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Fibroblasts as Target and Effector Cells in Japanese Patients with Sarcoidosis

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Abstract. Fibroblasts play a crucial role in progressive lung fibrosis, acting not only as target cells but also as effector cells. To clarify these functions in sarcoidosis, lung fibroblasts from Japanese sarcoid patients were studied for their proliferative capacity and cytokine productivity. Fibroblasts were cultured from transbronchial lung biopsy specimens from seven patients with sarcoidosis. As a comparison, fibroblasts from open lung biopsy specimens of four patients with idiopathic pulmonary fibrosis (IPF) were studied. For controls, fibroblasts were cultured from specimens of normal resected lung tissue of five patients with localized lung cancer. The proliferative activity of cultured fibroblasts from patients with sarcoidosis was highest among the three groups ($p < 0.05$). However, the proliferative capacity in all groups was suppressed when fibroblasts were cultured with interleukin-1β (IL-1β). No significant differences were noted in the degree of inhibition among the three groups. Addition of interferon-γ (IFN-γ) also resulted in inhibition of fibroblast growth in all groups, but the degree of inhibition was significantly greater in both the sarcoid and IPF groups than in controls ($p < 0.05$). The amount of interleukin-6 (IL-6) in the culture supernatants from sarcoid fibroblasts cocultured with IL-1β was significantly higher than in controls. Sarcoid fibroblasts are not only proliferatively active but also possess effector cell function to produce cytokines. IL-6 may enhance the immunologic reaction to sarcoidosis and cause the disease to become chronic. IFN-γ suppresses proliferation of sarcoid fibroblasts and may prevent fibrotic changes of the lungs in the Japanese sarcoid patients.

Key words: Sarcoidosis—Fibroblasts—Interleukin-6—Interleukin-1—Interferon-γ.

Introduction

In fibrosing lung diseases, peripheral blood mononuclear cells and alveolar macrophages (AM) work as effector cells to stimulate or inhibit fibroblast proliferation and

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collagen synthesis [3]. The regulation of the immune response in pulmonary fibrosis has been studied in terms of the interaction between AM obtained from bronchoalveolar lavage (BAL) of diseased lungs and fibroblast cell lines obtained from normal lungs or organs with specific diseases [1, 7, 25, 29]. Recent investigations have examined the function of fibroblasts derived from various diseases [19, 24, 28]. This attention has been focused mainly on fibrosing lung diseases other than sarcoidosis, thus it is unknown whether fibroblasts in patients with sarcoidosis in Japan have a role in the disease process.

Granulomas in sarcoidosis are characterized by a central collection of inflammatory cells surrounded by fibroblasts, leading to an interaction between these cells [21]. Most sarcoid fibrotic lesions are derived from solitary or confluent granulomas, with subsequent invasion of fibroblasts and collagen fibers into the granulomas [36]. One likely pathophysiology of this fibrotic inflammatory process is: (1) fibroblasts are recruited to the pulmonary parenchyma and proliferate locally; (2) mediators secreted by mononuclear cells in the lesions result in fibroblast proliferation; and (3) regulatory factors from the proliferating fibroblasts in turn modulate local inflammation. Recent studies support the production of various cytokines by fibroblasts, which may contribute to the inflammatory process [11, 22]. This cytokine-mediated fibroblast-inflammatory cell interaction may involve interleukin-8 (IL-8)-mediated chemotaxis [31], interleukin-6 (IL-6) [6], and the cellular effects of granulocyte-macrophage colony-stimulating factor (GM-CSF) [10].

From a clinical viewpoint, resolution of lung lesions occurs spontaneously in most Japanese patients with sarcoidosis, but in 15% of patients or fewer, pulmonary fibrosis develops [16]. Fibroblast inhibitory factors from AM supernatants have been speculated [7], but the possibility that the fibroblasts in these Japanese cases of sarcoidosis have different characteristics has yet to be investigated.

This study was initiated to determine whether fibroblasts from Japanese patients with sarcoidosis are functionally altered, and if so, whether this alteration affects the pathophysiology of sarcoidosis.

Materials and Methods

Study Population

The patients with sarcoidosis consisted of two men and five women with a mean age of 37.1 ± 13.5 years. Sarcoidosis was diagnosed by the clinical picture and the presence of epithelioid cell granulomas in specimens from lung, skin, or liver. Patients were subdivided according to the x-ray classification of their disease: two were stage 0, one was stage I, two were stage II, and two were stage III [34]. All patients had normal pulmonary function. Extrapulmonary manifestations of sarcoidosis were noted in one patient with an intramuscular lesion, one patient with a skin lesion, one with liver and stomach involvement, and four with ocular lesions.

As a comparison group, we studied three men and one woman (mean age 58.7 ± 14.8 years) with idiopathic pulmonary fibrosis (IPF) based on open lung biopsy specimens taken from the sites of milder lesions. All four were asymptomatic and were histopathologically confirmed to have usual interstitial pneumonia (UIP). Although all four had a normal Pao_2, their mean percent of vital capacity (%VC) was reduced to 62.7%.

Two men and three women (mean age 58.8 ± 6.5 years) who served as controls had resection of a

solitary lung cancer. These control patients were asymptomatic and had normal %VC and Pa_{O_2} values. No patients had received immunosuppressive drugs or corticosteroids before transbronchial lung biopsy (TBLB) or open lung biopsy.

TBLB Procedures

TBLB was performed for all sarcoid patients after local anesthesia with lidocaine through a flexible fiberoptic bronchoscope (BF type 1T20, Olympus Corp., Tokyo, Japan). The tip of the bronchoscope was introduced into the segmental bronchi, and an Olympus alligator forceps (model FOB20C) was inserted to obtain at least three specimens. The selection of biopsy sites was done with reference to the computed tomographic findings.

Fibroblast Culture

TBLB specimens were taken from radiologically normal areas in sarcoid patients with stages 0 and I and from affected sites in those with stages II and III. Open lung biopsy specimens in IPF were taken from the affected sites seen in computed tomogram. Control specimens were taken from macroscopically normal areas of resected tissue with lung cancer. Lung specimens were washed with phosphate-buffered saline, calcium- and magnesium-free (PBS(−) solution). After removing inappropriate tissue, the specimens were chopped into pieces with the size of about 1 mm^3 and washed three times with PBS(−), then twice with regular growth medium (RGM). RGM consisted of Dulbecco's modified Eagle's medium (DMEM, Sigma, St. Louis, MO) supplemented with 10% fetal bovine serum (FBS) (Life Technologies, Inc., Grand Island, NY), 1% nonessential amino acid solution (Life Technologies, Inc.), 0.75 mg/liter $NaHCO_3$ (Katayama, Nagoya, Japan), and 30 mg/liter gentamicin sulfate (Wako, Osaka, Japan).

The tissues were placed into a six-well polystyrene dish (Corning Glassworks, Corning, NY) with a coverslip so as to adhere to the dishes. After the addition of 2 ml of RGM, these tissues were incubated at 37°C and 5% CO_2. Medium was changed twice weekly. When the monolayer of cells reached confluence, usually after 4–6 weeks, the explanted tissue was removed. The cells were trypsinized for 10 min, after which they were resuspended in RGM and replated in 75-mm^3 tissue culture flasks with 20 ml of RGM. Subsequently, the cells were split 1:4 at confluence and then passaged. For the following experiments, we used a three to ten population doubling level for each assay.

For morphologic characterization of fibroblasts, cell monolayers were examined directly with a phase-contrast microscope (Olympus IMT-2). Cells were also examined by electron microscopy. Briefly, cell monolayers were fixed in 2% glutaraldehyde (Nakarai Desk, Kyoto, Japan), treated with 1.0% osmium tetraoxide (Wako), dehydrated in alcohols, and embedded in Questol 812 (Nissin EM Corp., Tokyo, Japan). Ultrathin sections were double stained with uranyl acetate (Merck Japan, Tokyo, Japan) and lead citrate (Katayama) and then examined with a JEM 100CX (JEOL, Tokyo, Japan) electron microscope. To exclude mycoplasma contamination, the cultured fibroblasts were checked with the MycoTect kit (Life Technologies, Inc.) prior to each assay.

Fibroblast Proliferation

Fibroblasts from confluent culture were trypsinized and resuspended at 5×10^4/ml in a flat bottomed 12-well plate (Corning). After 24 h, the RGM was removed, and each well was washed twice with serum-free DMEM to render the cells quiescent. One ml of serum-free DMEM was added to each well, and the cells were cultured for 48 h, after which cells were counted with a hemocytometer. This was the starting point for each experiment (day 0). The medium was changed to RGM, and cells were then incubated for various intervals up to 96 h (day 4) at 37°C and 5% CO_2. At the end of each incubation period, the cells were washed, trypsinized, and counted. Proliferation rate was defined as the ratio of cell numbers at each day to day 0.

To examine the response of fibroblasts to exogenous interleukin-1β (human recombinant IL-1β; Amersham Corp, Buckinghamshire, U.K.) and interferon-γ (human recombinant IFN-γ, Genzyme Research

Product, Cambridge), various concentrations of IL-1β and IFN-γ were added to the test medium at day 0. The highest concentration of IL-1β was 1 ng/ml, and the highest for IFN-γ was 1,000 units/ml. At days 1, 2, and 4, cells were trypsinized and counted. To determine whether changes in fibroblast growth in the presence of IL-1β related to the arachidonate pathway, exogenous indomethacin (1 μg/ml, Prasel Lorei, Frankfurt, Germany) was added to the culture medium.

All studies were performed in triplicate, and all three groups used the same lot number of FBS as a supplement. Cell viability was assessed with the trypan blue exclusion method. Viability was 95% or greater in all experiments.

Cytokine ELISA of Fibroblast Supernatants

After replacing the RGM with serum-free DMEM and incubation for 48 h to stabilize the cells, confluent fibroblasts were stimulated with 1 ng/ml IL-1β only, 1,000 units/ml IFN-γ only, or both for 24 h. IL-6, IL-8, and GM-CSF in the culture supernatants was determined using sensitive enzyme-linked immunosorbent assay (ELISA) kits (Amersham). Briefly, 100 μl of sample was added to microtiter wells coated with monoclonal antibody against IL-6, IL-8, or GM-CSF and incubated for 2 h. The plates were washed and incubated with 200 μl of horseradish peroxidase-conjugated anti-IL-6, IL-8, or GM-CSF for 2 h. After rinsing to remove excess conjugates, color development with tetramethylbenzidine and hydrogen peroxide for 20 min was measured with an automated ELISA plate reader at 450 nm optical density. The value of each cytokine in the supernatant was determined by referring to a standard curve.

Statistical Analysis

Data were analyzed with a Macintosh II computer using the correlation program of the Stat view 4.0 package (Abacus Concept, Inc., Berkeley, CA) and were expressed as mean ± S.D. Data were compared using the one-way analysis of variance (ANOVA).

Results

Characteristics of Fibroblast Cultures

Cell monolayers of tissue obtained from each group, i.e. controls, sarcoidosis, and IPF patients, were found to have typical spindle shapes by phase-contrast microscopy. Although cellular sizes varied from patient to patient, size was unrelated to the origin of the cultured cells (data not shown). Ultrastructural features of cultured cells showed a homogeneous nature with well developed endoplasmic reticulum, intracytoplasmic fibrils, and a nucleolus but poor phagolysosome and microvillous processes. These are regarded as the typical features of fibroblasts.

Fibroblast Proliferation

Preliminary experiments with fibroblasts in serum-free DMEM indicated an increase in proliferative activity of less than 10% after 1–4 days (data not shown). We therefore used RGM for subsequent assays of fibroblast proliferation, such that all cultured fibroblasts were subconfluent by day 4.

Figure 1 shows the fibroblast proliferation rates of the three groups in RGM over time. The fibroblast cultures derived from sarcoidosis were found to have a signifi-

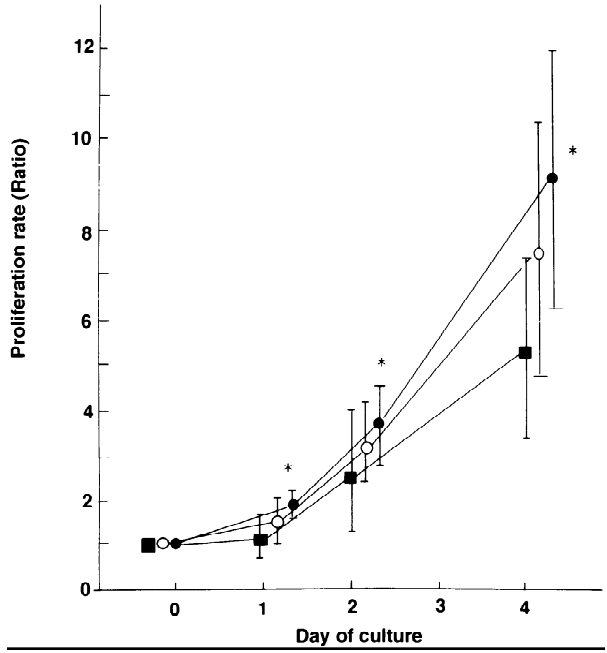

Fig. 1. Proliferative capacity of fibroblasts from sarcoidosis and IPF patients. Results are expressed as the mean ± S.D. of seven sarcoidosis patients (*closed circles*), four IPF patients (*open circles*), and five controls (*closed squares*). Fibroblasts from each group were cultured with DMEM, 10% FBS up to day 4. At days 0, 1, 2, and 4, cells were counted by hemocytometer, and the proliferative rate was defined as the ratio of cell number at each day to day 0. The proliferative rate of the sarcoid fibroblasts was greater than that of the controls at days 1, 2, and 4. The *asterisks* (*) indicate $p < 0.05$, in control vs sarcoidosis values.

cantly greater proliferative rate compared with controls at day 1 ($p < 0.05$), day 2 ($p < 0.05$), and day 4 ($p < 0.05$).

Effects of IL-1β and IFN-γ on Fibroblast Proliferation

Preliminary assays determined that IL-1β in the range from 0.01 to 1 ng/ml and IFN-γ from 10 to 1,000 units/ml inhibited fibroblast proliferation at day 2 and day 4 in a dose-dependent manner (data not shown). Thus, 1 ng/ml for IL-1β and 1,000 units/ml for IFN-γ were used. The inhibitory effect of both agents was not due to cytotoxic effects as confirmed by trypan blue assessment of viability (>95%).

On day 4, fibroblast proliferation in both the sarcoidosis and IPF groups after 1 ng/ml IL-1β tended to be lower than controls, but this difference was not significant (Fig. 2). However, day 4 proliferation after 1,000 units/ml IFN-γ was decreased significantly compared with controls (Fig. 2).

To determine the mechanism of the inhibitory action of IL-1β on fibroblast growth, indomethacin (1 μg/ml) was tested in three patients from each group (Table 1). Indomethacin alone at a concentration of 1 μg/ml had no effect on fibroblast prolif-

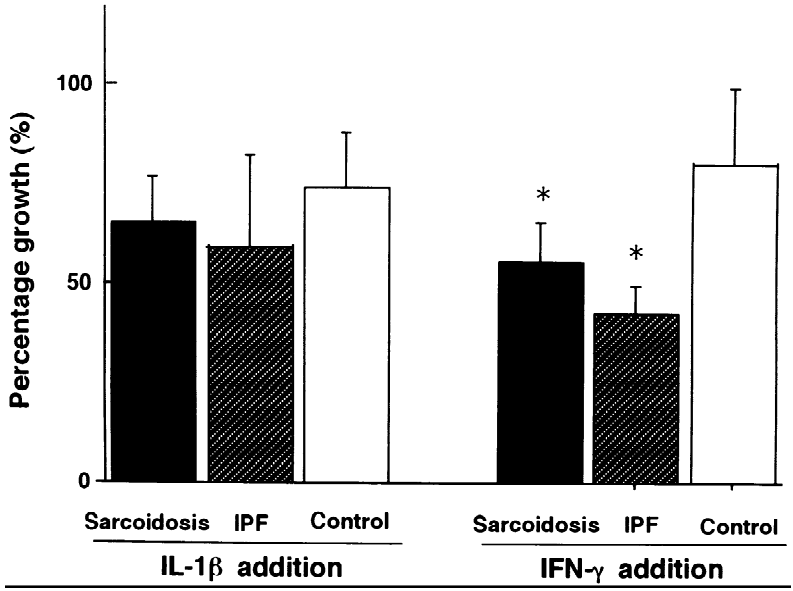

Fig. 2. Effects of the addition of IL-1β (1 ng/ml) or IFN-γ (1,000 units/ml) on fibroblast proliferation at day 4. The *vertical bar* represents percentage growth after stimulation with IL-1β or IFN-γ compared with growth in DMEM, 10% FBS. Each value represents the mean ± S.D. *Left,* in the DMEM, 10% FBS culture, the addition of IL-1β suppressed fibroblast proliferation of all groups compared with 10% FBS only. *Right,* in the DMEM, 10% FBS culture, IFN-γ suppressed fibroblast proliferation. The degree of suppression in the sarcoidosis and IFP patients was greater than in the controls. The *asterisks* (*) indicate $p < 0.05$ compared with controls.

Table 1. Effect of indomethacin and IL-1β on fibroblast growth at day 4

	No.	IL-1β[a] (%)	Indomethacin[b] (%)	IL-1β and Indomethacin (%)
Sarcoidosis	3	80.1 ± 6.3[c]	105.5 ± 4.5	116.0 ± 17.0
IPF	3	77.8 ± 11.5	101.9 ± 4.1	102.3 ± 8.6
Controls	3	73.9 ± 19.5	103.4 ± 7.4	105.0 ± 16.5

[a] IL-1β, 1 ng/ml
[b] Indomethacin, 1 μg/ml
[c] Percentage growth compared with DMEM, 10% FBS culture (100%)

eration, but on day 4, indomethacin reversed the inhibition caused by IL-1β, suggesting that the inhibitory effect of IL-1β may be related to the arachidonate pathway. As to the degree of suppression among the three groups, no significant differences were noted. When IL-1β and IFN-γ were added simultaneously to fibroblasts, there was an additive effect on fibroblast growth inhibition but without a significant difference among the three groups (data not shown).

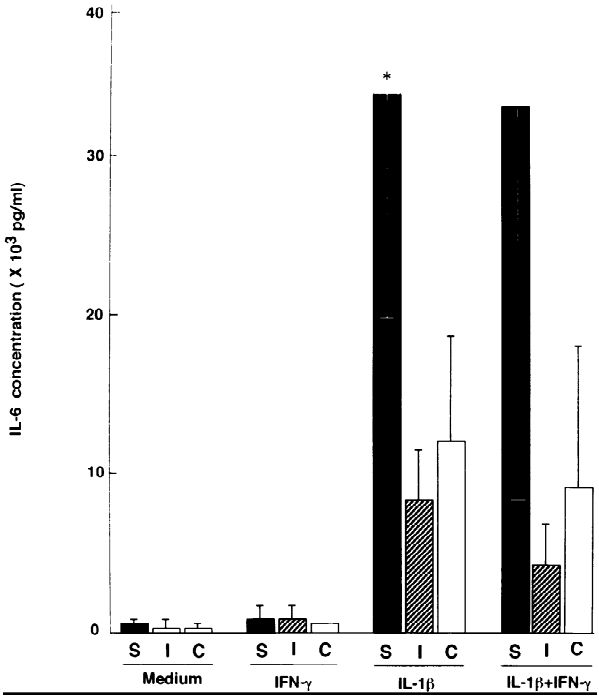

Fig. 3. IL-6 production by fibroblasts from sarcoidosis patients (*S*), IPF patients (*I*), and controls (*C*). Confluent fibroblasts with serum-free DMEM were stimulated with IFN-γ (1,000 units/ml), IL-1β (1 ng/ml), or both for 24 h. IL-6 in the culture supernatants was determined by ELISA. The amounts of IL-6 production by sarcoid fibroblasts following IL-1β increased significantly over that of the controls. Each *bar* represents the mean ± S.D. The *asterisk* (*) indicates $p < 0.05$ compared with controls. It is noted that IFN-γ did not suppress IL-6 production of sarcoid fibroblasts in the presence of IL-1β.

Cytokine Production by Fibroblasts

Based on a preliminary experiment to determine the optimal concentration of IL-1β and IFN-γ as stimulators as well as to determine the time course of the cell cultures, we performed the following experiments at an IL-1β concentration of 1 ng/ml and IFN-γ concentration of 1,000 units/ml with an incubation time of 24 h. Fibroblasts were incubated in serum-free DMEM prior to the assay. Because the increase in cell numbers during this experiment was less than 10%, any increase in cytokine production reflects an increase in production/cell rather than a general increase in cell number. Reproducibility of each cytokine production was achieved by examining at different population doubling levels in all groups (data not shown).

As seen in Figure 3, IL-6 production by sarcoid fibroblasts after IL-1β was significantly greater than controls ($p < 0.05$). Furthermore, in the presence of IL-1β, IFN-γ did not suppress IL-6 production by sarcoid fibroblast but showed a tendency to suppress IL-6 in IPF and controls. As shown in Figures 4 and 5, IL-8 and GM-CSF production induced by IL-1β and IFN-γ were not significantly different among the

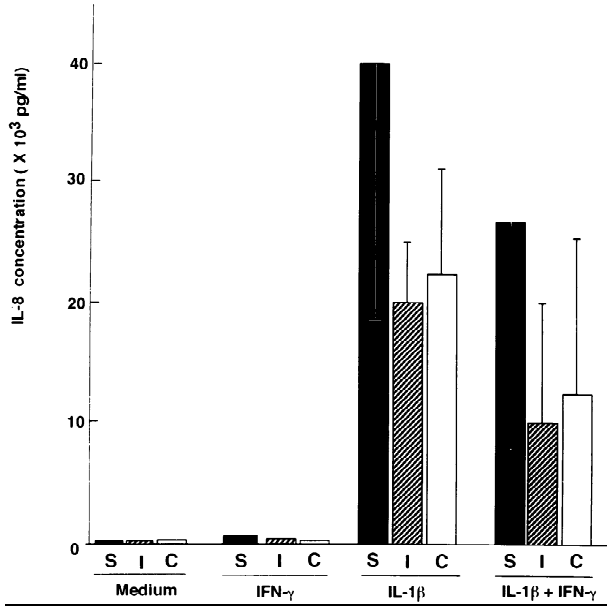

Fig. 4. IL-8 production by fibroblasts from sarcoidosis patients (*S*), IPF patients (*I*), and controls (*C*). Culture conditions were the same as for Figure 3. In the presence of IL-1β, the amount of IL-8 production from the sarcoid fibroblasts was not significantly different from other two groups. IFN-γ suppressed IL-8 production of fibroblasts from all groups in the presence of IL-1β.

three groups. Interestingly, IFN-γ suppressed IL-8 and GM-CSF production of fibroblasts in all groups in the presence of IL-1β (Figs. 4 and 5), but IFN-γ did not suppress IL-6 production in sarcoidosis (Fig. 3).

Figure 6 shows the relationship between the proliferation rate of sarcoid fibroblasts and their cytokine production. Only IL-6 production following stimulation with IL-1β for 24 h tended to show a correlation with the proliferation rate.

Discussion

This study examined the proliferative characteristics of fibroblasts established from tissue specimens from the lungs of patients with sarcoidosis. This proliferation was significantly suppressed by IFN-γ. Furthermore, IL-6 production in sarcoidosis fibroblasts stimulated by IL-1β was significantly increased, but IFN-γ did not suppress this increase in IL-6 production.

We chose IL-1β and IFN-γ because they have been implicated in the pathophysiology of sarcoidosis [33, 37], although conflicting data have been reported [13, 27, 30, 38] and also because changes in cytokine production by cultured fibroblasts after IL-1β and IFN-γ have not been investigated.

TBLB specimens were used for this study because this is the procedure of choice for patients with sarcoidosis, providing good diagnostic yields [12]. We minced TBLB

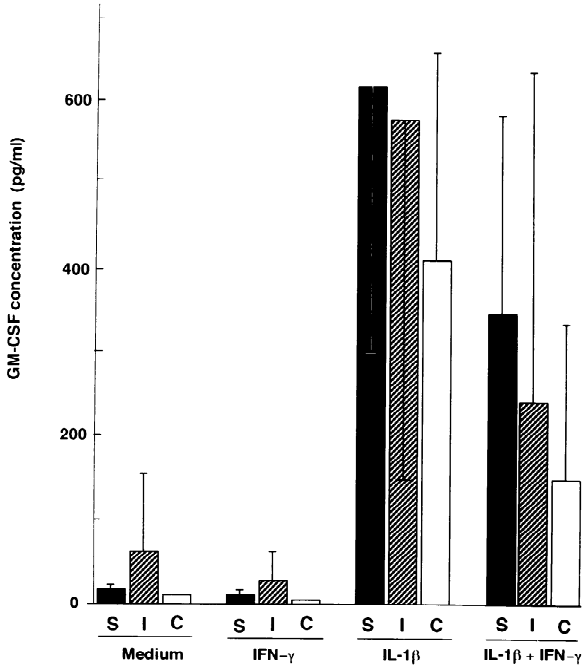

Fig. 5. GM-CSF production by fibroblasts from sarcoidosis patients (*S*), IPF patients (*I*), and controls (*C*). Culture conditions were the same as for Figure 3. In the presence of IL-1β, the amounts of GM-CSF production from the sarcoid fibroblasts was not significantly different from other two groups. IFN-γ suppressed GM-CSF production of fibroblasts from all groups in the presence of IL-1β.

specimens and surgically resected specimens into pieces with almost the same size; the time reached to confluence was not related to these sampling procedures. In fact, we found no morphologic differences between sarcoid fibroblasts from TBLB and those from specimens taken from IPF patients or controls obtained by open lung biopsy. Specimens from TBLB were not contaminated by colonizing bacteria from the upper airway.

The growth characteristics of sarcoidosis fibroblasts have not been fully examined. Fireman et al. have shown that AM supernatants from sarcoid patients suppressed the proliferative capacity of fibroblasts derived from the BAL of different patients with sarcoidosis [8]. In four patients with early fibrosis, two of whom had sarcoidosis, a heightened proliferative potential was noted [28]. Our findings in a larger number of patients are similar. The accelerated proliferation of cultured fibroblasts from sarcoidosis may result from polyclonal fibroblast activation or, alternatively, from fibroblast subpopulations with functional heterogeneity with respect to growth characteristics. Jordana et al. have speculated that the presence of a small but significant number of faster growing clones may be important in the development of fibrosis [19].

The effect of IL-1β on fibroblast proliferation was inhibitory in all tested groups. Jordana et al. have demonstrated that purified IL-1 could inhibit the proliferation of normal fibroblasts [18]. By contrast, other studies have shown that IL-1 stimulates

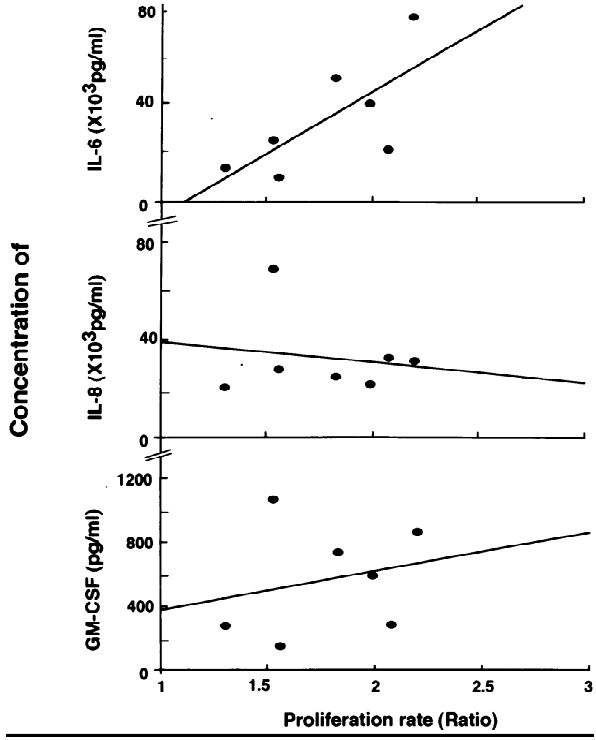

Fig. 6. Relationship between the rate of fibroblast proliferation and cytokine production in sarcoid fibroblasts. The rate of fibroblast proliferation was determined in DMEM-10% FBS culture at day 1. Cytokine production was measured in a culture of serum-free DMEM in the presence of IL-1β (1 ng/ml) at day 1. IL-6 production correlated slightly with the rate of fibroblast proliferation ($r = 0.507$).

fibroblast proliferation [2, 26, 32] or has no effect on fibroblast proliferation [5]. This discrepancy may be mainly because of differences in utilized cell lines, assay conditions, supplements, and the purity or concentration of IL-1. The concentration of serum used by Jordana et al. [18] and Elias et al. [5] was used in this study.

Elias et al. [5] found that IL-1β alone had a mild stimulatory effect on fibroblast proliferation, but dose-dependent inhibition of fibroblast proliferation occurred when fibroblasts were exposed simultaneously to recombinant IL-1β and tumor necrosis factor (TNF). These were shown to stimulate prostaglandin production synergistically in fibroblasts. Elias and colleagues concluded that contaminating TNF in partially purified IL-1 could explain Jordana's results. For our study, we used recombinant IL-1β to avoid any TNF contamination. To exclude the possibility of an effect of other cytokines that were contaminated or newly secreted by the fibroblasts following incubation with IL-1β, we measured the levels of TNF-α, TNF-β, and IL-1α by ELISA in fibroblast supernatants and found all values to be negligible. The indomethacin inhibition assay also suggests that the fibroblast proliferative response to IL-1β is mediated through the prostaglandin pathway without any TNF interaction.

The effect of IFN-γ on fibroblast proliferation was also inhibitory. This result is consistent with that of Elias [4] who performed his experiments at the same concentrations of serum and recombinant IFN-γ. Another study that used a lesser concentration of serum found that IFN-γ stimulated fibroblast proliferation [14]. In our study, IFN-γ inhibition of sarcoid fibroblasts was significantly greater than controls, suggesting that any IFN-γ released from lymphocytes in vivo may prevent pulmonary fibrosis in sarcoidosis patients. This phenomenon may not be specific for sarcoidosis since fibroblast proliferation in IPF patients was also significantly suppressed by IFN-γ. Since the report on IFN-γ in the pathogenesis of IPF is limited, unlike sarcoidosis, the significance of IFN-γ in IPF remained unclear.

IL-6 production from sarcoid fibroblasts after stimulation with IL-1β was significantly greater than controls and correlated slightly with fibroblast proliferation. IL-6 functions as a growth stimulatory factor for B cells, a costimulator of mitogen-dependent T cell proliferation, and an activator of neutrophils [35]. With regard to IL-6 levels in BAL fluid of patients with sarcoidosis, there are conflicting data that showed either increase [20] or no increase [17]. We have confirmed IL-6 production in patients with sarcoidosis. A slight correlation between IL-6 production and fibroblast proliferation fits the finding of Fries et al. [9] that IL-6 secreted from pulmonary fibroblasts stimulated fibroblast growth in an autocrine manner. We speculate the following interaction between AM and fibroblasts. IL-1 produced by AM or blood monocytes recruited into the lungs stimulates fibroblasts to produce IL-6; secreted IL-6 not only nonspecifically amplifies local and systemic inflammatory events but also stimulates fibroblast growth in an autocrine manner; this fibroblast-based amplification results in persistent pulmonary inflammation as seen in sarcoidosis.

In our study IL-6 production was not suppressed by IFN-γ in the presence of IL-1β. This suggests that IL-6 production and fibroblast proliferation are regulated differently with regard to IFN-γ response. In other words, IFN-γ may inhibit the progression to pulmonary fibrosis, but not inhibit IL-6-induced inflammation in sarcoidosis.

The production of IL-8 and GM-CSF was not statistically significant among three groups. Unlike IL-6, IL-8 and GM-CSF production from sarcoid fibroblasts did not correlate with fibroblast proliferation and was inhibited with the addition of IFN-γ as IPF and controls. These findings suggest that IL-8 and GM-CSF are not the main mediators with regard to fibroblast-related inflammation in sarcoidosis. However, messenger RNA expression of IL-8 and GM-CSF in BAL cells from patients with sarcoidosis have been reported [15, 23], and these cytokines have been associated with lung injury in such patients. Whether or not enhanced fibroblastic release of IL-8 and GM-CSF participates in the initiation or stimulation of an ongoing inflammatory process requires further study.

The average age of the sarcoid patients was slightly younger than that of controls because relatively younger people are liable to suffer from sarcoidosis. But these patterns of cytokine production were specific for three disease groups, not age dependent.

Proliferative fibroblastic activity and cytokine production did not correlate significantly with the value of serum angiotensin-converting enzyme, the percentage of BAL lymphocytes and the radiological staging. In addition, although the prognosis of

sarcoidosis did correlate slightly with these two functions, this correlation was not statistically significant.

We conclude that in addition to the ability of fibroblasts to proliferate in the lungs of patients with sarcoidosis, their augmented function as effector cells may be important to the pathophysiology of this disease.

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