Phase II Study of All-trans-retinoic Acid and α-Interferon in Patients with Advanced Non-Small Cell Lung Cancer

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ABSTRACT

Between April 1993 and June 1994, 29 patients (pts) with unresectable, locally advanced, or metastatic non-small cell lung cancer were treated with a combination of p.o. trans-retinoic acid (TRA), 150 mg/m²/day, in three divided doses and s.c. IFN-α, 3 x 10⁶ units/day. The age range was 41-80 years (median, 63 years). The Eastern Cooperative Oncology Group performance status was 0-1 (24 pts) and 2 (5 pts). Pts had advanced disease, refractory to conventional therapy (5 stage IIIB and 24 stage IV). Twenty-one pts had adenocarcinoma, six had squamous cell carcinoma, and two had large cell carcinoma. Only 3 pts completed 8 weeks of treatment, requiring neither interruption nor dose modification. Fatigue occurred in 88% of pts. A syndrome complex consisting of dry oral and nasal mucosa, recurrent sinus infections, and epistaxis occurred in 64% of pts. Grade II/III dermatitis was seen in 52%. Severe scrotal dermatitis was seen in 7 pts (47% of 15 males). Hypertriglyceridemia was moderate/severe in 11 pts, and 3 pts required gemfibrozil for levels up to 1660 mg/dl. Hematological toxicity was not encountered, and none of the pts had leukocytosis. One pt died with complications of myocardial infarction while on TRA/IFN-α. Twenty-five pts had more than 2 weeks of treatment and are evaluable for response; two pts died early with complications of cancer, and two pts declined to continue after only 3 and 5 days of treatment. Final assessment of response was by accepted clinical and radiological criteria at 8 weeks. There have been four objective responses: complete response, 2 (18+ and 17 months) and partial response, 2 (7 and 14 months). Responses were observed in all histologies. Combined differentiation treatment with TRA/IFN-α has modest but objective activity in non-small cell lung cancer. Toxicity is considerable. Additional studies to elucidate the biological basis of TRA/IFN-α and to define prognostic parameters predicting response are needed.

INTRODUCTION

Wolbach and Howe (1) performed a series of pioneering animal studies in the 1920s showing the significance of retinoids in control of both differentiation and proliferation of epithelial cells. In their experiments, rats deprived of vitamin A showed signs of abnormal cellular differentiation associated with excessive proliferation of dysplastic epithelia. Subsequently, it was shown that retinoids can suppress the process of carcinogenesis in experimental animals (2, 3). More recently, retinoids were shown to exert effects on invasive neoplastic cells, leading to suppression of proliferation and terminal differentiation, morphologically reflected in a more benign non-neoplastic phenotype (4-7).

Clinically, retinoids have been successfully applied in cancer therapy and chemoprevention (8). TRA2 induced complete remission in 88% of the patients with acute promyelocytic leukemia, a treatment effect that may occur on the molecular level through interaction with the RAR-α receptor gene, which is rearranged in all patients (9-11). CRA is an active therapy for oral leukoplasia (12), and has prevented the development of second primary epithelial malignancies in patients with resected primary cancers of the head and neck (13).

Retinoids and IFN-α act synergistically when applied to cancer cell lines (14-16). They seem to share independent anti-proliferative, differentiative, immunomodulatory, and antian- giogenic activities as well as different toxicity patterns, making a combination very attractive for clinical use (17, 18). The combination of CRA and IFN-α showed significant antitumoral activity in patients with SCC of the skin (19) and uterine cervix (20, 21), stimulating the design of Phase II trials in patients with other incurable epithelial malignancies.

NSCLC is a major public health problem, and 170,000 new cases and 158,000 lung cancer deaths are expected in the United States in 1995 (22). Although chemotherapy and radiation therapy have activity in NSCLC, surgery remains the only curative therapy. Unfortunately, only a small percentage of patients are expected to be cured. There is no systemic therapy currently available that can be administered with a curative intent for patients with recurrent or metastatic NSCLC, and chemotherapy remains a controversial tool for palliation (23). Development of innovative systemic strategies is urgently needed for this group of patients.

MATERIALS AND METHODS

Under the aegis of the National Cancer Institute, a single-institution, Phase II study of combined TRA and IFN-α in...

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NSCLC was initiated at Northwestern University in April 1993. Patients with recurrent, metastatic, or locally advanced inoperable NSCLC were enrolled. Histologies allowed included: adenocarcinoma, SCC, and large cell carcinoma. Histology was confirmed with biopsy or cytology. Sites of disease were measurable bidimensionally on a computerized tomographic scan or chest X-ray. Patients with brain metastases were excluded, and a normal brain computerized tomography was a requirement. The Eastern Cooperative Oncology Group performance status of 0, 1, or 2 was allowed, and patients required normal blood counts, hepatic and renal function, and baseline triglycerides less than 400 mg/dl. Patients were allowed up to two previous chemotherapy regimens and could have received previous radiation therapy as long as treatment was completed at least 6 weeks before initiation of TRA/IFN-α. Previously irradiated fields were acceptable as measurable disease, if tumor progression was documented. A signed informed consent was required.

Patients were treated with p.o. TRA (150 mg/m² (2/day)) in three divided doses and s.c. IFN-α (3 x 10⁶ units/day). Specific guidelines were developed for interruption of therapy and dose modifications in patients who experienced significant toxicity. The Cancer Treatment Evaluation Program of the National Cancer Institute provided both medications.

All patients were required to undergo radiographic imaging of measurable and evaluable disease. Complete restaging of the measurable and evaluable disease was planned after 8 weeks of therapy or at termination of therapy. Patients who completed 2 weeks of therapy were evaluable for response, and were characterized as having CR, PR, stable disease, or progressive disease using standard, clinical, and radiological criteria. Responding or stable patients continued therapy indefinitely, depending on the degree of associated toxicity. Patients with progressive disease were taken off study. All treated patients were followed to the time of death, and overall survival and time to progression were considered secondary end points. Survival was analyzed using Kaplan-Meier product line curves and the log rank test (24). Toxicity was evaluated with the Eastern Cooperative Oncology Group toxicity criteria (25).

RESULTS

Between April 1993 and June 1994, 29 patients were enrolled in the study. Patient characteristics are described in Table 1. All patients had advanced disease, incurable by conventional therapy.

Response was evaluable in 25 of 29 patients. Two patients died with complications of cancer within 2 weeks of therapy before they experienced any toxicity, and two patients discontinued therapy after 3 and 5 days. The median duration of treatment was 7 (range, 2–62) weeks. Interruption of therapy was common. Only 3 of 25 patients completed 8 weeks of uninterrupted therapy, requiring no dose modification.

Objective responses were seen in 4 (16%) of 25 patients: 2 CR (18+ and 17 months) and 2 PR (7 and 14 months). Responses were observed in all histologies. Four patients had stable disease. All responding patients showed signs of response within the first 8 weeks of therapy. Two patients with initial PR had another response more than 2 months after initiation of therapy, and one achieved and maintains a CR. An analysis of responding patients is shown in Table 2. Fig. 1 illustrates complete resolution of a lung mass in a responding patient (M. E.).

Toxicity was evaluable in 25 of 29 patients (Table 3). The four unevaluable patients had received minimal treatment without associated toxicity. In all other patients toxicity was significant but reversible shortly after therapy was discontinued. Fatigue was common and occurred in 88% (22/25) of patients. A syndrome complex consisting of dry oral and nasal mucosa, recurrent sinus infections, otitis, epistaxis, and conjunctivitis was encountered in 64% (16/25) of patients. Grade I/II dermatitis was seen in 76% (19/25); 7 of 15 male patients (46%) experienced severe scrotal dermatitis. Cheilitis was seen in 56% (14/25) of the patients, and almost all of the patients on study required local moistening agents. Chills and fever were reported in 24% (6/25); grade I/II headache in 36% (9/25); less common toxicities were diarrhea (3/25), alopecia (3/25), depression (3/25), and numbness (1/25). Hematological toxicity was minimal, and none of the patients developed leukocytosis or retinoic acid syndrome (e.g., dyspnea, leukocytosis, and pulmonary infarction). Hypertriglyceridemia was seen in 11 of 25 patients (44%), of whom required gemfibrozil for serum triglyceride levels up
Table 2  Analysis of responding patients

<table>
<thead>
<tr>
<th>Patient</th>
<th>Age/sex</th>
<th>Histology</th>
<th>Stage</th>
<th>Previous therapy</th>
<th>Responding site</th>
<th>Pre-, Posttreatment measurements</th>
<th>Response</th>
<th>Duration (mo)</th>
</tr>
</thead>
<tbody>
<tr>
<td>M. E.</td>
<td>57/F</td>
<td>SCC</td>
<td>IV</td>
<td>None</td>
<td>Lung mass</td>
<td>3.0 × 5.0 cm → 0</td>
<td>CR</td>
<td>17</td>
</tr>
<tr>
<td>J. D.</td>
<td>63/M</td>
<td>Adenocarcinoma</td>
<td>IV</td>
<td>Surgery, RT†</td>
<td>Flank mass</td>
<td>2.0 × 2.9 cm → 0</td>
<td>CR</td>
<td>18+</td>
</tr>
<tr>
<td>M. M.</td>
<td>58/F</td>
<td>Large cell</td>
<td>IV</td>
<td>Surgery, RT</td>
<td>Recurrent lung mass</td>
<td>1.5 × 2.0 cm → 0</td>
<td>PR</td>
<td>14</td>
</tr>
<tr>
<td>R. O.</td>
<td>52/M</td>
<td>Adenocarcinoma</td>
<td>IV</td>
<td>CDDP/CTX/Mito Taxol</td>
<td>Mediastinal l.n.</td>
<td>2.5 × 2.0 cm → 1.0</td>
<td>PR</td>
<td>7</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Pleural effusion</td>
<td>1.5 × 2.0 cm → 1.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Osteoblastic bone lesions</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* RT, radiation therapy; CDDP, cisplatin; CTX, cyclophosphamide; l.n., lymph node; Mito, mitomycin C.

Fig. 1  a, patient M. E., computerized tomographic scan demonstrating a left chest wall mass, biopsy-proven to be SCC. b, patient M. E., computerized tomographic scan at 4 months demonstrating complete resolution of the chest wall mass following TRA/IFN-α therapy.

Median follow-up time was 7 (range, 1–23) months. Median survival time was 8 months, as reflected in the survival curve for the entire group (Fig. 2). None of the four responders died, whereas 20 of the 25 nonresponders and patients inevaluable for response died with a median time to death of 7 months. There was a significant difference in survival between responders and nonresponders ($P = 0.008$) as shown in Fig. 3.
Table 3 Toxicity

<table>
<thead>
<tr>
<th>Grade*</th>
<th>I</th>
<th>II</th>
<th>III</th>
</tr>
</thead>
<tbody>
<tr>
<td>Systemic toxicity</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fatigue</td>
<td>3</td>
<td>15</td>
<td>4</td>
</tr>
<tr>
<td>Myalgia/arthralgia</td>
<td>2</td>
<td>7</td>
<td>0</td>
</tr>
<tr>
<td>Chills</td>
<td>5</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Mucocutaneous toxicity</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mucositis</td>
<td>1</td>
<td>15</td>
<td>0</td>
</tr>
<tr>
<td>Dermatitis</td>
<td>6</td>
<td>13</td>
<td>0</td>
</tr>
<tr>
<td>Chelitis</td>
<td>3</td>
<td>11</td>
<td>0</td>
</tr>
<tr>
<td>Alopecia</td>
<td>2</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Neurotoxicity</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Headache</td>
<td>5</td>
<td>4</td>
<td>0</td>
</tr>
<tr>
<td>Depression</td>
<td>3</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Numbness</td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Hematological toxicity</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Anemia</td>
<td>4</td>
<td>4</td>
<td>0</td>
</tr>
<tr>
<td>Leukopenia</td>
<td>0</td>
<td>0</td>
<td>0</td>
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<tr>
<td>Thrombocytopenia</td>
<td>0</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Metabolic/gastrointestinal toxicity</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Hypertriglyceridemia</td>
<td>0</td>
<td>8</td>
<td>3</td>
</tr>
<tr>
<td>Diarrhea</td>
<td>2</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Abnormal liver function studies</td>
<td>5</td>
<td>5</td>
<td>2</td>
</tr>
</tbody>
</table>

*No grade IV toxicities were observed.

DISCUSSION

In this study, objective responses to TRA/IFN-α were observed in 4 of 29 treated patients with advanced NSCLC; the associated toxicity was significant, but reversible. Since NSCLC represents the leading cause of cancer deaths in the industrialized world, the development of an effective systemic therapy is of obvious importance.

Retinoids and IFN-α have had minimal activity when applied to patients with NSCLC as a single-agent therapy (26-28). The efficacy of retinoids in reversing sputum atypia and in decreasing squamous metaplasia created high expectations, but in Phase II lung cancer studies of CRA (26, 27) and IFN-α (28) have demonstrated only infrequent responses.

The rationale for the combination of retinoids and IFN-α was demonstrated in the laboratory. Synergism was initially shown in leukemic cell lines (6, 7, 29) and later in teratocarcinoma (5), and in suprapharmacological doses in SCC cell lines (15, 16). Molecular mechanisms underlying the synergism of retinoids and IFN-α are not clear. Some investigators have emphasized the antiproliferative properties (4), while others consider the induction of terminal differentiation (5-7) as the basis of biological activity. It seems that the combination induces the affected cells to terminal differentiation, which results in diminished proliferative potential. The inhibition of angiogenesis is supported as a primary mechanism of activity by some investigators (30).

Renewed clinical interest in retinoids led to the development of a multitude of new retinoid molecules with differing metabolism and biological half-life, varying affinity to retinoid receptors, and a varying spectrum of clinical activity (31). In preclinical studies, TRA proved to be the most active inducer of differentiation (32). IFN-α, already known to possess clinical activity in hairy cell leukemia, chronic myelogenous leukemia, and lymphomas (33), proved to be an important enhancer of the differentiative effects of retinoids, and emerged as the obvious cytokine to combine with retinoids for clinical studies (32). Other cytokines (IFN-γ, granulocyte-colony-stimulating factor, interleukin 1-α, and interleukin 4) were found to be synergistic with retinoids in the laboratory (29), and will be studied in future clinical trials.

The combination of CRA-IFN-α was first evaluated in the treatment of cutaneous T-cell lymphoma, producing objective responses in four of seven patients (34). The first positive results in epithelial malignancies were observed in a Phase II study of CRA-IFN-α in advanced SCC of the skin (19) with a response rate of 68% (19/28), including 7 CRs. The responses were impressive in locoregional disease (17/20 responses), but less so (2/8 responses) with distant metastases. In SCC of the uterine cervix, CRA-IFN-α was reported to have a 50% response rate, but most patients relapsed shortly after therapy was discontinued (20, 21). Studies in recurrent head and neck cancer (35) and disseminated melanoma (36) were disappointing, showing only rare PRs.

Finally, CRA-IFN-α has previously been studied in advanced NSCLC in two Phase II studies. The experience from the M. D. Anderson Cancer Center in SCC was disappointing, with only one PR observed among 24 patients (37). In a study con-
duced by the Canadian National Cancer Institute, CRA-IFN-α was administered to 34 patients with NSCLC histologies, and only one PR in a patient with SCC was observed (38). Our study is encouraging, although the basis for enhanced activity of the TR/A-IFN-α combination therapy is uncertain.

TRA bioavailability is variable (39). Serum levels may decrease after the initiation of therapy, possibly through induction of the cytochrome P450 system (40, 41). Our study was not designed to monitor serum retinoid levels, but patients appeared to have a predictable toxicity pattern associated with retinoids, even after more than 2 months of therapy. Use of cytochrome P450 inhibitors such as p.o. ketokonazole and H2 blockers administered concurrently with retinoids may be considered for future trials (40, 41).

The optimum dose and duration of TRA-IFN-α therapy is not defined. Most patients experienced significant toxicity and required dose reductions, suggesting that future studies should be initiated with lower doses of TRA and IFN-α. Responding patients had signs of clinical and radiological response during the first 8 weeks of therapy, but further response evolved over time, even in a patient who had discontinued therapy. Based on our experience, patients who have not responded by 8 weeks of therapy are unlikely to benefit by continued TRA-IFN-α therapy. In addition, responding patients may not benefit from prolonged therapy.

The identification and cloning of nuclear RARs (RAR-X) has prompted theories about the potential mechanisms of retinoic acid and IFN-α interactions. TRA is a high-affinity ligand for the RARs, while 9-cis-retinoic acid, a recently identified retinoid, is specifically active on RAR-Xs (42, 43). Retinoic acid response elements are actually located in the promoter area of retinoic acid-responsive genes, which are regulated by retinoids. IFN activity is also affected through nuclear receptors, and IFN-stimulated genes and transcription factors have been described (18).

RAR-β is coded in the short arm of chromosome 3 in an area of frequent chromosome deletions in lung cancer (44). Gebert et al. (45) examined RAR-β mRNA in specimens of normal lung, 33 lung cancer cell lines, and 9 primary lung tumors. Although normal lung expresses RAR-β, 50% of the cell lines and 30% of the tumor samples showed altered RAR-β expression and/or inducibility, including examples of the absence or a specific loss of one of the RAR-β transcripts (45). Furthermore, Houle et al. (46, 47) showed that transfection of RAR-β to epidermoid lung cancer cell lines not expressing RAR-β resulted in less tumorigenicity, reduced growth rate, and increased latency. These experiments clearly implicate the RAR-β gene as a potential tumor suppressor gene.

The underlying mechanism of interaction between TRA and IFN-α remains unclear. Exposure of cancer cells to the two agents separately decreases the expression of oncogenes (i.e., N-myc; Refs. 48 and 49). TRA also induces the expression of IFN-stimulated genes and transcription factors in cancer cell lines (50). These observations could partially explain the synergistic antiproliferative activity of the combination.

Another common link between TRA and IFN-α is the 2′-5′-oligoadenylate synthetase enzyme system, which has been implicated in cell growth control (51, 52) and differentiation (53). Studies in human neuroblastoma cells (54) and human histiocytic lymphoma (55) cell lines showed that TRA enhanced the induction of 2′-5′-oligoadenylate synthetase by IFN-α, mostly through a posttranscriptional amplification effect. Finally, retinoids and IFN-α showed synergistic antiangiogenic activity in human papilloma virus-harboring tumor cell lines (30). These laboratory observations could partially explain the clinical antitumor activity of TRA-IFN-α.

Future studies of TRA-IFN-α under consideration in NSCLC include a combination of TRA-IFN-α with chemotherapy or concurrent administration of TRA-IFN-α with radiation therapy, since both agents have a known radiosensitizing effect (56, 57).

Our clinical experience with TRA-IFN-α has shown occasional but striking responses. Toxicity was significant, but reversible, consisting mostly of mucocutaneous side effects and fatigue. Improved understanding of underlying mechanisms and definition of prognostic parameters is needed.

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REFERENCES

Retinoids/IFN in Non-Small Cell Lung Cancer


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