ability of IL2 to decrease the amount of prebound IL2 associated with the cell may represent dissociation of IL2 and prevention of rebinding of IL2 rather than displacement of bound IL2.

Since washing of suramin prior to the addition of \(^{125}\)I-labeled IL2 markedly decreased the effect of suramin on \(^{125}\)I-labeled IL2 binding, it seems most likely that suramin decreases binding of \(^{125}\)I-labeled IL2 to cells by binding \(^{125}\)I-labeled IL2 directly. However, it is also possible that suramin binds to the IL2 receptor with a low affinity and that washing displaces much of the suramin from the cell surface. This is supported by the requirement for high quantities of suramin to prevent IL2 binding.

Little is understood about the mechanisms whereby growth factors bind to their receptors and alter receptor conformation, so that a signal is transmitted across the cell membrane. The ability of the highly charged suramin molecule to block binding of a variety of growth factors to their receptors suggests that a common charge motif may be involved in receptor binding. This is also suggested by the ability of low pH to dissociate binding of growth factors from their specific cell surface receptors. Suramin similar to heparin sulfate, which also binds to many growth factors, is a polysulfonated molecule. This may explain the ability of suramin and heparin sulfate to bind to a variety of growth factors (9). Suramin therefore provides a probe to study interaction of growth factors with their receptors and also a tool to determine the role of receptor internalization and the kinetic requirements for IL2 receptor interaction and cell proliferation.

Although IL2 rapidly (seconds) induces biochemical changes in cells including increases in intracellular pH and tyrosine phosphorylation (22–25, 36), experiments wherein IL2 is washed from cells indicate that prolonged (hours) incubation with IL2 is required to induce cell cycle progression (10, 12, 38, 39). Since suramin inhibits IL2-induced proliferation in
CTLL-2 cells primarily by competing for the IL2 receptor, suramin was used as an alternative method to determine if prolonged interaction of IL2 with its receptor is required for cell cycle progression. When suramin was added 1 h prior to addition of IL2 or 1 h after addition of IL2, similar degrees of growth inhibition occurred. Suramin inhibited cell cycle progression despite rapid internalization of IL2 at 37°C (10, 15) and inability of delayed addition of suramin to decrease radio-labeled IL2 associated with cells incubated at 37°C (not presented). This likely represents internalized IL2, as washing with citrate (pH 3.5) induced a similar decrease in 125I-labeled IL2 binding. This confirms previous reports that prolonged IL2 receptor occupancy is required to induce entry into S phase (12, 38, 39). However, in contrast to results wherein IL2 was washed from cells, even a 3-h incubation with IL2 is sufficient to induce some CTLL-2 cells to enter S phase. The difference between the results with suramin and the washing experiments of Cantrell and Smith (12) probably results from the species utilized and the more rapid entry into S phase of murine CTLL-2 cells compared with human lymphocytes. In addition, in both the experiments with human and murine cells (12, 28, 39), washing with medium is not likely to be sufficient to displace IL2 bound to its high-affinity receptor, and some IL2 was likely carried over into the secondary incubations. Similar to suramin, washing with citrate (pH 3.5), which would remove IL2 bound to its high-affinity receptor (37), several hours after addition of IL2 decreases entry into S phase (not presented). Therefore, addition of suramin may give a more accurate reflection of the duration of interaction of IL2 with its receptor which is required to induce cell cycle progression.

Suramin has been demonstrated to be a potent inhibitor of in vitro cell growth (6). This in vitro activity of suramin has led to its use in therapy trials of malignant disease (9). Although suramin appears to be active in vivo in preventing tumor progression, it is not clear whether this is due to its antiproliferative activity or to its ability to induce coagulation abnormalities (9). Nevertheless, the results presented herein suggest that, in cancer patients treated with suramin, lymphoid function must be monitored to ensure that patients do not become immunocompromised with a worse prognosis of their cancer.

Due to its ability to inhibit reverse transcriptase, suramin has also been used in preliminary trials in AIDS (7, 8). In these patients, suramin induced a profound immunosuppression which appeared to worsen the disease (7). This correlated with the ability of suramin to deplete splenocytes and cause thymic atrophy in mice without exerting obvious deleterious effects on other organ systems (9). Since IL2 is necessary for thymic function and lymphoid expansion (13, 14), at least a portion of the immunosuppressive effects of suramin may be due to the ability of suramin to prevent binding of IL2 to its cell surface receptor. However, it is difficult to dissociate the effects of suramin on IL2 function from its effects on enzymes required for cell growth and function. The lymphoid tropism of the in vivo toxicity of suramin does suggest that the activity may be directed against IL2 function or against a lymphoid-specific enzyme. Although previous studies have demonstrated that suramin inhibits terminal deoxynucleotide transferase, which is a relatively lymphoid-specific enzyme, the presence or absence of terminal deoxynucleotide transferase in lymphoid cell lines did not correlate with the ability of suramin to inhibit cell growth in vitro, arguing against inhibition of terminal deoxynucleotide transferase as the mechanism for the preferential action of suramin on lymphoid cells (9). Similarly, although we have demonstrated that suramin inhibits IL2-induced growth of T-lymphocytes, suramin also inhibits growth of lymphoid cell lines which do not require exogenous IL2 (9). Inhibition of IL2 function in T-lymphocytes may combine with the generalized enzyme inhibition to explain the in vivo tropism of suramin for lymphoid cells.

In summary, suramin prevents binding of IL2 to both IL2Rα and IL2Rβ. Similar concentrations of suramin were required to block binding of 125I-labeled IL2 to its receptor, IL2-induced tyrosine phosphorylation, and IL2-induced proliferation. Suramin thus provides a tool to probe the mechanism by which IL2 binds to its receptor and by which IL2 cell proliferation.

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