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Cytokines in Cancer Therapy

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Contents

Interleukin-2 in Cancer Treatment

Gene Transfer of Cytokines

Biological Function of Cytokines and Adhesion Molecules

Miscellaneous

Immunotherapy in Cancer: Concepts and Prospects

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Interferon-α in the Treatment of B-Cell Chronic Lymphocytic Leukemia

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Introduction

The majority of previously untreated patients with early phase B-cell chronic lymphocytic leukemia (B-CLL) expects a long-lasting indolent course of the disease. Long-time survival of these patients does not differ from that of an age- and sex-matched control population [1]. However, approximately 30% of the patients will have progressive disease within 2 years after establishment of diagnosis. Diffuse bone marrow involvement [2], lymphocyte doubling time < 12 months [3], and serum levels of thymidine kinase > 5 U/l [4] have been identified as risk factors for progression. Treatment with chlorambucil during the early phase of the disease revealed no benefit with respect to survival, but led to an increased risk for secondary malignancies [1]. Therefore, early stage B-CLL usually is not treated until progression has occurred.

In pilot studies, interferon-α (IFN-α) was shown to have antileukemic activity in early stage B-CLL [5–8] whereas only marginal or no beneficial effects were reported in advanced stage disease [9, 10]. Here, we report on our own experiences obtained from a pilot study with IFN-α in early stage B-CLL. Furthermore, we present first results of a still ongoing randomized

multicenter trial. Encouraged by the results of our pilot study, this trial was activated to examine the possible benefit of an IFN-α treatment in early stage disease at high risk for progression. We will also discuss future concepts dealing with IFN-α in the treatment of B-CLL.

IFN-α in a Pilot Study on B-CLL

Patient Characteristics

Prompted by the obvious efficacy of IFN-α in hairy cell leukemia [11], a pilot study in B-CLL was initiated [7, 12]. Nine patients with previously untreated early stage B-CLL (4 females, 5 males) were included in this phase II trial. Median age was 48 years (range 42–58 years). They all had Binet stage A disease [13], according to Rai stages varying from 0 to II [14]. Median time from first diagnosis was 16 months (range 2–65 months). Four patients had a lymphocyte doubling time (LDT) < 12 months. A diffuse bone marrow involvement was found in another 2 patients. Overall, the tumor load was considered to be low, since the total circulating lymphocyte count in each patient did not exceed 50,000 l/μl at the time of recruitment for the study.

Treatment

Treatment consisted of 5×10^6 IU IFN-α subcutaneously (Intron A®, supplied by Essex-Pharma, Munich, FRG) 3 times weekly. Duration of treatment has been in the range of 15–36 months. Treatment was overall well tolerated. In 6 patients toxic side effects included flu-like symptoms, which did not exceed WHO grade II. While in 2 patients depression was observed, another 2 patients did not experience any toxic side effects.

Results

All patients responded to IFN-α with a decrease of their lymphocyte count. However, the lymphocyte count increased again despite continued IFN-α treatment in 1 patient 3 weeks after initiation of therapy. Splenomegaly resolved in 1 patient, and this particular patient turned from Rai stage II to stage I disease. A rise of immunoglobulin levels was observed in 3 patients, and in 4 patients HLA-DR expression on monocytes was doubled. Four patients achieved a partial remission according to the remission criteria proposed by Cheson et al. [15], whereas another 4 patients had stable disease.

Conclusions

This pilot study showed the antileukemic efficacy of IFN-α in early stage B-CLL. The high rate of responding patients (8 out of 9) was in

Table 1. Pilot studies on IFN-α in B-CLL

Author	Patients	CR	PR	Ref.
Foon et al.	18	0	2	9
O'Connell et al.	4	0	1	16
Pangalis and Griva	10	1	4	6
Talpaz et al.	10	0	3	10
Rozman et al.	10	0	8	8
Ziegler-Heitbrock et al.	9	0	8	7

accordance with some previous reports [6, 8], but in striking contrast to other findings [9]. However, that latter report discussed data obtained in a trial on patients with advanced stage and in some cases pretreated but refractory B-CLL. One may therefore speculate that a low tumor load and lack of prior cytotoxic therapy as in our patients could be a prerequisite condition for the effectiveness of IFN-α. (A summary of published remission rates in B-CLL treated with IFN-α is listed in table 1.)

Of major interest was the finding that patients with a LDT < 12 months and with a diffuse bone marrow involvement also responded to treatment. Beside high levels of serum thymidine kinase [4], a short LDT [2] as well as a diffuse bone marrow involvement [3] were identified as risk factors for progression in early stage B-CLL.

Thus, in consequence of the results of this pilot study, a phase III trial on IFN-α in early stage B-CLL at high risk for progression was initiated.

IFN-α in Early Stage B-CLL – Preliminary Results of a Phase III Trial

Materials and Methods

Inclusion Criteria and Study Design

Patients who were not older than 75 years and having a morphologically and immunologically proven, previously untreated B-CLL in Binet stage A were included in this trial. All the patients had to give their written informed consent. The lymphocyte count had to be < 100,000 l/μl. Patients were stratified by risk factors. A diffuse bone marrow infiltration and a LDT < 12 months and /or serum levels of thymidine kinase > 5 U/l qualified for the high-risk group. Patients in the high-risk group were randomized into an arm A with IFN-α

therapy, and an arm B without treatment. Patients with a nodular bone marrow infiltration, and all those with a diffuse infiltration but without an additional risk factor, were allocated to the low-risk group (arm C, no treatment). The aims of the study are freedom from progression and long-time survival.

Patient Characteristics

At the time of this analysis (April 15, 1992), 13 patients were evaluable in arm A, 13 patients in arm B, and another 28 patients in arm C. Median age of the three study arms was 60 years. No severe concomitant diseases were evident. Median serum levels of the thymidine kinase were 7 U/l in arm A and 8.3 U/l in arm B, but 3.8 U/l in arm C ($p < 0.01$). Median LDT was 10 months in arm A, 24.5 months in arm B (no significant difference), and 42 months in arm C ($p < 0.05$).

Treatment

High-risk patients randomized into arm A were treated with IFN-α (Intron A®) subcutaneously. The planned dosage per week was $3 \times 5 \times 10^6$ IU IFN-α. Dose modifications for individual reasons like toxic side effects were allowed. Dose escalation up to $3 \times 8 \times 10^6$ IU IFN-α was proposed in case of increasing lymphocyte counts during IFN treatment.

Definitions

Complete remission (CR) was defined as the complete disappearance of peripheral signs of CLL, i.e. lymphocytosis, lymphadenopathy, and hepatosplenomegaly. Partial remission (PR) was defined as the 50% or more reduction of the tumor load. Beside an upstaging to Binet B or Binet C disease, progression of disease was defined as the increase of the total lymphocyte count >100,000 l/μl, or occurrence of new enlarged lymph nodes, or increase of prior enlarged lymph nodes, liver, or spleen.

Results

Status of Disease

Five out of the 13 high-risk patients with IFN-α treatment (arm A) achieved a PR with time to reach remission varying from 1 to 5 months. Two patients had stable disease. The remaining 6 patients had progressive disease, and 2 of them had to be treated with conventional chemotherapy. It has to be noted, however, that for individual reasons only 45% of the patients in arm A received the 100% dosage of IFN-α according to the study protocol, i.e. $3 \times 5 \times 10^6$ IU/week. In arm B (high risk, no treatment) 7 out of 13 patients had stable disease, whereas the disease was progressive in the remaining 6 patients. Four of the latter ones had to be treated with conventional chemotherapy. In contrast to the high-risk patients, the vast majority (23 out of 28 patients) of the low-risk patients had stable disease. Even though 5 patients experienced disease progression, none of

Table 2. Results

Arm A: n = 13	treatment with IFN-α, 'high-risk'[1]
	PR 5
	NC 2
	PD 6, with 2 in need for chemotherapy
Arm B: n = 13	no treatment, 'high risk'
	PR 0
	NC 7
	PD 6, with 4 in need for chemotherapy
Arm C: n = 28	no treatment, 'low risk'
	PR 0
	NC 23
	PD 5, with 0 in need for chemotherapy

PR = partial remission; NC = no change; PD = progressive disease.

[1] Only 45% of the patients were treated with the 100% dosage of IFN-α ($3 \times 5 \times 10^6$ IU sc) as outlined in the protocol.

them was in need for chemotherapy. Time until progression differed significantly ($p < 0.05$) between the high-risk and the low-risk patients. No influence of IFN-α became evident so far, while median time to progression was 12 months in arm A, and 13 months in arm B. The median was not yet reached in arm C within an observation time of 30 months. The results are listed in table 2.

Toxic Side Effects

IFN-α was well tolerated in the majority of patients. Flu-like symptoms, alopecia, and weight loss did not exceed WHO grade II toxicity. A slight increase of serum levels of GOT and creatinine, respectively, was observed only in 1 patient (WHO grade I toxicity). Nevertheless, 3 patients were excluded from further IFN-α treatment due to toxic side effects. Two of them experienced depression (WHO grade II). One patient suffered from diarrhea and nausea to be classified as WHO grade III toxicity. This particular patient had to definitely discontinue IFN-α therapy despite a dosage reduction from $3 \times 3 \times 10^6$ FU IFN-α/week to $3 \times 1 \times 10^6$ IU/week, and despite concomitant treatment with antiemetic drugs. The toxic side effects are listed in table 3.

Table 3. Toxic side effects

Side effects	WHO grade			
	I	II	III	IV
Fever	2	1		
Myalgia	5			
Headache	1			
Alopecia	4			
Weight loss	2			
Diarrhea		1	1	
Nausea	2		1	
Dizziness	1	2		
Depression	1	2		
SGOT	1			
Creatinine	1			

Only 1 patient with WHO > II toxicities; 3 patients off treatment due to toxicities.

Discussion

The preliminary results of this randomized trial on IFN-α in early stage B-CLL seem to confirm the concept of stratification by risk factors. Five out of 28 patients at low risk for progression had progressive disease, whereas 12 out of 26 patients at high risk (13 patients in arm A, another 13 patients in arm B) had progressive disease. Moreover, a significant difference in time until progression became evident. In the two high-risk groups, median time to progression was 12 and 13 months, respectively. In the low-risk group, however, the median was not yet reached within an observation period of 30 months. Therefore, the occurrence of a diffuse bone marrow infiltration together with a LDT < 12 months and/or levels of serum thymidine kinase > 5 U/l define in fact a condition of high risk for progression in early stage B-CLL.

Antileukemic effects of IFN-α were evident in some patients. Five out of 13 patients in our study achieved a PR. Foon et al. [9] reported on 2 patients out of 12 achieving a PR, and O'Connell et al. [16] found 1 PR in 4 patients. Somewhat better results were obtained by Pangalis et al. [6] who reported on 5 out of 10 patients achieving a remission (1 CR and 4 PR). In that particular study, the 4 patients who attained a PR had Binet stage A dis-

ease and had not recieved prior chemotherapy. A high rate of responding patients was reported by Rozman et al. [8]. All the 10 patients examined in their study had a transient reduction of the number of circulating lymphocytes. Moreover, an increase of the granulocyte count was detected in 8 patients. Interestingly, all the patients in the study of Rozman et al. had Binet stage A disease without prior treatment, according to Rai stages O–I.

While some efficacy of IFN-α in early stage B-CLL is obvious, little is known until today about the mechanisms of action. Efficacy of IFN-α may depend on the number of receptors on the target cells. While low numbers of receptors were found in advanced stage B-CLL, high numbers detected in early stage could be prerequisite for the antileukemic effects of IFN-α [17]. Moreover, induction of HLA-DR expression with activation of monocytes [7], recruitment of NK cells [16], and a relative increase of the T_4-positive lymphocyte subset [16] during IFN-α treatment were reported. These latter findings may also be related to the responsiveness of early stage B-CLL to IFN-α.

IFN-α may interact with other cytokines. Tumor necrosis factor-α (TNF-α) was found to act as an autocrine growth factor in B-CLL [18, 19], and high serum levels of TNF-α are thought to be a characteristic feature in early stage disease [20]. The disruption of the TNF-α dependent proliferation in B-cell malignancies by IFN-α was postulated to represent the mechanism by which IFN-α could counteract cell proliferation [21]. However, IFN-α does not seem to have an influence on the production of TNF-α in B-CLL cells, and furthermore, it enhances TNF-α release in hairy cell leukemia [22]. Thus, efficacy of IFN-α seems to be mediated by other pathways.

The prognostic impact of the response to IFN-α treatment in early stage B-CLL remains undefined, since no studies on the long-time follow-up of the responding patients were published so far. A larger number of patients and a follow-up for several years in our study may contribute to elucidate that point.

Future Concepts

So far, efficacy of IFN-α in B-CLL was shown in a number of pilot studies (see table 1). A consistent observation was that the effectiveness of IFN-α was higher in early stage than in advanced stage disease. Induction of remission in case of a low tumor load is therefore possible, but the ben-

efit with respect to survival and freedom from progression remains to be defined.

Maintenance of remission is another goal in the treatment of B-CLL. IFN-α was shown to prolong the duration of remission in multiple myeloma after prior 'inductive' chemotherapy [23]. In analogy, IFN-α could provide the chance of maintained remission in early as well as in advanced stages of B-CLL after conventional chemotherapy. Montserrat et al. [24] published first results of a pilot study, in which the tumor load of 11 patients with early stage B-CLL was reduced by intermittent chlorambucil. Then, IFN-α was administered, and the quality of remission was improved in 5 patients. Additional studies on IFN-α after prior chemotherapy in advanced stage B-CLL are urgently warranted.

IFN-α is currently examined in combination with 5-fluorouracil (5-FU) in a number of trials on solid tumors of the gastrointestinal tract. The rationale of these trials is to modify the cytotoxicity of 5-FU by IFN-α [25]. Modulation of cytotoxicity could be an approach also in B-cell malignancies. Pini and Foa [26] reported on 2 patients with B-CLL having received chlorambucil in combination with IFN-α. The progressive disease in both patients could not be further controlled by chlorambucil alone. But, the additional application of IFN-α led to a reduction of circulating lymphocytes followed by a stable disease. This preliminary observation should initiate pilot studies to assess the feasibility and efficacy of combined treatments with IFN-α and cytotoxic agents.

A number of cytokines seem to be involved in the regulation of cell growth in B-CLL. Interleukin-4 (IL-4) shows antiproliferative activity [27], while, for example, TNF-α and interleukin-2 (IL-2) are known to stimulate proliferation [18, 19, 28]. Therefore, studies on the combination of IFN-α with IL-4 are needed and might be helpful for understanding the cytokine network in B-CLL.

In summary, IFN-α provides some antileukemic activity in B-CLL. However, its modes of action and most effective modalities of treatment still remain to be clarified.

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