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Novel Gamma-Chain Cytokines as Candidate Immune Modulators in Immune Therapies for Cancer

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Abstract

Cytokines that signal through the common-gamma chain are potent growth factors for T cells and natural killer cells. Interleukin (IL)-2, the γ c prototype, can mediate antitumor effects as a single agent or in the context of multimodality regimens but is limited by side effects and a propensity for expansion of regulatory T cells. IL-7, IL-15, and IL-21 each possess properties that can be exploited in the context of immunotherapy for cancer. Each has been demonstrated to mediate potent vaccine adjuvant effects in tumor models, and each can enhance the effectiveness of adoptive immunotherapies. Although the overlap among the agents is significant, IL-7 is uniquely immunorestorative and preferentially augments reactivity of naive populations, IL-15 potently augments reactivity of CD8⁺ memory cells and natural killer cells, and IL-21 preferentially expands the inflammatory Th17 subset and may limit terminal differentiation of effector CD8⁺ cells. Clinical trials of IL-7 and IL-21 have already been completed and, so far, demonstrate safety and biologic activity of these agents. Clinical trials of IL-15 are expected soon. Ultimately, these agents are expected to be most effective in the context of multimodal immunotherapy regimens, and careful clinical trial design will be needed to efficiently identify the proper doses, regimens, and settings in which to exploit their biologic properties for therapeutic gain.

Keywords

gamma c cytokines; IL-7; IL-15; IL-21; cancer immunotherapy

Immune responses to tumor antigens can be induced in patients with cancer using a variety of vaccine strategies, including genetic vectors, peptides, and dendritic cells. Despite this, tumor vaccines alone are generally not potent enough to shrink established tumors, and although some trials of tumor vaccines have demonstrated clinical benefit adjuvantly, this is not a universal finding.¹ New approaches have become available to modulate endogenous immune responses using genetically modified T cells to target a wide array of tumors, but the effectiveness of these therapies remains limited, in part, by poor persistence of the transferred cells in vivo. Immunomodulatory agents have also shown promise against some

established tumors, but response rates of single agents remain low.² Together, the clinical data gleaned from a variety of models suggest that future progress in tumor immunotherapy will require multimodal regimens that incorporate agents to augment the strength and duration of immune responses. This can be accomplished by enhancing the number, quality, diversity, or longevity of tumor immune effectors and/or by diminishing suppressive factors that mediate tumor immune escape.

Cytokines are among the most promising agents available for potentiating the weak immune responses induced by current tumor vaccines and immunomodulators, and for augmenting persistence and functionality of adoptively transferred cells. Cytokines that signal through the common cytokine gamma chain (γ_c) can enhance the number, quality, diversity, and longevity of T cells and natural killer (NK) cells. Interleukin (IL)-2, the γ_c prototype, mediates antitumor effects in some settings but has significant limitations, including augmentation of regulatory T cell number and function.³ Recent studies demonstrate that other γ_c cytokines, namely IL-7, IL-15, and IL-21, have properties that could be exploited to amplify antitumor T cell and NK immunity. In this review, we will briefly discuss the basic biology of IL-7, IL-15, and IL-21 and then review translational and early clinical studies illustrating their potential to amplify cell-based immunity toward cancer.

INTERLEUKIN-7

IL-7 Biology and Preclinical Studies

IL-7 is required for development and survival of T cells, is important but not required for human B-cell development, and plays no significant role in NK-cell development and survival. IL-7 is produced primarily by stromal and parenchymal cells⁴ with lesser amounts produced by dendritic cells; IL-7 is not produced by B cells or T cells. The IL-7 receptor is a heterodimer comprising the common γ_c and IL-7R α . IL-7R α also signals thymic stromal lym-phopoietin (TSLP). Unlike receptors for most γ_c cytokines, (absent on resting cells but upregulated on activation), the IL-7R is expressed on most resting lymphocytes, downregulated after T-cell activation, then reexpressed on effector cells destined for the memory cell pool⁵ and on central memory T cells. Because IL-7 is produced by nonimmune cells in the absence of immune activation, is measurable in sera from healthy individuals, and the IL-7R heterodimer is present on most T cells, IL-7 can be conceptualized as a homeostatic cytokine whose signals are continuously available and play an important role in maintaining T-cell homeostasis.

The corollary to the central role of IL-7 in T-cell homeostasis is that the availability of IL-7 is increased during lymphopenia.^{6–8} Increased IL-7 availability plays a central role in driving immune reconstitution through homeostatic peripheral expansion, which involves exaggerated responses to cognate antigen,⁹ increased antigen-independent cycling of memory populations, and proliferative responses to low affinity antigens comprising both self-antigens and crossreactive environmental antigens.^{10–12} IL-7 is notable for its immunorestorative properties, its ability to augment both CD4 and CD8 expansion, its tendency to induce cycling of naive T cells with broad repertoire diversity, and its ability to amplify low affinity, subdominant immune responses such as those required for antitumor immunity. In contrast to the immune activating effects described earlier, IL-7 signaling on

dendritic cells has also been recently implicated in a regulatory axis that controls CD4 homeostatic expansion during lymphopenia.¹³ Cell-specific effects of IL-7 are summarized and compared with other γ c cytokines in Table 1.

Several studies have demonstrated the capacity of IL-7 to enhance antitumor effects of adoptively transferred cells when used to culture effectors ex vivo^{14,15} or when tumors were genetically engineered to express IL-7 in vivo.^{16–18} In several studies, IL-7 outperformed IL-2 for ex vivo expansion of antigen-specific T cells, in terms of both cell yield and functionality after adoptive transfer.^{19,20} When administered systemically in animal models, IL-7 is also a potent vaccine adjuvant,^{21,22} which outperforms IL-2 when the 2 agents are compared directly. In the study by Melchionda et al, both IL-7 and IL-15 augmented immunodominant CD8 responses to a similar degree; however, IL-7 had more potent effects on CD4 effectors and more potently augmented subdominant antigens than IL-15.²³ Similar to IL-15, IL-7 therapy provided as a short course during vaccination provided long-term benefits for memory cell survival, which persisted long after the cytokines were discontinued. A more recent study confirmed the capacity of IL-7 to enhance long-term memory cell responses, noting that this occurred after lentiviral-based tumor vaccination but not after peptide-based tumor vaccines,²⁴ presumably because of upregulation of IL-7R α on effector cells after lentiviral but not peptide-based vaccination. Nanjappa et al²⁵ noted that IL-7 enhancement of memory cell responses is dependent on timing of administration, with IL-7 therapy administered during the contraction phase more effective than when the cytokine was administered during the initial expansion phase of lymphocyte choriomeningitis virus (LCMV) infection.

Work by Pellegrini et al dissected the effects of IL-7 in a murine islet cell tumor model that expresses LCMV glycoprotein. In this model, LCMV infection yields limited antitumor effects, but coadministration of IL-7 with LCMV infection prolonged survival, decreased tumor burden and increased tumor T-cell infiltration. As expected based on the capacity of IL-7 to expand T cells, this was associated with substantial increases in the number of antigen-specific T cells in vivo. In addition, however, these authors identified immunomodulatory effects of IL-7 that had not previously been appreciated. Specifically, increased numbers of tumor infiltrating Th17 cells and NK cells and increased serum levels of IL-6, IL-12, IL-17, and IL-1 α were found when IL-7 was administered after LCMV infection compared with LCMV infection without cytokine. Interestingly, IL-7 did not induce inflammatory mediators in the absence of viral infection. T cells from IL-7-treated mice in this model were less susceptible to Treg and TGF β -mediated inhibition and show diminished *Cbl-b* expression as a result of Nedd4 upregulation. Given that *Cbl-b* is a negative modulator of TCR signaling, IL-7 modulation of *Cbl-b* may be the mechanism by which IL-7 enhances T-cell responses to weak antigens. In summary, in addition to confirming the known effects of IL-7 on T-cell expansion and persistence, this work identified several novel mechanisms through which IL-7 augmented antitumor effects. It also clearly illustrated that potent IL-7-mediated antitumor responses are exquisitely dependent on concomitant administration of an effective vaccine because IL-7 had no significant antitumor effect in the absence of LCMV infection in these studies.

IL-7 Clinical Trials

Several clinical studies of rhIL-7 have been completed and are currently underway, and therefore, emerging data are available regarding the clinical potential of this agent. Thus far, rhIL-7 has been well tolerated, with no evidence for capillary leak syndrome. Toxicities have been predominantly low-grade fever, myalgias, transient flu-like symptoms, and self-limited transaminitis.²⁶ Biologic effects have been observed in doses ranging from 3 to 60 µg/kg, the highest dose studied thus far. Essentially, all studies have demonstrated dose-dependent increases in T-cell number following rhIL-7, with substantial increases in both CD4 and CD8 cells.^{27–30} Early B-cell progenitors are also transiently increased, usually within the bone marrow but occasionally seen in the peripheral blood,²⁶ and lesser increases in NK-cell numbers have been observed. In addition to changes in peripheral blood lymphocytes, rhIL-7 increases the size of the secondary lymphoid organs, including spleen and lymph nodes, which can be visualized by computed tomography scanning or positron emission tomography imaging.²⁷ Increases in peripheral T-cell numbers seems to result primarily from peripheral effects including enhanced cycling and diminished apoptosis, rather than increased thymopoiesis. Notable is preferential expansion of rhIL-7 of naive T cells, which can enhance the repertoire diversity independent of age and independent of thymopoiesis.²⁷ In addition, rhIL-7 has minimal effects on expansion of Tregs because of low IL-7Ra on this subset,^{31,32} which results in relative decreases in the frequency of Tregs in rhIL-7-treated subjects.²⁸ This is in contrast to rhIL-2, which preferentially expands Tregs.^{3,33} Thus far, mature clinical data are not yet available to evaluate its effectiveness as a vaccine adjuvant or as a supportive cytokine for adoptive immunotherapy, but these studies are anxiously awaited. Clinical trials with IL-7 are summarized and compared with other γ c cytokines in Table 2.

INTERLEUKIN-15

IL-15 Biology and Preclinical Studies

IL-15 was identified as a non-IL-2 growth factor that signaled through IL-2R β and γ c.^{34–36} It has structural similarities to IL-2 but distinct biologic properties.^{37–39} Although IL-2 is produced by activated T cells, IL-15 is produced by a myriad of cell types, both bone marrow derived and nonbone marrow derived.^{40,41} Unlike the other γ c cytokines, which function as soluble ligands, IL-15 largely functions as a cell associated cytokine bound to IL-15R α expressing cells, predominantly monocytes^{42–45} and dendritic cells.⁴⁶ Thus, interleukin-15 signaling occurs when a cell-associated IL-15/IL-15R α complex binds the IL-2R β γ c heterodimer on activated and memory T cells or NK cells. IL-15 is required for NK-cell development and maintenance and maintenance of the CD8 memory pool.^{47,48} IL-15 is not required to generate memory CD8⁺ T cells, but their survival depends on this factor.^{49–52} IL-7 can substitute for IL-15 in this regard in lymphopenic hosts where elevated IL-7 levels occur⁵³ or when IL-7 is overexpressed in T-cell replete hosts⁴⁹ but not under normal conditions. IL-15 is also necessary for homeostasis of NKT cells.⁵⁴

IL-15 potently costimulates CD8⁺ T cells, and IL-15 administration expands the memory pool. Several studies have demonstrated that IL-15 therapy increases the size of the effector and memory pool generated in response to infection or immunization.^{23,55–57} For instance,

IL-15 enhances CD8⁺ T-cell immunity in mice infected with *T. gondii*,⁵⁶ IL-15 transgenic mice generate higher numbers of effectors after infection with *L. monocytogenes*,⁵⁷ and therapy with either IL-7 or IL-15 improves survival of *M. tuberculosis* infected mice.²² Thus, IL-15 enhances the immune response generated to intracellular pathogens, leading to increased eradication in the short term. Although augmentation of NK-cell expansion and survival may contribute to these effects, T cells contribute substantially as well. IL-15 therapy also enhances T-cell responses to DNA vaccines,^{55,58} and dendritic cell vaccines in mice,^{23,59} and influenza and tetanus vaccines in rhesus macaques.⁶⁰ The transpresentation of IL-15 has led to the insight that therapies aimed at enhancing availability of this agent in vivo are more effective when IL-15 is provided access to IL-15R α .^{61–63}

In several studies evaluating vaccine adjuvant and antitumor effects, IL-15 was directly compared with IL-2 and demonstrated to be superior.^{23,55,60} In 1 study, evaluation of the effect of IL-15 on immunodominant and subdominant antigens demonstrated that IL-15 more potently augmented responses to subdominant antigens, when compared with dominant antigens, similar to the effects observed when IL-7 is administered in the setting of vaccination. This is particularly pertinent for tumor antigens, which are generally weak antigens, and which may be better modeled by subdominant antigens. In addition to potent effects on the T-cell effector pool, 2 studies undertook careful analysis of the effects of IL-15 on the long-lived memory pool. In both, treatment with rhIL-15 at the time of immunization led to durable increases in the survival of the memory pool, an effect which remained long after the therapeutic levels of IL-15 had cleared.⁵⁵ Interestingly, a recent study also implicated IL-15 in enhancing humoral immune responses to a dendritic cell vaccine in a CD4⁺ independent manner, which contributed to antitumor effects.⁶⁴

The potent survival of IL-15 and differentiation signals for CD8 memory cells and NK cells render it an attractive candidate for use in the context of adoptive immunotherapy for cancer. Preclinical models have confirmed its activity in this regard. For instance, T-cell receptor transgenic cells specific for the gp100 melanoma antigen cells cultured in the presence of IL-15 ex vivo were more potent than cells cultured in the absence of cytokines and more potent than cells cultured with IL-2 in inducing regression of established melanomas in a murine melanoma model.⁶⁵ Similarly, IL-15 was superior to IL-2 after adoptive transfer of antigen-specific T cells in murine plasmacytoma,⁶⁶ and transduction of tumors with IL-15 or tumor-specific T cells with IL-15⁶⁷ induces potent antitumor effects.^{65,67} Similarly, ex vivo culture of tumor reactive CD8⁺ T cells found in tumor draining lymph nodes with IL-15 led to differentiation toward a central memory phenotype, directly implicating IL-15 in this process.⁶⁸ Thus, substantial preclinical data demonstrate that IL-15 is a potent adjuvant for adoptive T-cell therapy, both when used to culture cells ex vivo and when administered in vivo after transfer.

NK cells are less proliferative than T cells; however, recent work suggests that substantial numbers of NK cells can be generated for use in adoptive therapy using artificial antigen-based culture conditions.⁶⁹ IL-15 substantially augments ex vivo NK expansion in these systems, and although NK cells expanded with IL-2 also show enhanced cytotoxic activity, preliminary studies suggest that IL-15-expanded NK cells may be more potent.⁷⁰ Similarly, in vivo therapy with IL-15 seems to induce more substantial NK expansion than IL-2

therapy.⁷¹ Thus, the central role of IL-15 as a growth and survival factor for developing and mature NK cells has led to the prediction that it will be a potent adjuvant for NK-cell-based immunotherapy. However, IL-2, IL-15, and IL-21, all have substantial effects on NK cells, and optimal use of these factors for ex vivo expansion and in vivo survival of adoptively transferred NK cells could potentially involve 1 or all these agents, and no clearly superior agent or combination has emerged thus far.^{72,73}

Clinical studies have not yet been conducted with rhIL-15, but studies in primates demonstrated that the administration of primate IL-15 at a dose of 10 µg/kg every 2 to 7 days increased responses to tetanus and influenza vaccine.⁶⁰ In simian immunodeficiency virus-infected macaques receiving antiretroviral therapy, IL-15 (10–100 µg/kg) administered twice weekly for 4 weeks increased circulating NK-cell numbers and CD8⁺ and CD4⁺ effector memory populations.⁷⁴ When rhIL-15 was administered to a macaque using a daily dose of 15 µg/kg/d, Berger et al⁷⁵ reported reversible neutropenia related to bone marrow suppression in the first animal, 20% weight loss in a second animal and a nonspecific inflammatory dermatitis in a 3rd animal treated with a reduced daily dose of 5 µg/kg. Despite these clinical toxicities, there was clear evidence for biologic activity with increased numbers and cycling rates of peripheral blood CD4⁺ T cells, CD8⁺ T cells, γδ T cells (CD4⁺CD8⁺), and NK cells, with preferential expansion of memory T cells, when compared with naive subsets. Subsequent studies in a total of 7 animals used an intermittent regimen (2.5–10 µg/kg every 3 days), which was well tolerated and also associated with substantial expansion of CD4⁺ and CD8⁺ memory T cells but lesser effects on NK cells and γδ T cells. Thus, preclinical data in primates suggest that a safe dose and regimen of IL-15 can be defined wherein the effects of IL-15 on memory T cells and NK cells could be exploited clinically.

INTERLEUKIN-21

IL-21 Biology and Preclinical Studies

IL-21 is the newest member added to the γc cytokine family, discovered in 2000.^{76,77} IL-21 is structurally similar to IL-2 and has many similar properties. The receptor for IL-21 comprises a unique subunit (IL-21R) and the common γc.^{78,79} The IL-21R complex is expressed on most mature lymphocytes, including T, B and NK cells, but IL-21 production is restricted to activated CD4⁺ T cells, especially Th17 cells^{80–82} and T follicular helper (Tfh) cells. Because IL-21 production is restricted, cytokine availability likely plays an important role in regulating the effects of IL-21. IL-21 signaling on CD4⁺ T cells, in combination with TGFβ, drives development of the Th17 subset and restricts induced Treg generation.^{80,81} The combination of IL-21 signaling on, and IL-21 production by, activated Th17 cells results in an autocrine loop, which plays an important role in autoimmune and inflammatory states such as rheumatoid arthritis, SLE, psoriasis,⁸³ and GVHD.⁸⁴ In CD8 T cells, IL-21 signaling costimulates for activation and enhances effector function. In NK cells, IL-21 induces maturation and enhances cytotoxic killing.⁸⁵ In B cells, IL-21 signaling alone induces apoptosis, but in the presence of CD40 and anti-IgM, IL-21 can promote class switching and differentiation of memory B and plasma cells. Moreover, IL-21 signaling on CD4⁺ Tfh cells, which reside within the germinal center, provides cognate help for isotype

switching and high-affinity antibody production.⁸⁶ Thus, although IL-21 induces apoptosis of B cells, it plays a major role in the generation of effective humoral immunity through indirect effects within the germinal center. In contrast to the immune activating effects described earlier, IL-21 inhibits dendritic cell differentiation and activation.^{87,88}

The capacity of IL-21 to direct differentiation of inflammatory CD4⁺ T cells while inhibiting Tregs and enhancing cytolytic function of CD8 and NK cells makes it an attractive cytokine for use in cancer immunotherapy. IL-21 treatment can inhibit growth of syngeneic tumors including Rensselaer renal carcinoma and B16 melanoma, both with⁸⁹ and without^{90,91} adoptive transfer of tumor-specific T cells. Similarly, antitumor effects have been observed in several models where murine tumors were genetically modified to secrete IL-21.^{92–95} In these models, tumors modified to secrete IL-21 make potent vaccines that induce memory responses and a capacity to respond to parental cell lines not modified with IL-21.^{95,96} IL-21 also enhances the effectiveness of antibody-dependent cell-mediated cytotoxicity, potentially by upregulation of CD247 on NK cells.^{97–100}

The costimulatory properties of IL-21 have led to its use in expansion of antitumor effectors ex vivo. Combination treatment with IL-21 and IL-15 yielded more CD8⁺ and CD56⁺ cells, fewer Tregs, and enhanced antigen-specific cytotoxicity, when compared with treatment with IL-2.^{101,102} Similarly, recent work demonstrated that IL-21 enhanced the effectiveness of human cells bearing a chimeric antigen receptor targeting CD19 in a xenograft model, compared with IL-15 and IL-2.¹⁰³ In 1 study, IL-21 outperformed both IL-2 and IL-15 as an adjuvant for adoptive transfer of tumor antigen-specific T cells.¹⁰⁴

Based on evidence that IL-21 can enhance both CD8 and NK-cell function, the results described earlier are not surprising. However, more detailed work demonstrates that IL-21 also induces a distinct pathway of CD8 differentiation that differs fundamentally from IL-2 mediated differentiation. Although IL-2 enhances the generation of CD44⁺ granzyme⁺ CD8⁺ cells that express Eomesodermin, IL-21 generates CD8⁺ T cells with repressed IL-2R α but enhanced L-selectin expression and enhanced antitumor effects.¹⁰⁵ Thus, through as yet poorly understood mechanisms, IL-21 may prevent terminal differentiation of cytolytic effectors ex vivo, and in turn enhance subsequent functionality of IL-21 cultured cells on transfer in vivo. In addition, to its distinct pathway of CD8 differentiation, another study administered IL-21 in concert with IL-2 as an adjuvant to T-cell transfer and noted simultaneous enhancement of cellular and humoral responses by IL-21 administration. Importantly, IL-21 mediated enhancement of humoral immunity contributed to the antitumor effects in this model.¹⁰⁶ Thus, in addition to the capacity of IL-21 to enhance CD8 and NK-cell function and augment Th17 cells, IL-21 mediated effects on Tfh cells, which provide help for antibody generation in the germinal center, and may also contribute to the antitumor effects of IL-21.

Clinical Trials With IL-21

Three clinical trials have been conducted with IL-21. The first phase I study was an open-labeled, 2-armed trial of recombinant human IL-21 in patients with metastatic melanoma. Twenty-nine patients were given an intravenous bolus injection at doses from 1 to 100 μ g/kg using 2 treatment regimens: thrice weekly for 6 weeks (3/wk) or 3 cycles of daily dosing for

5 days followed by 9 days of rest (5 + 9). IL-21 was generally well tolerated with a maximally tolerated dose of 30 µg/kg. Toxicities included increased alanine and aspartate aminotransferases, neutropenia, fatigue, thrombocytopenia, and lightheadedness with fevers and rigors. One partial response was observed after 3 weeks of treatment and became a complete response 3 months later.¹⁰⁷ Pharmacodynamic studies showed elevated serum levels of several cytokines, chemokines, acute-phase proteins, and cell adhesion proteins in a dose-dependent fashion.¹⁰⁸

In another phase I trial, 43 patients with metastatic melanoma or renal cell carcinoma were given an intravenous bolus injection of IL-21 at dose levels from 3 to 100 µg/kg, administered for 2 5-day cycles (days 1–5 and 15–19). The most common adverse events included flu-like symptoms, pruritis, and rash. Twelve patients received 5 additional 2-cycle courses of treatment without cumulative toxicity, except 1 patient with reversible grade 4 hepatotoxicity. Of 24 patients treated for malignant melanoma, there was 1 complete response and 11 patients with stable disease. Of 19 patients treated for renal cell carcinoma, there were 4 partial responses and 13 patients with stable disease.¹⁰⁹ In a phase II open-labeled, single-armed, 2-staged trial, IL-21 was administered at 30 µg/kg/d in 5-day cycles every 2nd week. The primary objective of this study was antitumor efficacy. Of 24 patients treated, there was 1 complete response and 1 partial response.¹¹⁰ Thus, IL-21 is well tolerated and has shown a reproducible rate of antitumor effects when administered as a single agent to patients with malignant melanoma and renal cell carcinoma. Future studies are awaited combining IL-21 with vaccines and adoptive cell therapies (summarized in Table 2).

GAMMA-C CYTOKINES AND LYMPHOID MALIGNANCIES

Given the role that γ c cytokines play in lymphoid development, incorporation of these agents into regimens used to treat lymphoid malignancies could be problematic. Indeed, mice transgenic for IL-7 develop significant lymphoproliferation and mice transgenic for IL-15 develop large granular cell leukemia. Furthermore, most immature and mature lymphocytes that comprise lymphoid leukemias and lymphomas can be expected to proliferate in response to these agents, raising a note of caution for use in this context. IL-21, however, is an exception to this rule. Because of the unique ability of IL-21 to induce apoptosis in B cells, a logical application of this cytokine in cancer immunotherapy would be against B-cell malignancies. Accordingly, in a murine model, IL-21 can induce caspase-dependant apoptosis of diffuse large B-cell lymphoma cells, which exhibit widespread expression of the IL-21 receptor.¹¹¹ In another study, IL-21 was shown to induce apoptosis in vitro in mantle cell lymphoma, a subtype of B-cell lymphoma, by a STAT1-dependent pathway,¹¹² and IL-21 has also been reported to induce apoptosis of CLL, through upregulation of the BH3 family member BIM.^{98,113} In a model using CD19⁺ lymphoma and adoptively transferred CD19-specific T-cells that constitutively express IL-2, IL-7, IL-15, or IL-21, each cytokine enhanced eradication of B-cell malignancy, but IL-7 and IL-21 were most efficacious in vivo.¹⁰³ Importantly, however, T-cell lymphomas do not apoptose after IL-21 signaling, and in anaplastic lymphoma kinase-positive anaplastic large cell lymphoma, IL-21 contributed to JAK/STAT3 activation and cell growth.¹¹⁴ Similarly, growth of Hodgkin lymphoma cell lines, although involving a B-cell lineage, can be potentiated by

IL-21.^{115,116} Thus, special consideration should be given to the effects of γ c cytokines on the tumors themselves when considering incorporation of these agents as adjuvant to immune based therapies targeting these diseases.

SUMMARY

Promising results continue to be generated with the use of immunomodulators, tumor vaccines, and adoptive cell therapy for cancer, but the overall effectiveness of immunotherapies against established tumors remains limited. Preclinical work demonstrates that the γ c cytokines, which are potent T-cell and NK-cell growth factors, can augment the potency and antitumor effects of a variety of immune based therapies. Although IL-2 is the γ c prototype, there is now ample evidence that IL-7, IL-15, and IL-21 are more effective adjuvants for vaccine-based and adoptive immunotherapy than IL-2. Their properties overlap somewhat, and each has been demonstrated to enhance long-term responses to vaccines, long after the cytokine has been discontinued. Furthermore, all have been demonstrated to induce antitumor activity when tumor cells are genetically engineered to secrete the cytokine, and all have augmented adoptive therapies when used to culture cells ex vivo or when administered in vivo in conjunction with cell transfer. Beyond this, each has unique properties that should serve to guide their incorporation into multimodal regimens in the clinic. Thus far, their toxicity profiles seem acceptable although only limited clinical studies have been conducted with rhIL-7 and rhIL-21, and rhIL-15 has not yet been tested in humans. Careful clinical trials are needed to define the dose, schedule, and setting where their biologic effects can be maximally leveraged to enhance the effectiveness of immunotherapy.

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TABLE 1.Cell-Specific Effects of γ c Cytokines

	B Cells	CD4 T Cells	CD8 T Cells	NK Cells
IL-2	↑	↑ Especially Tregs	↑↑	↑↑
IL-7		↑ Especially non-Tregs	↑↑	
IL-15		+/-	↑↑↑	↑↑↑
IL-21	↓	↑ Th17 ↑ Tfh	↑ Cytotoxicity	↑ Cytotoxicity

TABLE 2.

Clinical Trials With γ c Cytokines

	Dose	Toxicity	Biological Activity	Antitumor Activity
IL-2	Subcutaneous: 700,000 IU–5,000,000 IU/m ² (43 μ g–306 μ g/m ²) $\approx$ (1–7.7 μ g/kg)	Capillary leak, hepatic dysfunction, flu-like malaise, low blood pressure, fever, nausea, vomiting, diarrhea, chills, edema, weight gain, confusion, skin rashes, and changes in blood chemistry.	Early lymphopenia due to trafficking followed by expansion of peripheral lymphoid cells with continued administration. Treg proliferation. Increased serum levels of gamma interferon.	Renal cell carcinoma, melanoma, and possible enhanced activity of adoptively transferred cells.
IL-7	Intravenous: 360,000–720,000 IU/kg (22–44 μ g/kg) 3–60 μ g/kg	Fever, myalgias, $\uparrow$ liver function tests, injection site erythema.	Lymphocytosis Increased T-cell cycling and broadening of circulating TCR repertoire diversity. Increased size of spleen and lymph nodes. Bcl-2 upregulation.	None
IL-15	Recently approved for phase I trials, data not yet available			
IL-21	3–100 μ g/kg	Fever, myalgias, $\uparrow$ liver function tests, neutropenia, and thrombocytopenia.	Increased soluble CD25 Increased frequencies of NK and CD8 ⁺ T cells Increased cytotoxicity of NK cells and CD8 ⁺ T cells	Renal cell carcinoma, melanoma