Int. J. Radiation Oncology Biol. Phys., Vol. 63, No. 5, pp. 1448–1454, 2005

0360-3016/05/$–see front matter

ELSEVIER

doi:10.1016/j.ijrobp.2005.05.032

CLINICAL INVESTIGATION **Lung**

CYTOKINE PROFILING FOR PREDICTION OF SYMPTOMATIC RADIATION-INDUCED LUNG INJURY

JUSTIN P. HART, PH.D.,* GLORIA BROADWATER, M.S.,‖ ZAHID RABBANI, M.D.,† BENJAMIN J. MOELLER, B.S.,† ROBERT CLOUGH, B.A.,† DALE HUANG, B.S.,† GREGORY A. SEMPOWSKI, PH.D.,‡¶ MARK DEWHIRST, D.V.M., PH.D.,*†§ SALVATORE V. PIZZO, M.D., PH.D.,* ZELJKO VUJASKOVIC, M.D., PH.D.,† AND MITCHELL S. ANSCHER, M.D.†

Departments of *Pathology, †Radiation Oncology, ‡Medicine, and §Biomedical Engineering, ‖Cancer Center Biostatistics, and ¶Duke Human Vaccine Institute, Duke University Medical Center, Durham, NC

Purpose: To analyze plasma cytokine profiles before the initiation of radiation therapy to define a cytokine phenotype that correlates with risk of developing symptomatic radiation-induced lung injury (SRILI).
Methods and Materials: Symptomatic radiation-induced lung injury was evaluated in 55 patients (22 with SRILI and 33 without SRILI), according to modified National Cancer Institute common toxicity criteria. These plasma samples were analyzed by the multiplex suspension bead array system (Bio-Rad Laboratories; Hercules, CA), which included the following cytokines: interleukin (IL)-1β, IL-2, IL-4, IL-5, IL-6, IL-7, IL-8, IL-10, IL-12p70, IL-13, IL-17, granulocyte/macrophage colony-stimulating factor, interferon-γ, monocyte chemotactic protein 1, macrophage inflammatory protein 1β, tumor necrosis factor α, and granulocyte colony-stimulating factor.
Results: Significant differences in the median values of IL-8 were observed between patients with and without SRILI. Patients who did not develop SRILI had approximately fourfold elevated levels of IL-8 as compared with patients who did subsequently develop SRILI. Significant correlations were not found for any other cytokine in this study, including transforming growth factor β1.
Conclusions: Patients with lower levels of plasma IL-8 before radiation therapy might be at increased risk for developing SRILI. Further studies are necessary to determine whether IL-8 levels are predictive of SRILI in a prospective trial and whether this marker might be used to determine patient eligibility for dose escalation.

Interleukin-8, Cytokines, Symptomatic radiation-induced lung injury, Transforming growth factor β1.

INTRODUCTION

Radiation therapy is an important therapeutic modality in the treatment of lung cancer. Although tumor control might require high doses of radiation, treatment is often limited by the radiation tolerance of normal tissues, Indeed, traditional radiation doses have been considerably lower than the estimated dose of 80–100 Gy needed to eradicate large non–small-cell lung cancers (1). In support of these estimates, improved local control and survival have been observed after dose escalation for patients with non–small-cell lung cancer and improved survival in patients with small-cell lung cancer (2–4).

Traditional normal tissue dose guidelines have been estimated on the basis of the sensitivity of large populations and might not accurately reflect any one individual's risk of developing symptomatic radiation-induced lung injury (SRILI) after radiation therapy. Therefore, current research has focused on identifying biologic markers or physical dose parameters that might aid in determining each individual patient's risk of developing SRILI. If identified, these markers might aid in the selection of patients who might benefit from dose escalation. To date, no biologic pretreatment predictor of SRILI has been conclusively demonstrated to identify patients at high risk of toxicity. Previous studies indicate that both physical and biologic parameters, namely dose–volume histograms and changes in plasma transforming growth factor (TGF)-β1 levels during radiation therapy, can be used to stratify patients into low-, intermediate-, and high-risk groups (5, 6). Other markers, such as interleukin (IL)-6 have been suggested from preliminary studies but have failed to provide predictive information in a larger clinical setting (7).

Reprint requests to: Mitchell S. Anscher, M.D., Duke University Medical Center, Department of Radiation Oncology, Box 3085, Durham, NC 27710. Tel: (919) 668-5637; Fax: (919) 668-7345; E-mail: anscher@radonc.duke.edu

Supported by a grant from the Laboratory Directors Fund for Innovation, the Department of Pathology.

Received Mar 14, 2005, and in revised form May 12, 2005. Accepted for publication May 12, 2005.

Previous studies have demonstrated that SRILI can be correlated with certain inflammatory events within the host, including white blood cell recruitment and subsequent activation (8). Therefore, it seems plausible that analysis of plasma inflammatory cytokine profiles might provide a pretreatment assessment of a patient's risk of developing SRILI. The goal of the present analysis was to investigate whether pretreatment cytokine levels are predictive for the development of SRILI.

METHODS AND MATERIALS

Patient eligibility

In this retrospective study, the records of all lung cancer patients treated with radiotherapy with curative intent from 1991 to 2003 at Duke University Medical Center on a series of prospective normal tissue injury studies were reviewed. A total of 55 patients (22 with SRILI and 33 without) were selected randomly from the database, according to the following inclusion criteria: (*1*) newly diagnosed lung cancer of any histology treated with radiation therapy (RT) with or without chemotherapy with curative intent, (*2*) follow-up time >6 months, and (*3*) availability of plasma samples obtained before the initiation of RT. Additionally, archival plasma samples from 39 non–cancer-bearing patients were used as controls. All studies were approved by the Duke University Medical Center Institutional Review Board. Patients gave written informed consent before enrollment.

Clinical evaluation

Pretreatment evaluation included a history and physical examination, as well as blood work, including automated blood count and kidney and liver function tests. Chest X-ray and computed tomography (CT) scan of the chest and upper abdomen were routinely performed for staging purposes. Patients with locally advanced disease also had bone scans and brain CT scans. A few patients also had mediastinoscopy and positron emission tomography scanning performed. Patients were staged according to the American Joint Committee on Cancer Staging criteria (9).

After the completion of RT, patients were generally seen every 3 months for 2 years, then every 6 months. At these follow-up visits, a history, physical examination, chest X-ray, and CT scan were obtained. Other tests, such as bone scans and brain CT scans, were done if required by protocol or by the clinical situation. As part of the protocols, patients were carefully assessed for signs of pulmonary toxicity. Additional studies, including pulmonary function testing and single photon emission computed tomography, were obtained at regular intervals, as prescribed by the individual study.

Plasma TGFβ1 quantification

The methods used for measuring the concentration of plasma TGFβ1 have been previously described (10–14). Before analysis, plasma samples were thawed and subjected to ethanol–acid extraction, as described previously (15). The enzyme-linked immunosorbent assay (ELISA) used for quantifying the concentration of plasma TGFβ1 was a basic sandwich assay. In the early years of the study, the following procedure was used. The monoclonal antibodies 12H5 (0.5 mg/mL), a non-neutralizing anti-TGFβ1 immunoglobulin (Ig)G_{2b}, *k* antibody and 4A11 (2 mg/mL), a neutralizing anti-TGFβ1 IgG_1, *k* antibody were used as capture and probe antibody, respectively (Genentech, South San Francisco, CA). The secondary antibody was horseradish peroxidase-conjugated rabbit antimouse IgG_1 (1:6000 dilution) (Zymed Laboratories, South San Francisco, CA); 2,2′-azino-di-(3-ethylbenzthiazoline 6-sulfonic acid) (Bio-Rad Laboratories, Hercules, CA) was the substrate. The 96-well microtiter plates were read at 405 nm with an automatic plate reader (V-max; Molecular Devices, Menlo Park, CA). Because the plasma TGFβ1 was activated during extraction, this procedure measures total TGFβ1 (including both active and latent forms). In addition, a commercially available sandwich ELISA was used (R&D Systems, Minneapolis, MN) (16–18). The sensitivity of both assays is <1 ng/mL. The upper limit of normal was 7.5 ng/mL with the early assay and 4.5 ng/mL with the R&D assay. In both cases, this value is 2 standard deviations above the mean for normal controls.

Cytokine analysis

These plasma samples were analyzed by the multiplex suspension array system using Luminex beads (Bio-Rad Laboratories), which included the following cytokines: IL-1β, IL-2, IL-4, IL-5, IL-6, IL-7, IL-8, IL-10, IL-12p70, IL-13, IL-17, granulocyte/macrophage colony-stimulating factor, interferon γ, monocyte chemotactic protein 1, macrophage inflammatory protein 1β, tumor necrosis factor α, and granulocyte colony-stimulating factor. Plasma samples were thawed, diluted 1:1 with diluent, and run according to the manufacturer's protocol. All samples were run in duplicate and were measured as picograms per milliliter of plasma.

Endpoints

The endpoint of these studies was the development of SRILI at any time after completion of radiotherapy. Symptomatic radiation-induced lung injury was prospectively diagnosed on the basis of the development of clinical symptoms, the evaluation of which did not reveal pneumonia, tumor recurrence, or any other specific etiology. Radiographic changes alone were insufficient to make the diagnosis of SRILI (10, 14, 19). With this modification of the common toxicity criteria of the National Cancer Institute (NCI), patients with clinical symptoms were required to demonstrate a worsening of ≥1 grade on the NCI scale to make a diagnosis of SRILI.

Statistics

In addition to descriptive statistics, our primary goal was to determine whether any cytokine or clinical variable was significantly different in the group of patients who developed SRILI compared with those who did not. We used the Kruskal-Wallis test to compare the median IL-1β, IL-2, IL-4, IL-5, IL-6, IL-7, IL-8, IL-10, IL-12p70, IL-13, IL-17, granulocyte/macrophage colony-stimulating factor, interferon γ, monocyte chemotactic protein 1, macrophage inflammatory protein 1β, tumor necrosis factor α, granulocyte colony-stimulating factor, and TGFβ1 levels in the three groups of patients (those who developed SRILI, those who did not develop SRILI, and controls). We used the Wilcoxon rank–sum test to compare TGFβ1 levels between the SRILI and non-SRILI groups. Fisher's exact test (for small cell sizes) and the χ^2 test were used when comparing SRILI with TGFβ1 levels in patients with SRILI and patients without SRILI. We used a receiver operating characteristics (ROC) graph to find the best cut-point for IL-8. On the basis of the sensitivity and specificity of the IL-8 cut-off value determined by ROC, the relative risk of SRILI and the odds ratio for patients with low IL-8 levels were deter-

mined with Clinical Calculator 2: Predictive Values and Likelihood Ratios (20).

RESULTS

Patient and treatment characteristics

The pretreatment clinical characteristics and treatment parameters of the study populations are summarized in Table 1. Patients with SRILI were slightly younger, smoked less, and had slightly better lung function than patients who did not develop SRILI. Furthermore, the group without SRILI had more male patients and more Caucasians, fewer received chemotherapy, and more had surgery. Total RT doses were somewhat higher in the SRILI group, but dose–volume histogram analysis revealed that the percent of total lung volume receiving >30 Gy (V30) was similar in patients who did or did not develop SRILI. There were no statistically significant differences between the two groups.

Cytokine profiles

A summary of 17 cytokines evaluated in the patient plasma before the initiation of curative RT is displayed in Table 2. Cytokine levels in normal, cancer-free control subjects were also measured. Among cancer patients, this analysis demonstrated a significant difference in the levels of IL-8 between patients that did or did not develop SRILI after treatment. Patients who did not develop SRILI had a median IL-8 level of 1.49 pg/mL, approximately four times higher than the IL-8 levels in patients who did develop SRILI (Fig. 1). Patients who developed SRILI had IL-8 levels that were significantly elevated as compared with normal control subjects (Table 2). Of note, the patients who developed SRILI had IL-8 levels closer to those of normal, cancer-free control subjects (medians of 0.34 pg/mL and 0.0 pg/mL, respectively) (Fig. 1). Statistical significance was not observed between these two patient populations for any of the remaining cytokines examined in this assay.

On the basis of the significance observed for IL-8 levels between cancer patients who did or did not develop SRILI, we used a ROC graph to determine an optimal cut-point for this cytokine as a predictor of SRILI (Fig. 2). The cut-point determined by this method was 1.2 pg/mL; this value provides the maximum sensitivity and specificity (0.682 and 0.576, respectively). Assuming an SRILI incidence of 15%, the positive predictive value is 22.1% if a patient has a pre-RT IL-8 level <1.2 pg/mL (20). The negative predictive value is 91.1% if a patient has a pre-RT IL-8 level >1.2 pg/mL. The odds ratio of developing SLIRI if the pre-RT IL-8 level is <1.2 pg/mL is 2.90, as compared with patients with an IL-8 level >1.2 pg/mL.

TGFβ1 analysis

Previous studies have demonstrated that TGFβ1 parameters are predictive of SRILI (10, 14, 19). Therefore, previously collected TGFβ1 levels from patients in this study were analyzed to determine whether any correlation existed between TGFβ1 and IL-8, the cytokine identified above as a predictor of SRILI. In the current analysis, TGFβ1 parameters provided no statistically significant predictive value for the development of SRILI. This includes analysis of TGFβ1 ratio (post-RT levels divided by pre-RT levels) and elevated TGFβ1 levels at the end of therapy (greater than either the upper limit of normal or the pretreatment value). The χ^2 test for association between SRILI and the TGFβ1 ratio produced a p value of 0.50. The Fisher exact test for association between SRILI and elevated TGFβ1 levels at the end of RT produced a p value of 0.20 (Table 3).

On the basis of our findings, a potential correlation between IL-8 and TGFβ1 levels was then examined. Patients with elevated treatment TGFβ1 had higher pretreatment IL-8 levels than patients with low TGFβ1, with medians of 2.06 vs. 0.73 pg/mL, respectively (p = 0.02; Wilcoxon rank–sum) (Fig. 3A). Thus, patients' pretreatment IL-8 levels were higher in patients with elevated TGFβ1 levels during therapy. Furthermore, pretreatment IL-8 was found to be a significant predictor of treatment TGFβ1 levels with a positive correlation (p = 0.0008; analysis of variance) (Fig. 3B).

Table 1. Patient and treatment characteristics

Characteristic	No SRILI	SRILI	p
Age (yr)	67.0 (33)	62.5 (22)	0.08
PCKYR	60 (30)	38.5 (18)	0.52
FEV1	57 (15)	70 (7)	0.89
DLCO	57 (15)	58 (6)	1.00
Sex (% male)	64 (33)	50 (22)	0.32
Race (% Caucasian)	88 (33)	82 (22)	0.55
% wt loss	10 (9)	11 (3)	0.47
Hist (Adeno/NSCLC/ Sq/Large/Sarcoma)	8/9/13/2/1	4/11/6/1/0	
Chemo (% yes)	42 (33)	58 (12)	0.36
Surgery (% yes)	33 (33)	25 (12)	0.61
RT dose (cGy)	6600 (33)	7240 (13)	0.25
Hemoglobin (g/dL)	12.2 (15)	12.2 (19)	0.61
V30	24.0 (10)	24.8 (12)	0.64

Abbreviations: PCKYR = Pack year smoking history; FEV1 = forced expiratory volume at 1 s; DLCO = carbon monoxide diffusing capacity; Hist = histology; Adeno = adenocarcinoma; NSCLC = non–small-cell lung cancer; Sq = squamous cell carcinoma; Large = large-cell carcinoma; Chemo = chemotherapy; RT = radiation therapy; V30 = percent of total lung volume exceeding 30 Gy.

Values are median or percentage. Numbers in parentheses are sample sizes (n).

DISCUSSION

This is the first study to evaluate the use of plasma cytokine profiling as a tool to identify patients at increased risk of SRILI. After screening patients for 17 proinflammatory cytokines, only reduced levels of IL-8 were found to correlate with the risk of developing this toxicity. A significant correlation between SRILI and previously reported TGFβ1 parameters was not found in the current study;

Table 2. Median cytokine and TGFβ1 levels

	No SRILI (n = 33)		SRILI (n = 22)		Controls (n = 39)		
	Median	IQR	Median	IQR	Median	IQR	p
Cytokine levels (pg/mL)							
IL-2	0	0–7.9	0	0–4.02	0	0–9.0	0.81
IL-4	0	0–0	0	0–0	0	0–24.1	0.91
IL-6	18.2	8.2–42.2	11.7	7.5–34.9	11.7	4.6–35.7	0.81
IL-8	1.49	0.8–3.3	0.34	0–1.81	0	0–0.78	<0.0001*
IL-10	0.23	0–0.7	0.46	0–2.69	0.6	0.06–3.23	0.32
GM-CSF	0	0–94.6	0	0–172	0	0–157.9	0.58
IFN-γ	0	0–1.3	0	0–2.65	0	0–28.6	0.33
TNF-α	0.24	0–1.2	0	0–1.27	0.08	0–1.6	0.44
IL-1β	0.03	0–0.24	0.06	0–0.18	0.1	0–0.7	0.33
IL-5	0.18	0.1–0.5	0.17	0.13–0.47	0.26	0.04–0.69	0.89
IL-7	0.72	0.3–1.6	1.03	0.40–2.32	0.74	0.41–1.17	0.48
IL-12p70	0	0–0	0	0–0	0	0–0	0.64
IL-13	0.87	0–3.0	1.30	0.4–7.0	2.6	0–5.0	0.41
IL-17	0	0–0	0	0–0	0	0–0	0.69
G-CSF	0	0–0	0	0–5.2	0	0–0	0.47
MCP-1	48.1	37.8–80.2	49.1	37.3–88.8	49.1	39.9–72.6	0.77
MIP-1β	29.9	23.1–48.5	28.0	24.8–31.7	25.9	20–34.5	0.43
TGFβ1 levels (ng/mL)							
TGFβ1 ratio	0.74	0.38–1.30	0.98	0.71–1.50			0.23
Pre-T-TGF1β	9.1	5.5–13.4	5.65	2.07–8.0			0.055
Trt-TGFβ1	6.7	2.8–9.9	3.9	2.18–6.30			0.27
Sum TGFβ1	1	1–2	1	1–2			0.72

Abbreviations: GM-CSF = granulocyte/macrophage colony-stimulating factor; IFN-γ = interferon γ; TNF-α = tumor necrosis factor α; G-CSF = granulocyte colony-stimulating factor; MCP-1 = monocyte chemotactic protein 1; MIP-1β = macrophage inflammatory protein 1β; TGFβ1 ratio = end-of-treatment TGFβ1 level/pretreatment TGFβ1 level; Pre-T-TGFβ1 = pretreatment TGFβ1 level; Trt-TGFβ1 = end-of-treatment TGFβ1 level; IQR = interquartile range.

p values for cytokines determined by Kruskal-Wallis test; for TGFβ1 levels by Wilcoxon rank-sum test.

* Significant p value.

however, elevated levels of IL-8 correlated with increased levels of TGFβ1 during therapy.

Symptomatic radiation-induced lung injury is an important and serious complication of curative RT, and the risk of this side effect routinely limits the total prescribed dose of radiation delivered to patients with tumors in and around the thorax. The incidence of SRILI ranges from 5% to 20% (21–26), depending on the clinical setting. Concern over this potential toxicity, which can be fatal, might in fact limit the ability to adequately treat large thoracic tumors (1). In light of these issues, current research has focused on identifying biologic and physical parameters that might aid in determining an individual's relative risk of developing SRILI before the initiation of RT.

On the basis of the role of inflammatory cytokines (10, 14, 27–29) and white blood cells (8) in the development of SRILI, we hypothesized that a patient's general inflammatory status before treatment might affect their normal tissue response to thoracic radiation. Therefore, in the present study we investigated each patient's inflammatory status before RT, using a screen for plasma levels of various inflammatory cytokines.

Here we demonstrate that there is a significant difference in plasma IL-8 levels before the initiation of RT between patients who will and patients will not go on to develop SRILI. The ROC-determined cut-point for IL-8 levels between these two groups was 1.2 pg/mL. The resulting sensitivity, specificity, and odds ratio were 0.682, 0.576, and 2.90, respectively. Although we hypothesized that patients with increased levels of inflammatory cytokines before RT

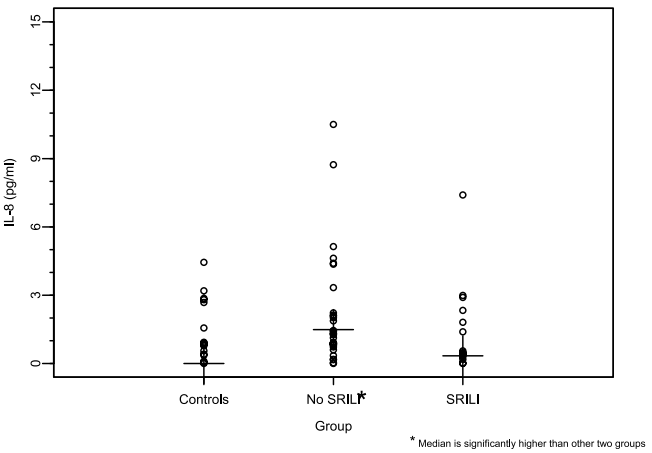

Fig. 1. Distribution of interleukin (IL)-8 levels between control subjects, patients with no symptomatic radiation-induced lung injury (SRILI), and patients with SRILI. Observed and median IL-8 levels are indicated for patients in the control, no-SRILI, and SRILI groups. *$p < 0.05$, SRILI vs. no-SRILI.

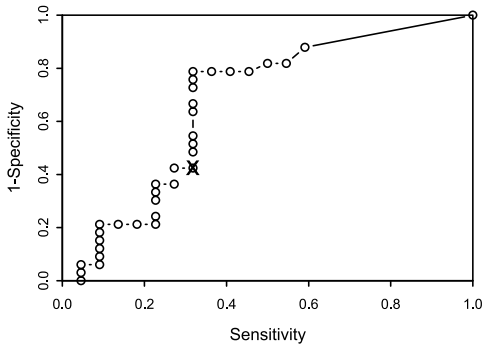

Fig. 2. The receiver operating characteristics curve allows exploration of the relationship of sensitivity and specificity of interleukin-8 for predicting the target clinical outcome of symptomatic radiation-induced lung injury for a variety of different cut-points, thus allowing the determination of an optimal cut-point.

might be at increased risk of normal tissue injury after radiation, the data demonstrate that patients with increased levels of IL-8 have a decreased risk of developing SRILI. Consistent with previous reports, both sets of cancer patients (with and without SRILI) had increased levels of IL-8 as compared with normal control subjects (30). However, the patients who developed SRILI seemed to have IL-8 levels that were closer to the levels present in normal, cancer-free control subjects. These observations suggest that IL-8 might be protective against SRILI.

The fact that IL-8 levels are higher in cancer patients suggests that this cytokine might be produced within the tumor environment and possibly as a result of tumor–host interaction. Indeed, previous studies have suggested that non–small-cell lung cancer cells produce IL-8 as a result of interaction with infiltrating macrophages (31). Our data suggest that the degree of IL-8 upregulation might affect the sensitivity of the lung to RT.

Interleukin-8 was originally identified as a neutrophil chemotactic factor that was isolated from stimulated human mononuclear cells (32, 33). Since its identification, a number of structurally related cytokines have been characterized. Because of their ability to exhibit chemotactic activity for specific leukocytes, this family of cytokines has been recognized as chemokines (34, 35). Additionally, IL-8 induces chemotactic responses in basophils and T lymphocytes (36). More recently, IL-8 has demonstrated angiogenic activities through inducing migration of endothelial cells (37, 38). This chemokine is also capable of inducing a loss of focal adhesion, followed by chemotaxis and chemokinesis of fibroblasts (39).

Although IL-8 demonstrates chemotactic activity for neutrophils, this is believed to only transiently regulate the influx of these cells during initiation of the inflammatory process. Chronic upregulation of this chemokine, on the other hand, results in long-term impaired neutrophil migration (40). Simonet *et al.* suggest that prolonged exposure to increased levels of circulatory IL-8 decreases neutrophil expression of L-selectin, a surface receptor believed to mediate leukocyte rolling on endothelial cells (41). Rodents overexpressing human IL-8 demonstrated increased accumulation of neutrophils in the microcirculation of lung, liver, and spleen and decreased migration into sites of inflammation. Therefore, it is possible that cancer patients with increased levels of circulating IL-8 might also have impaired neutrophil migration to sites of inflammation and tissue injury, including sites of radiation-induced tissue damage. This reduction of neutrophil migration might then ameliorate the proliferation of profibrotic changes in SRILI. More recent evidence demonstrates that L-selectin-deficient knockout mice exhibit significantly increased survival as compared with wild-type littermates after 20 Gy irradiation to both lungs (42). Although the exact mechanism of this protection was not elucidated, it is possible that the effect was due to reduced neutrophil migration to the lung after radiation.

Interleukin-8 levels have also been identified as a poor prognostic indicator for patients with non–small-cell lung cancer (30, 31, 43, 44). Yuan *et al.* (44) demonstrated that high expression of IL-8 in tumor tissue was highly associated with advanced-stage disease, distant lymph node metastasis, increased microvessel count, short survival, and early relapse. Therefore, although it might indicate a decreased risk of developing SRILI, these patients are likely to have more aggressive disease with decreased overall survival.

Although IL-8 levels are a significant predictor of SRILI in this retrospective study, we were unable to find a significant association between the previously established TGFβ1 parameters and the incidence of SRILI in this patient subset. Furthermore, although both an increasing TGFβ1 ratio (post-RT TGFβ1/pre-RT TGFβ1) and elevated levels of TGFβ1 during treatment have been shown to predict SRILI (12), our data indicate that increased levels of IL-8 are associated with increased TGFβ1 ratio and elevated TGFβ1 levels during RT. It is noted that TGFβ1 parameters were not predictive of SRILI in this patient subset, so the relevance of the relationship between IL-8 and TGFβ1 remains uncertain for purposes of predicting SRILI.

These data seem to be very promising in the search for parameters that might provide predictive risk assessments for SRILI before the initiation of RT. It is notable that the sensitivity and specificity of predicting SRILI on the basis of pretreatment IL-8 levels (68.2% and 57.6%, respectively) approach the sensitivity and specificity of predicting prostate cancer on the basis of prostate-specific antigen levels

Table 3. Analysis of dichotomous TGFβ1 variables

	No SRILI	SRILI	*p*
TGFβ1 ratio (% elevated)	49	45	0.51
Trt-TGFβ1 (% elevated)	39	23	0.20

Abbreviations as in Table 2.

TGFβ1 ratio = 1 if TGFβ1 ratio $>$ 1, 0 if TGFβ1 ratio $<$ 1; Trt-TGFβ1 = 1 if Trt-TGFβ1 $>$ cutoff value, 0 if Trt-TGFβ1 $<$ cutoff value; cutoff value = 7.5 ng/mL or 4.5 ng/mL, depending on TGFβ1 assay used for measurement.

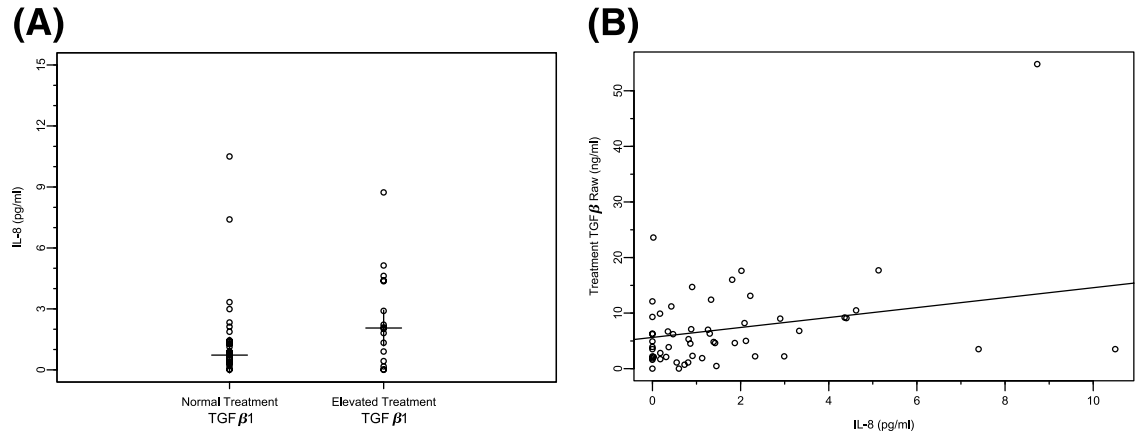

Fig. 3. Correlations between transforming growth factor (TGF)β1 and interleukin (IL)-8. (A) Distribution of IL-8 levels between patients with normal and elevated treatment levels of TGFβ1. Patients with treatment TGFβ1 levels less than predetermined cutoff values (7.5 ng/mL or 4.5 ng/mL, depending on the TGFβ1 assay used) were identified as normal treatment TGFβ1. Patients with treatment TGFβ1 levels greater than predetermined cutoff values (7.5 ng/mL or 4.5 ng/mL, depending on the TGFβ1 assay used) were identified as elevated treatment TGFβ1. (B) Scatterplot of IL-8 levels by raw treatment levels of TGFβ1, fitted with least-squares regression line ($n = 54$). The positive slope of the regression line indicates IL-8 increases as TGFβ1 increases.

(67.5–80.0% and 60–70%, respectively) (45). Presently, the use of this parameter requires further investigation in a larger, prospective study for validation. These studies will provide positive and negative predictive values for IL-8 levels in determining the risk of SRILI. Evaluation of a larger number of patients might also provide additional multivariate factors (e.g., V30 and other physical parameters) that could be acquired before treatment to more accurately assess an individual's risk of developing SRILI. Additionally, these data suggest that there might be biologic variables present among cancer patients, potentially due to the extent of tumor–host interaction, that might affect the inflammation and wound response that occurs after therapeutic radiation.

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