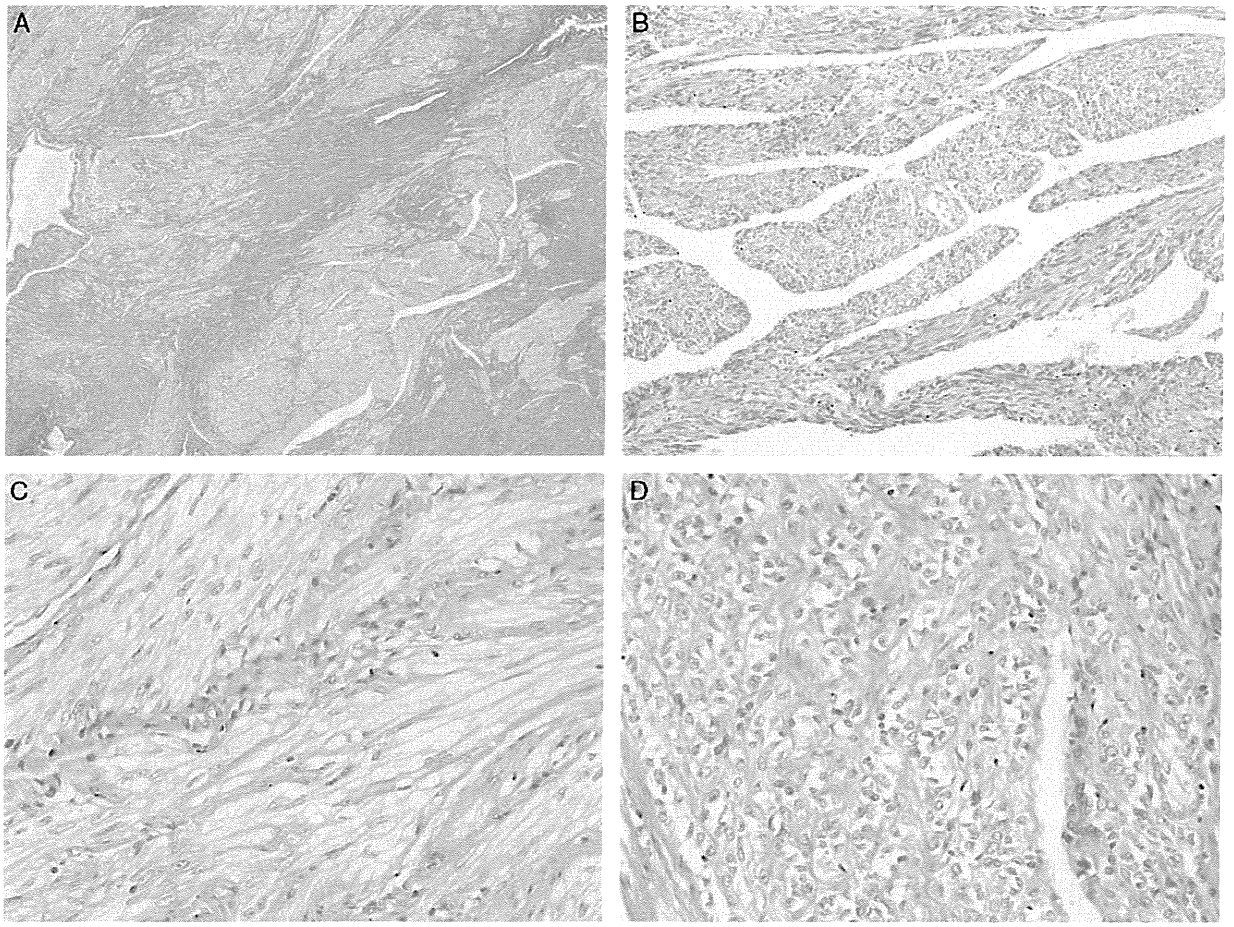

FIGURE 3. Representative pathologic findings of uterine LAM lesions in TSC-LAM patients. The multiple uterine lesions of patients with TSC were ill defined and intermixed with myometrium, although focal LAM lesions including slit-like spaces were detected (A). Some areas contained spindle-shaped LAM cells with pale eosinophilic cytoplasm arranged in fascicles around slit-like spaces (B). In addition to spindle-shaped LAM cells with clear cytoplasm and nuclei devoid of pleomorphism or mitotic activity (C), epithelioid polygonal cells with mild nuclear atypia (D) were also seen in patients with TSC (A-D, hematoxylin-eosin stain).

DISCUSSION

This study demonstrates for the first time that women with pulmonary LAM are commonly afflicted with LAM in their genital organs as well, most frequently (90%) within the uterus. To our knowledge, Lack et al[15] were the first to describe the existence of uterine LAM lesions in a patient who had pulmonary and extrapulmonary LAM with bilateral renal angiomyolipomas. However, until now, LAM has rarely been considered to involve the uterus. To date, few case reports regarding uterine LAM have been published, and most of them have yielded similar results: (1) all patients were TSC; (2) uterine involvement was evident only microscopically; and (3) the myometrium contained multiple LAM lesions.[11,16,23] Although the results of those case reports resemble ours, this study reveals that uterine LAM frequently accompanies not only TSC-LAM but also sporadic LAM. Furthermore, the uterine lesions of TSC-LAM have histologic features that are somewhat different from those of sporadic LAM. In TSC-LAM, uterine LAM lesions had no clear margins, and a tongue-like growth pattern was more apparent; however, compared with sporadic LAM, slit-like spaces outlined by VEGFR-3–positive lymphatic endothelial cells were less frequent. In addition, epithelioid-shaped LAM cells with immunopositivity for HMB45 were more prominent in TSC-LAM patients.

An important consideration for the differential diagnosis of uterine LAM includes diffuse leiomyomatosis, intravascular leiomyomatosis, disseminated peritoneal leiomyomatosis, benign metastasizing leiomyoma, epithelioid leiomyosarcoma, and endometrial stromal sarcoma. The microscopic size of lesions, their multifocality, and their intimate admixture with dilated lymphatic vascular channels are the histologic features most helpful for distinguishing LAM from other leiomyomatous tumors of the uterus.[11] The immunoreactivity for HMB45 is also a helpful marker,[3] although several reports have shown that a subset of conventional leiomyomas, conventional leiomyosarcomas, epithelioid leiomyosarcoma, low-grade endometrial stromal sarcomas, and mixed stromal/smooth

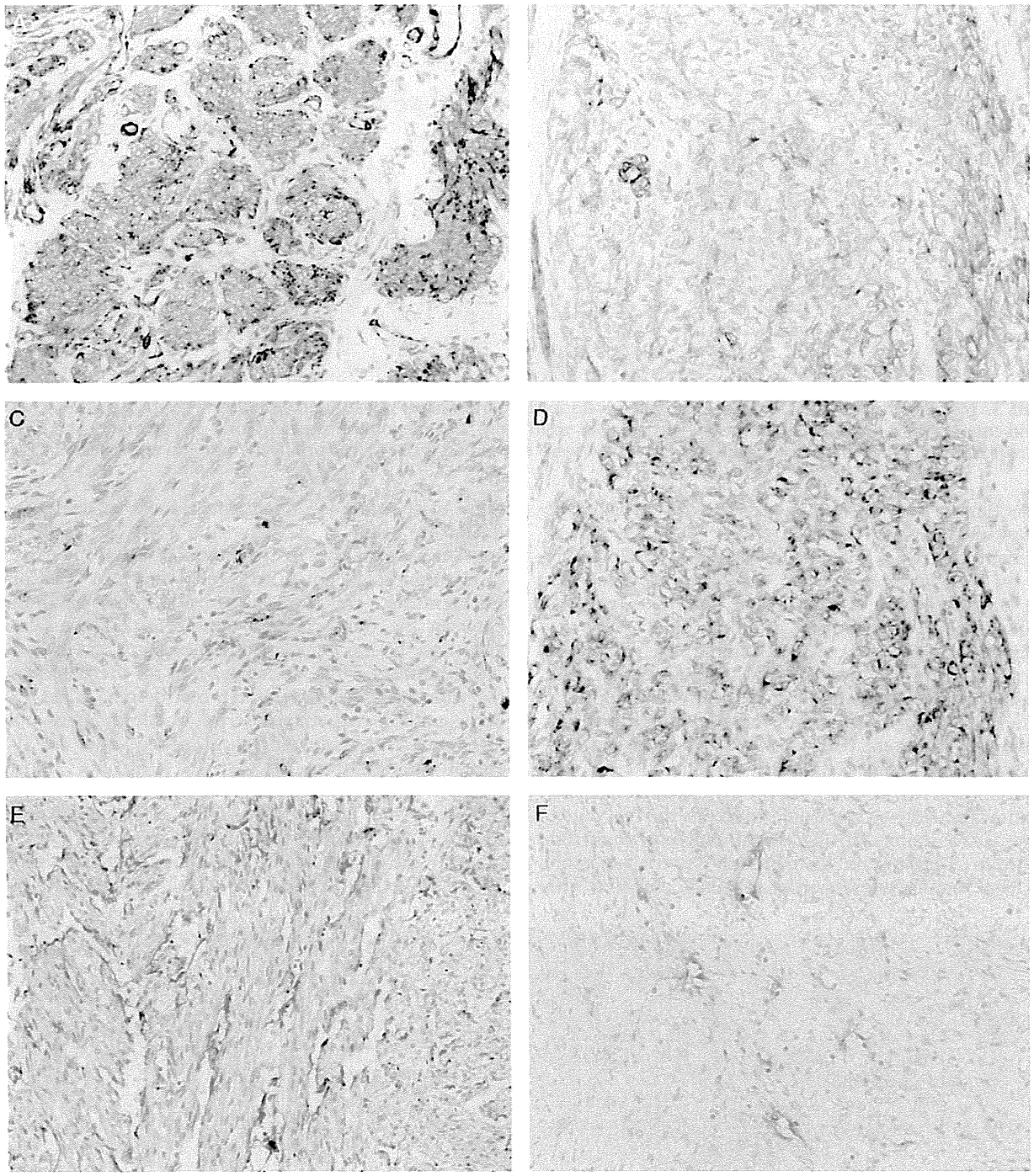

FIGURE 4. Representative immunohistologic findings of uterine LAM lesions. Immunohistochemically, spindle-shaped cells of sporadic LAM (A) and epithelioid-shaped cells of TSC-LAM patients (B) showed cytoplasmic staining with α-SMA. The spindle-shaped cells of sporadic LAM were focally positive for HMB45 (C), but epithelioid-shaped LAM cells of TSC patients were diffusely positive for HMB45 (D). Abundant slit-like spaces in lesions of patients with sporadic LAM were outlined by VEGFR-3–positive lymphatic endothelial cells (E). A contrasting few, but distinctive, VEGFR-3–positive lymphatic spaces were present in the uterine LAM lesions of TSC-LAM patients (F).

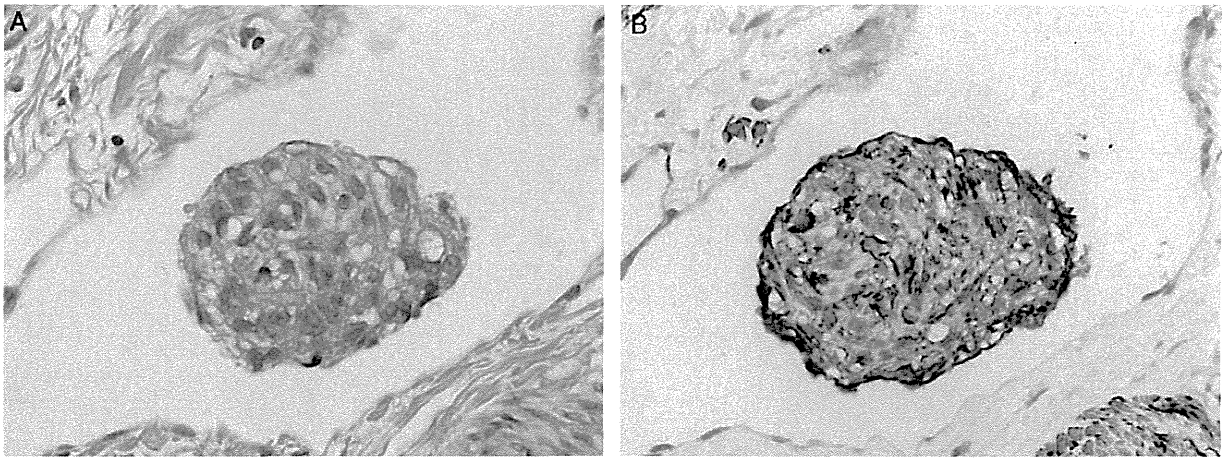

FIGURE 5. LCC in lymphatics. A cluster of LAM cells enveloped by a monolayer of flattened endothelial cells is seen freely floating with no connection to surrounding tissue (A, hematoxylin-eosin stain). Double immunostaining for α-SMA (brown) and VEGFR-3 (red) revealed that LAM cells immunopositive for α-SMA were enveloped by VEGFR-3–positive lymphatic endothelial cells (B).

muscle tumors can be immunopositive for melanocytic markers including HMB45.[6,21,22,28]

Zamboni et al[27] introduced the term "PEComa" in 1996 for the clear cell "sugar" tumor of the pancreas. Vang and Kempson[24] described 2 different types of uterine "PEComa." The first, "group A tumors," showed a tongue-like growth pattern similar to low-grade endometrial stromal sarcomas and comprised cells with abundant clear-to-eosinophilic pale granular cytoplasm, diffuse HMB45 expression, and focal immunoexpression of myogenic markers. In contrast, "group B tumors" comprised epithelioid cells with prominent clear cell features, less HMB45 immunopositivity, and extensive immunoexpression of myogenic markers. In this study, uterine LAM lesions of TSC-LAM patients would fall into group A because of their growth pattern and more prominent immunoexpression of HMB45 compared with those of sporadic LAM patients. However, as Vang and Kempson[24] stated in their study, LAM lesions may contain a small subset of epithelioid cells, but they are rarely numerous enough to be classified as a "PEComa." Furthermore, we showed an additional relationship between the uterus, pelvic, and retroperitoneal lesions and those of the lung in both sporadic LAM and TSC-LAM patients. Thus, we believe that the uterine lesions in the women described here are best designated as LAM. Some pathologists may describe our findings as uterine "PEComa." It is an emerging concept and the World Health Organization says that the "PEComa" family of tumors includes angiomyolipoma, clear cell "sugar" tumor of the lung, LAM, clear cell myomelanocytic tumor of the falciform ligament/ligamentum teres, and unusual clear cell tumors of the pancreas, rectum, abdominal serosa, uterus, vulva, thigh, and heart.[8] Thus,

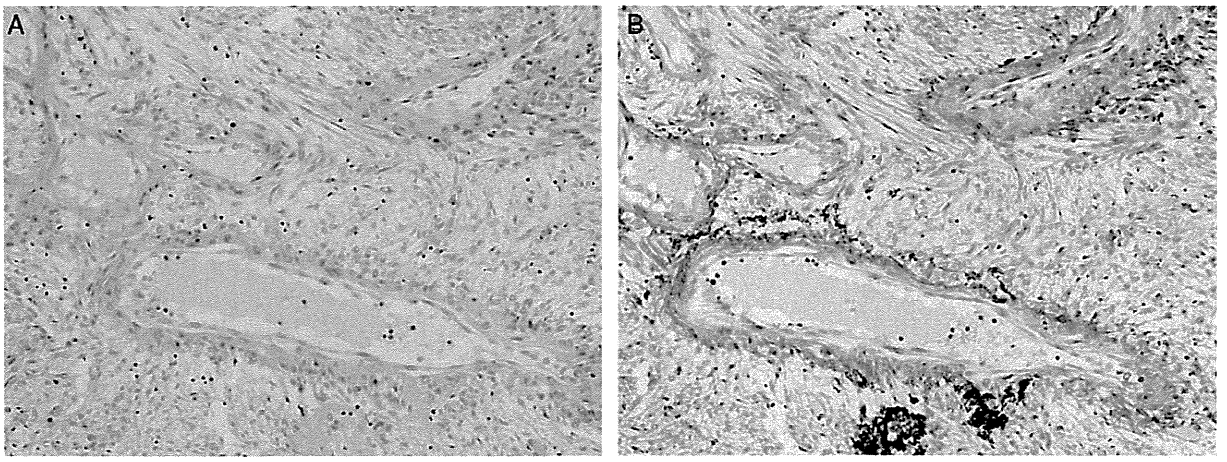

FIGURE 6. Vascular involvement of proliferating LAM cells. Spindle-shaped and epithelioid-shaped LAM cells with clear-to-pale eosinophilic cytoplasm invaded the wall of spiral arteries in the uterine myometrium (patient 9) (A, hematoxylin-eosin; B, Elastica-Masson trichrome). Note that elastic fibers in the arterial wall disappeared partially or completely upon invasion of LAM cells (B).

we agree with the use of the term uterine "PEComa" for a patient with pulmonary LAM if their lesions are composed primarily of epithelioid-shaped cells and cannot be categorized into LAM.[25] Furthermore, we partly agree with the suggestion that both the term "PEComa" and smooth muscle tumor terminologies be used to make clinicopathologic documentation of these rare tumors concretely. For this aim, uterine lesions in the women described here may be designated as "LAM PEComa," as described in another study.[4]

The mechanism for the development of uterine LAM remains undetermined, but one may speculate as follows. First, the uterus could be the primary site of origin of LAM. Our results and those from a patient who had microscopic uterine LAM lesions but no other medical history indicating pulmonary or renal disease[4] suggest that uterine lesions are common and are the earliest manifestations of LAM. Yet, several arguments dispute this hypothesis. In this study, a patient with sporadic LAM (patient 6) did not have uterine LAM lesions, although we could not completely exclude the possibility that such lesions might have been too tiny for our level of detection. The occurrence of LAM shows an extreme predilection for women, but 1 case in a man has been published.[1] The expression of melanosomal proteins by LAM cells suggests that a precursor of LAM cells could originate from a neural crest lineage, a cell type that is reported to migrate into various organs during development.[10] Second, the uterus may be one of the most frequent metastatic or disseminated sites of LAM. The metastatic potential of LAM cells was clearly demonstrated by their presence in circulating blood.[5] LAM cells also express lymphangiogenic growth factors including VEGF-D and may spread through lymphatic channels.[14] Judging from these combined findings, LAM can metastasize to the uterus by hematogenous or lymphatic spread. In this study, all LAM patients who had uterine LAM lesions concomitantly suffered a proliferation of LAM cells in retroperitoneal or pelvic lymph nodes and LCCs in lymphatics. LAM lesions in pelvic lymph nodes were detected even in the LAM patient (patient 6) who had no uterine LAM lesion. In addition, the frequency of extrapulmonary LAM lesions was highest in lymph nodes of the retroperitoneum and pelvic cavity along the axial lymphatics,[14] indicating that LAM cells may originate in this area. Therefore, the uterine involvement of LAM could result from a lymphatic spread emanating somewhere in the retroperitoneum or pelvic cavity. However, this pattern of spread seems extraordinarily rare in conventional pelvic tumors, and the proliferating pattern of uterine LAM in TSC-LAM patients (that is, a tongue-like growth structure) seems rare when tumors metastasize to the uterine corpus. Third, the mechanisms for the development of uterine LAM may differ between TSC and the sporadic form. In this study, uterine lesions of sporadic LAM had slit-like spaces outlined by lymphatic endothelial cells more abundantly than TSC-LAM, suggesting differing extents of lymphangiogenesis. Speculation follows that lymphangiogenesis has a more important role in the progression of uterine lesions from sporadic LAM. However, no apparent differences between TSC-LAM and sporadic LAM have been reported in the literature regarding the histologic features of extragenital organs including lungs and lymph nodes. Finally, we cannot exclude the possibility of multifocal LAM lesions in the context of a field effect, nor the possible association between hormonally sensitive smooth muscle and the development of these lesions in women.

In summary, we have demonstrated for the first time a high incidence of uterine LAM in patients with pulmonary LAM. Furthermore, the histologic features of uterine LAM associated with TSC-LAM were unlike those of sporadic LAM. Our results raise the intriguing possibility that the uterus or a nearby locale in the retroperitoneum or pelvic cavity may be the primary site where LAM arises, although the origin and pathobiology of this disease remain subjects of continued investigation.

ACKNOWLEDGMENT

The authors thank Phyllis Minick for excellent assistance in the review of English.

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Pulmonary Endothelial Impairment During Gefitinib Therapy: A Preliminary Assessment with Iodine-123-Metaiodobenzylguanidine (123I-MIBG) Scintigraphy

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Abstract: Iodine-123-metaiodobenzylguanidine (^{123}I-MIBG) kinetics in the lung could serve as a novel diagnostic tool to evaluate endothelial damage. Interstitial lung disease (ILD) associated with gefitinib, an epidermal growth factor receptor (EGFR)-tyrosine kinase inhibitor (TKI), has been reported as a serious adverse effect. This study was performed to examine the possibility that gefitinib induces pulmonary endothelial damage. Serial ^{123}I-MIBG scintigraphy was performed in 5 patients with non-small cell lung cancer before and one month after initiation of gefitinib treatment. Anterior planar images were acquired 15 min after injection of ^{123}I-MIBG and the total lung to upper mediastinum ratio (L/M) was calculated in both lungs. None of the patients developed ILD during the study. There were no significant differences in the values of L/M before and after gefitinib therapy. These findings suggest that gefitinib has little influence on the pulmonary endothelium in patients with no signs of ILD.

Keywords: Interstitial lung disease, acute lung injury, non-small cell lung cancer, EGFR, drug-induced lung injury.

INTRODUCTION

Iodine-123-metaiodobenzylguanidine (^{123}I-MIBG) has been widely used for the detection of various neuroendocrine tumors and evaluation of adrenergic dysfunction in the heart [1,2]. ^{123}I-MIBG is taken up by the lung through a saturable, energy-requiring, sodium-dependent transport mechanism similar to biogenic amines, such as serotonin and norepinephrine [3,4]. It is well known that transport of these biogenic amines requires normal endothelial cell integrity. Thus, ^{123}I-MIBG is regarded as an indicator of pulmonary endothelial function [3-6], because it behaves in a quantitatively similarly manner to norepinephrine in the pulmonary circulation [4]. Slosman *et al.* initially evaluated lung uptake of ^{123}I-MIBG using animal models and demonstrated decreased ^{123}I-MIBG lung extraction in bleomycin-induced endothelial injury [6]. Furthermore, several clinical studies using this property of ^{123}I-MIBG scintigraphy showed that decreased ^{123}I-MIBG kinetics in the lung could serve as a novel diagnostic tool to evaluate the endothelial damage in patients with pulmonary fibrosis [7], myeloperoxidase anti-neutrophil cytoplasmic antibody-positive vasculitis [8], high altitude-related hypoxia [9], and high altitude pulmonary edema [10].

Gefitinib is an epidermal growth factor receptor (EGFR)-tyrosine kinase inhibitor (TKI) and shows antitumor activity in patients with advanced non-small cell lung cancer (NSCLC) [11]. In particular, higher response rates have been reported in Asian EGFR mutation-positive NSCLC patients [12,13]. However, cases of fatal gefitinib-induced interstitial lung disease (ILD) have been reported [14-16], and higher incidence rates of ILD have been reported in Japanese NSCLC patients receiving gefitinib than in other countries [15, 16]. The underlying mechanisms of gefitinib-induced ILD remain unclear. EGFR is expressed on a number of cells of the lungs, including epithelial cells, smooth muscle cells, fibroblasts, and endothelial cells [17, 18]. Gefitinib-induced ILD shows diffuse alveolar damage, including epithelial and endothelial areas.

Our hypothesis was that gefitinib caused pulmonary endothelial damage in patients with NSCLC, even in those with no clinical respiratory manifestations and/or abnormal chest radiographic findings. Thus, the present study was performed to determine the effects of gefitinib administration on the pulmonary uptake of ^{123}I-MIBG before and after gefitinib therapy.

MATERIALS AND METHODS

This study conformed with the provisions of the Declaration of Helsinki 1995. All subjects were informed of the procedures and risks of this study, and informed consent was obtained prior to participation in the study.

Subjects

Patients with histologically or cytologically confirmed NSCLC were enrolled in this study. Patients with active infection, interstitial pneumonia on chest computed tomography (CT), pericardial effusion that required drainage, active brain metastasis, pregnancy, or Eastern Clinical Oncology Group (ECOG) performance status > 2 were excluded from the trial. In addition, patients who showed atelectasis due to tumor mass or pleural effusion on chest radiograph were also excluded. Other concomitant

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Table 1. The Clinical Characteristics of Patients Enrolled in this Study and the ^{123}I-MIBG Lung Uptake (L/M)

Case	Age	Gender	Smoking (Pack · yrs)	Histology	Previous Chemotherapy (current Line)	Response to Gefitinib	(rt.L/M+lt.L/M)/2	
							Before Treatment	After 1 Month
1	61	M	82	Sq	+(4th)	SD	1.33	1.35
2	73	M	106	Sq	−(1st)	SD	1.26	1.36
3	47	F	12	Ad	+(3rd)	SD	1.61	1.82
4	79	M	0	Ad	+(3rd)	SD	1.19	1.26
5	69	F	0	Ad	+(4th)	SD	1.77	1.78
average	65.0						1.35	1.44
±SD	14.1						0.18	0.25

anticancer therapy or experimental drug administration of any type was not permitted. Furthermore, none of the subjects were receiving medications that could affect ^{123}I-MIBG uptake and metabolism, such as beta-adrenergic blocking drugs, catecholamine, and angiotensin converting enzyme inhibitors.

^{123}I-MIBG scintigraphy was performed before and one month after the initiation of gefitinib therapy. Patients were started on treatment with gefitinib at a dose of 250 mg daily after initial ^{123}I-MIBG scintigraphy. Patients were monitored by chest radiography and blood gas analysis during gefitinib therapy. If the enrolled patient showed a good response and achieved partial response assessed by the response evaluation criteria in solid tumors (RECIST Version 1.0) within one month after commencement of gefitinib therapy, second examination of ^{123}I-MIBG scintigraphy was canceled, because we speculated that radiographic improvement could affect the ^{123}I-MIBG lung uptake.

MIBG Scintigraphy

A dose of 111 MBq of ^{123}I-MIBG (Daiichi Radioisotopes Labs, Tokyo, Japan) was administered intravenously after a 15-min rest period with the patient lying undisturbed in bed. Anterior planar images were acquired 15 min after injection of ^{123}I-MIBG, and stored in a 64×64 matrix by a scintillation camera (ZLC 7500; Siemens, Solna, Sweden) equipped with a low-energy, general purpose collimator interfaced to a minicomputer (SCINTIPAC 2400; Shimadzu, Kyoto, Japan). The energy window was set at the 159 keV photopeak of ^{123}I. The region of interest (ROI) was placed over the upper mediastinum, the right and left lung in planar images. ROIs corresponding to the contours of the right and left lungs were manually assigned with reference to the isocount line. Total counts of each lung and heart were measured and the geometric mean was calculated as counts per pixel. To quantify the degree of lung uptake of ^{123}I-MIBG, the lung to upper mediastinum ratio in ^{123}I-MIBG uptake (L/M) was measured for the right (R) and left (L) lungs. These data were used to calculate the mean value (R+L/2; L/M).

Statistical Analysis

Data are expressed as means ± SD. The paired *t* test was used to analyze differences in ^{123}I-MIBG scintigraphic data. In all analyses, $P < 0.05$ was taken to indicate statistical significance.

RESULTS

In the present study, total nine patients with NSCLC (5 men and 4 women, with a mean age of 61.0 years, range 34-79 years) were enrolled in the present study. Among them, 4 subjects showed good response to gefitinib within one month and excluded from the following examination of ^{123}I-MIBG lung uptake. Thus, the comparative analysis of serial ^{123}I-MIBG lung uptake was performed using 5 patients. The clinical characteristics and the ^{123}I-MIBG lung uptake (L/M) in the present study are summarized in Table 1. There were three men and two women, with a mean age of 65.0 years, range 47 – 79 years. Three patients were ex-smokers. The histological types were squamous cell carcinoma (2 cases) and adenocarcinoma (3 cases). The response in all patients within one month was stable disease. The value of L/M in nine enrolled patients before gefitinib therapy was 1.44 ± 0.25, which was not statistically significant from that in selected 5 subjects for comparison with following ^{123}I-MIBG examination. In the 5 subjects, there were no statistically significant differences in ^{123}I-MIBG lung uptake before (1.35 ± 0.18, ranging from 1.19 to 1.77) and after (1.44 ± 0.25, ranging from 1.26 to 1.82) gefitinib treatment.

DISCUSSION

In this study, serial evaluations of ^{123}I-MIBG lung uptake were performed in patients with NSCLC before and after gefitinib treatment. None of the subjects developed ILD or showed new abnormal radiographic findings in the lungs. There were no statistically significant differences in ^{123}I-MIBG lung uptake before and after gefitinib treatment, suggesting that gefitinib administration did not cause pulmonary endothelial damage.

In terms of gefitinib-induced ILD, the interaction between epithelial cells or fibroblasts and the expression of EGFR has been discussed [19,20]. Indeed, the levels of epithelial expression of EGFR and its ligands, EGF and transforming growth factor-α, were increased in fibrotic lung disease [19]. However, the overexpression of EGFR in tumor-associated endothelial cells from clinical specimens of human lung cancer has also been reported [18]. The appearance of ILD in patients treated with gefitinib was

uncommonly rapid and the pathological finding was diffuse alveolar damage, similar to acute lung injury. The pathophysiology mainly involved damage to both epithelial and endothelial cells. EGFR-TKI could affect biological status of pulmonary endothelial cells, including apoptosis [18]. Therefore, in the present study, we focused on whether gefitinib could influence pulmonary endothelial cell impairment in a clinical situation using ^{123}I-MIBG lung uptake. The onset of gefitinib-induced ILD is frequency observed within one month [15,16]. Therefore, second examination of ^{123}I-MIBG lung uptake in the present study was selected at one month after the start of gefitinib therapy.

There were limitations to interpretation of our results in the present study. First, in contrast to our hypothesis, we found that gefitinib itself does not directly influence endothelial cells and that gefitinib-induced ILD is dependent on the individual. That is, pulmonary endothelial damage may be induced secondary to the development of gefitinib-induced ILD. Indeed, animal experiments showed no evidence that gefitinib induces lung injury in intact rat lungs [20]. Second, the number of patients enrolled in this study was too small to detect endothelial damage by ^{123}I-MIBG lung uptake. We have demonstrated that only 6 cases of myeloperoxidase anti-neutrophil cytoplasmic antibody-positive vasculitis were enough to detect the impaired pulmonary endothelial function [8]. However, Takabatake *et al.* [7] summarized ^{123}I-MIBG lung uptake in 23 patients with idiopathic pulmonary fibrosis and demonstrated significantly decreased ^{123}I-MIBG lung uptake in patients with idiopathic pulmonary fibrosis compared with controls. They concluded that endothelial cell injury played a significant role in the pathogenesis of idiopathic pulmonary fibrosis. Based on these studies, further case studies might be needed to draw definitive conclusions.

Risk factors for gefitinib-induced ILD have been identified in a number of studies in the Japanese population, including sex, history of smoking, concurrent interstitial pneumonitis, history of prior chemotherapy, and poor performance status [15,16]. However, little information is available regarding predictive factors for gefitinib-induced ILD. Thus, it is necessary to find markers to elucidate which patients are at high risk of ILD before gefitinib therapy. The present study failed to demonstrate the usefulness of ^{123}I-MIBG lung uptake. However, we believe that pulmonary endothelial impairment due to exposure to the administered drug could be involved. Determination of the scintigraphic kinetic behavior of ^{123}I-MIBG in the lung may yield some new insight for detecting pulmonary endothelial damage.

CONFLICT OF INTEREST

None

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Received: November 11, 2010 Revised: January 1, 2011 Accepted: January 10, 2011

The NEW ENGLAND JOURNAL of MEDICINE

ESTABLISHED IN 1812 APRIL 28, 2011 VOL. 364 NO. 17

Efficacy and Safety of Sirolimus in Lymphangioleiomyomatosis

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ABSTRACT

BACKGROUND

Lymphangioleiomyomatosis (LAM) is a progressive, cystic lung disease in women; it is associated with inappropriate activation of mammalian target of rapamycin (mTOR) signaling, which regulates cellular growth and lymphangiogenesis. Sirolimus (also called rapamycin) inhibits mTOR and has shown promise in phase 1–2 trials involving patients with LAM.

METHODS

We conducted a two-stage trial of sirolimus involving 89 patients with LAM who had moderate lung impairment — a 12-month randomized, double-blind comparison of sirolimus with placebo, followed by a 12-month observation period. The primary end point was the difference between the groups in the rate of change (slope) in forced expiratory volume in 1 second (FEV_1).

RESULTS

During the treatment period, the FEV_1 slope was −12±2 ml per month in the placebo group (43 patients) and 1±2 ml per month in the sirolimus group (46 patients) ($P<0.001$). The absolute between-group difference in the mean change in FEV_1 during the treatment period was 153 ml, or approximately 11% of the mean FEV_1 at enrollment. As compared with the placebo group, the sirolimus group had improvement from baseline to 12 months in measures of forced vital capacity, functional residual capacity, serum vascular endothelial growth factor D (VEGF-D), and quality of life and functional performance. There was no significant between-group difference in this interval in the change in 6-minute walk distance or diffusing capacity of the lung for carbon monoxide. After discontinuation of sirolimus, the decline in lung function resumed in the sirolimus group and paralleled that in the placebo group. Adverse events were more common with sirolimus, but the frequency of serious adverse events did not differ significantly between the groups.

CONCLUSIONS

In patients with LAM, sirolimus stabilized lung function, reduced serum VEGF-D levels, and was associated with a reduction in symptoms and improvement in quality of life. Therapy with sirolimus may be useful in selected patients with LAM. (Funded by the National Institutes of Health and others; MILES ClinicalTrials.gov number, NCT00414648.)

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This article (10.1056/NEJMoa1100391) was published on March 16, 2011, at NEJM.org.

N Engl J Med 2011;364:1595-606.

LYMPHANGIOLEIOMYOMATOSIS (LAM) IS AN uncommon systemic disease that is associated with cystic destruction of the lung, chylous pleural effusions, and abdominal tumors such as renal angiomyolipomas.[1,2] LAM affects women almost exclusively and occurs sporadically, developing in about 5 persons per 1 million; it also affects 30 to 40% of women with tuberous sclerosis complex (TSC). Lung function, measured as the forced expiratory volume in 1 second (FEV_1), declines at the rate of 75 to 118 ml per year[3-5]; clinically important respiratory impairment, recurrent pneumothoraxes, and hypoxemia develop in most patients within a decade after the onset of symptoms.[6]

Smooth-muscle cells that infiltrate the lung in patients with LAM appear to be benign histologically,[7] arise from an unknown source, circulate in the blood,[8] and harbor biallelic, inactivating TSC gene mutations.[9] Loss of TSC gene function constitutively activates the mammalian target of rapamycin (mTOR) signaling pathway, which regulates multiple cellular functions, including growth, motility, and survival.[10] LAM cells also express two lymphangiogenic growth factors, vascular endothelial growth factor C (VEGF-C) and vascular endothelial growth factor D (VEGF-D), and spread through lymphatic channels.[11,12] Current evidence, together with reports of recurrence of LAM after lung transplantation,[13,14] suggests that LAM is a low-grade, metastatic neoplasm that selectively targets the lung (see video).

A video showing a Cine MRI of a woman with TSC and LAM is available at NEJM.org

Sirolimus (also called rapamycin) blocks mTOR activation of downstream kinases and restores homeostasis in cells with defective TSC gene function.[10] The cells that make up LAM lesions in the lung exhibit activation of the mTOR pathway and ex vivo sensitivity to the antimitogenic effects of sirolimus.[15] Administration of sirolimus in rodent models of TSC has been shown to cause regression of neoplastic growths in the liver and kidney.[16,17] Recent phase 1–2 trials[18,19] of sirolimus in patients with TSC or LAM showed that there was a reduction in the size of angiomyolipomas and, in some cases, improvement in lung function; however, the relative risks and benefits of sirolimus in patients with LAM remain unclear.[20] We conducted an international, multicenter, randomized, placebo-controlled study to test the hypothesis that treatment with sirolimus for 1 year would improve lung function in patients with LAM.

METHODS

STUDY PATIENTS

Patients were eligible for inclusion in the study if they were women 18 years of age or older, had an FEV_1 after bronchodilation of 70% of the predicted value or less, and had received a diagnosis of LAM on the basis of findings of compatible cystic change on high-resolution computed tomography plus at least one of the following criteria: confirmation of LAM by means of a biopsy, a serum VEGF-D level of 800 pg per milliliter or higher,[21] or clinically consistent findings (an existing diagnosis of TSC, a prior chylous pleural effusion, or a history of renal angiomyolipoma). Exclusion criteria were a current or planned pregnancy, large chylous fluid collections, and prior lung transplantation. All patients provided written informed consent on documents approved by the local committee charged with oversight of human subjects research. Further details of the inclusion and exclusion criteria are provided in the Supplementary Appendix, available with the full text of this article at NEJM.org.

STUDY DESIGN AND END POINTS

The study was designed by the investigators, approved by the data and safety monitoring board at the National Center for Research Resources and the institutional review board at each participating site, and conducted within the National Institutes of Health Rare Lung Diseases Consortium. The LAM Foundation assisted with recruitment of patients and with study logistics. The data, collected with the use of Internet-based electronic case-report forms, were reported to the data management and coordinating center, where they were securely held and analyzed. All the authors participated in the writing of the first and subsequent drafts of the manuscript and in the decision to submit the manuscript for publication and vouch for the completeness and veracity of the data and data analyses. Pfizer provided the drug and the money for the costs of study visits but had no role in the design or conduct of the study or the analysis or reporting of the data. The protocol, including the statistical analysis plan, is available at NEJM.org.

The study design included a screening visit and a 12-month, double-blind, placebo-controlled treatment period, followed by a 12-month observation period during which no patients received a

study drug and all patients remained unaware of their treatment assignment. Patients who met the eligibility criteria were randomly assigned by the data management and coordinating center, in a 1:1 ratio, to receive oral sirolimus, at an initial dose of 2 mg per day, or matched placebo. Sirolimus levels were measured at each follow-up visit; the results of these measurements were revealed only to an independent medical monitor, who made dosing recommendations to maintain sirolimus trough levels between 5 and 15 ng per milliliter, as well as corresponding sham dose adjustments in the placebo group.

The primary outcome measure was the FEV_1 response, which was assessed as the rate of change in FEV_1 (FEV_1 slope) in milliliters per month. Secondary outcome measures included responses in forced vital capacity (FVC), measured as changes from baseline to 12 months; lung volumes (residual volume, functional residual capacity, and total lung capacity); the distance covered on a 6-minute walk test; diffusing capacity of the lung for carbon monoxide; serum VEGF-D levels; and scores on the St. George's Respiratory Questionnaire, the Medical Outcomes Study 36-Item Short-Form Health Survey (SF-36), the Functional Performance Inventory, the General Well-Being Questionnaire, and the EuroQOL visual-analogue scales assessing fatigue, dyspnea, and quality of life. Study visits occurred at baseline, at 3 weeks, and at 3, 6, 9, 12, 18, and 24 months. Primary and secondary end points were measured at baseline and at every visit after the 3-week visit, as described in the study calendar in the Supplementary Appendix.

Adverse events were assessed according to the Common Terminology Criteria for Adverse Events (version 3.0). Laboratory testing to assess safety included hematologic, serum chemical, and urine chemical tests.

STATISTICAL ANALYSIS

A planned interim analysis was conducted with the use of the O'Brien–Fleming stopping boundary when 40 patients had completed the 12-month visit. A significance level of 0.002 was chosen to preserve a nominal significance level of 0.049 for efficacy at the end of the study. The interim analysis did not occur until late in the study, owing to regulatory and contracting hindrances that delayed the opening of some sites and prolonged the enrollment period. Although the interim stopping rule met the threshold for early termination, the data and safety monitoring board recommended that the trial be continued until all the patients had completed the 12-month visit. The investigators later learned that this action was taken to ensure that a full complement of efficacy and safety data would be available for the primary analysis in the event that the effect size was small. The data and safety monitoring board also endorsed an investigator-initiated proposal to truncate the observation phase of the study, owing to the impending termination of the funding period. The treatment assignments and the deliberations of the data and safety monitoring board remained concealed until the release of the final analysis.

The analyses were performed according to the intention-to-treat principle. The primary outcome, the FEV_1 response measured in milliliters per month over the course of 1 year (termed the FEV_1 slope), was analyzed as the difference in the FEV_1 slope between the placebo group and the sirolimus group. This was calculated with the use of spirometric data obtained at baseline and at 3, 6, 9, and 12 months during the treatment phase. A linear mixed-effects model was used to evaluate the between-group and within-group differences in the FEV_1 slope. The model included the time since enrollment, the treatment assignment, and the interaction between time and treatment. The PROC MIXED procedure with the Kenward–Roger correction (SAS Institute) was used to fit the model, without imputation of missing data. A general linear model was used to compare the difference between the two groups in the mean change from baseline to 12 months, after adjustment for baseline values. A Wilcoxon signed-rank test was used to assess the difference from baseline to 12 months within each group. For categorical outcomes, the data were compared with the use of Fisher's exact test or the chi-square test, as appropriate. For continuous variables, the medians were compared with the use of the Wilcoxon rank-sum test. P values of less than 0.05 were considered to indicate statistical significance. All reported P values are two-sided, and have not been adjusted for multiple testing. All analyses were performed with the use of SAS software, version 9.2.

RESULTS

ENROLLMENT AND BASELINE CHARACTERISTICS OF THE PATIENTS

From December 2006 through September 2010, a total of 111 patients consented to participate in the study (Fig. 1). Of these, 89 patients were eligible for the study and underwent randomization — 43 to the placebo group and 46 to the sirolimus group. The baseline characteristics of the patients were similar in the two groups (Table 1, and Table A in the Supplementary Appendix). The patients who were enrolled in the study had moderately severe lung disease; the mean (±SD) FEV_1 was 47.7±14.4% of the predicted value in the placebo group and 49.3±13.3% of the predicted value in the sirolimus group (P=0.77). There was also evidence of airflow obstruction, gas trapping, and impaired gas exchange. For additional details regarding the baseline characteristics of the patients and the results of the trial see the Supplementary Appendix.

SERUM LEVELS OF SIROLIMUS AND ADHERENCE TO MEDICATION

The serum levels of sirolimus in the placebo group were below the detection limit throughout the study. Other than brief out-of-range excursions, the sirolimus levels in the active-treatment group were maintained between 5 and 15 ng per milliliter, except in the case of four patients in whom levels were intentionally kept below therapeutic levels for 3 months or more in order to control side effects.

PRIMARY ANALYSIS

In the placebo group, the FEV_1 slope from baseline to 12 months was −12±2 ml per month; the slope was significantly less than zero (P<0.001), a finding that was consistent with declining lung function (Table 2). The FEV_1 slope in the sirolimus group was 1±2 ml per month, which was not significantly different from zero; this was indicative of the stabilization of lung function during treatment (Fig. 2A). There was a significant difference between the two groups in the FEV_1 slope (P<0.001). The absolute difference in the mean change in FEV_1 during the treatment period, calculated as the difference between the mean change in the placebo group (−134±182 ml) and the mean change in the sirolimus group (19±124 ml), was 153 ml (P<0.001 for the between-group difference) (Fig. 2B). A total of 12% of the patients in the placebo group, as compared with 46% of the patients in the sirolimus group, had FEV_1 values at or above baseline values at the 12-month visit (P<0.001) (Fig. 2C).

Figure 1 (facing page). **Screening, Randomization, and Follow-up.**

The treatment period was 12 months in duration and was followed by a 12-month observation period during which no patients received a study drug and all patients remained unaware of their treatment assignment. Spirometry was performed every 3 months during the treatment year and every 6 months during the observation year. Patients who were unable to undergo pulmonary-function (PF) testing at one visit could undergo testing at a subsequent visit. The reasons for withdrawal of patients from the study included a decision to use sirolimus outside the study (3 patients in the placebo group and 5 in the sirolimus group), pneumothorax (2 patients in the placebo group), infection (3 patients in each group), placement on a list for transplantation (2 patients in the placebo group), nonadherence to visits or testing (2 patients in the sirolimus group), rash (1 patient in the sirolimus group), anxiety (1 patient in the sirolimus group), and death (2 deaths in the placebo group, 1 due to stroke and 1 in a house fire). FEV_1 denotes forced expiratory volume in 1 second and LAM lymphangioleiomyomatosis.

SECONDARY ANALYSES

The FVC slope during the treatment phase was −11±3 ml per month in the placebo group, as compared with 8±3 ml per month in the sirolimus group (P<0.001) (Table 2). The FVC slope was significantly less than zero in the placebo group (P=0.001), which was consistent with a decline in lung function, and the slope was significantly greater than zero in the sirolimus group (P=0.009), which was consistent with an improvement in lung function during treatment. The absolute difference in the mean change in FVC during the treatment phase, calculated as the difference between the mean change in the placebo group (−129±233 ml) and the mean change in the sirolimus group (97±260 ml), was 226 ml (P=0.001 for the between-group difference) (Fig. 2B). A total of 23% of the patients in the placebo group, as compared with 54% of patients in the sirolimus group, had FVC values that were at or above baseline values at the 12-month visit (P<0.001). The between-group difference in the slope for functional residual capacity during

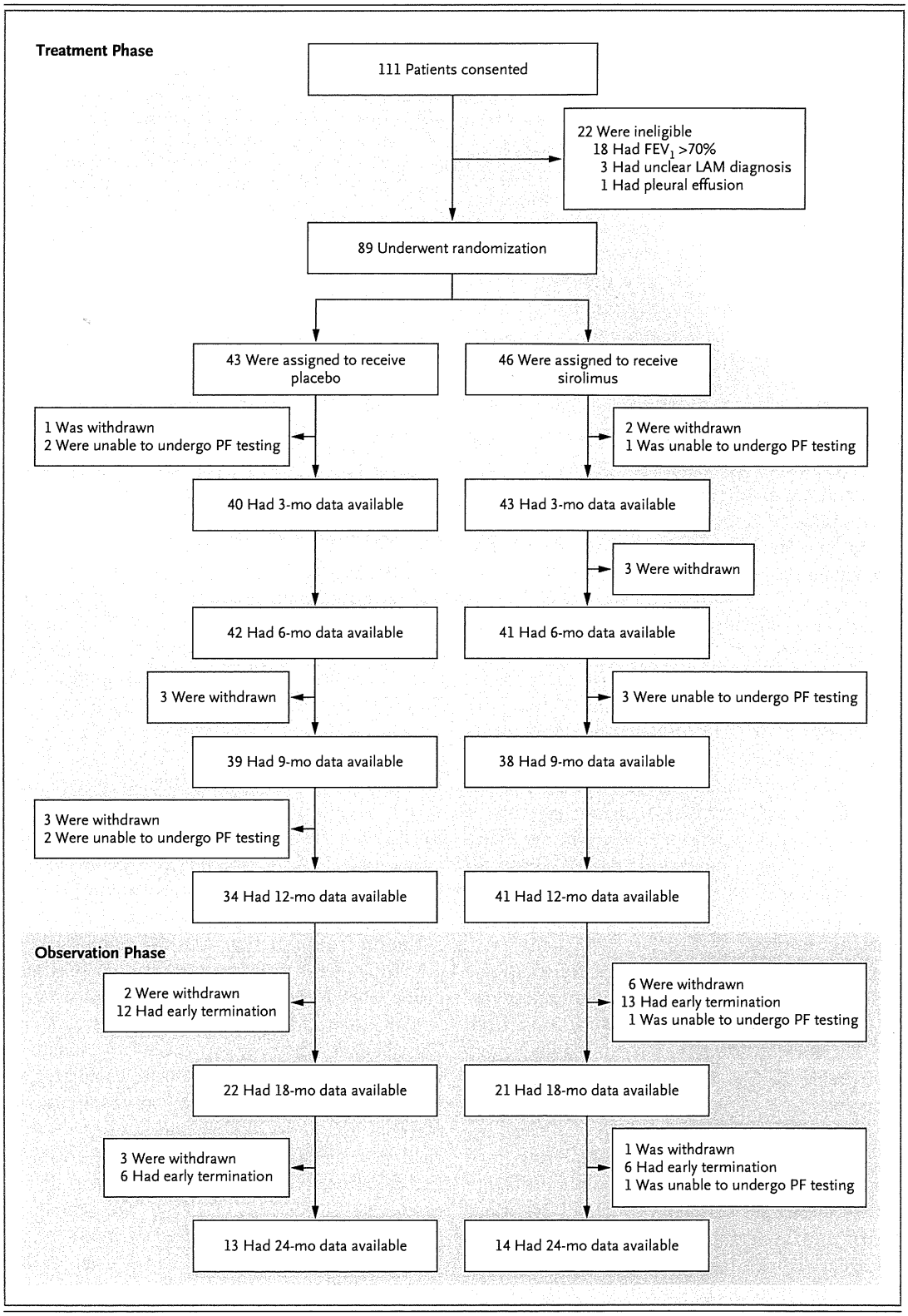

Table 1. **Baseline Demographic and Clinical Characteristics of the Patients.***

Characteristic	All Patients (N=89)	Placebo Group (N=43)	Sirolimus Group (N=46)	P Value
Age — yr	45.4±10.6	45.9±10.3	45.0±10.9	0.74
Race — no. (%)†				
White	59 (66)	30 (70)	29 (63)	0.58‡
Asian	27 (30)	12 (28)	15 (33)	
Other	3 (3)	1 (2)	2 (4)	
Clinical features — no. (%)				
Tuberous sclerosis complex	8 (9)	4 (9)	4 (9)	1.00§
Postmenopause	30 (34)	16 (37)	14 (30)	0.50‡
History of angiomyolipoma	44 (49)	22 (51)	22 (48)	0.75‡
History of pneumothorax	53 (60)	29 (67)	24 (52)	0.14‡
Oxygen-therapy requirement				
Continuous use	28 (31)	14 (33)	14 (30)	0.83‡
Intermittent use	52 (58)	23 (53)	29 (63)	0.36‡
Pulmonary-function testing				
FEV_1				
Volume — ml	1367±420	1378±446	1357±400	0.69¶
% of predicted value	48.54±13.77	47.73±14.37	49.29±13.31	0.77¶
FVC				
Volume — ml	2791±692	2909±749	2682±622	0.14¶
% of predicted value	79.71±16.60	80.77±17.62	78.73±15.70	0.55¶
Ratio of FEV_1 to FVC	0.50±0.15	0.48±0.15	0.52±0.16	0.35¶
Total lung capacity — % of predicted value	105.21±25.63	106.70±29.45	103.83±21.71	0.61¶
Functional residual capacity				
Volume — ml	3000±905	3175±1059	2838±710	0.20¶
% of predicted value	112.49±31.32	116.61±38.29	108.67±22.97	0.43
Residual volume — % of predicted value	141.42±59.22	147.48±69.25	135.78±48.15	0.80¶
D_{LCO}				
Diffusing capacity — ml/mm Hg/min	10.23±4.61	10.42±4.82	10.05±4.47	0.52¶
% of predicted value	43.43±18.97	43.77±20.56	43.12±17.66	0.70¶
6-Minute walk distance — m	403±105	399±115	407±96	0.78¶
Health-related symptom scores				
EuroQOL visual-analogue scale for quality of life‖	67.82±19.25	67.09±20.11	68.5±18.61	0.83¶
Functional Performance Inventory**	2.29±0.50	2.35±0.49	2.25±0.51	0.35¶
Serum VEGF-D concentration — pg/ml	2029±2343	2223±2997	1848±1514	0.57¶

* Plus–minus values are means ±SD. D_{LCO} denotes diffusing capacity for carbon monoxide, FEV_1 forced expiratory volume in 1 second, FVC forced vital capacity, and VEGF-D vascular endothelial growth factor D.
† Race was self-reported.
‡ P value was calculated with the use of the chi-square test.
§ P value was calculated with the use of Fisher's exact test.
¶ P value was calculated with the use of the Wilcoxon rank-sum test.
‖ The EuroQOL visual-analogue scale measures self-reported ratings of health status. Scores range from 0 to 100, with lower scores indicating worse functioning.
** Scores on the Functional Performance Inventory range from 1 to 4, with lower scores indicating lower health status.

Table 2. **Effects of Sirolimus on Primary and Selected Secondary Outcome Variables during the Treatment Period.***

Variable	Value at 12 Months		Change from Baseline			Rate of Change per Month		
	Placebo (N=34)	Sirolimus (N=41)	Placebo (N=34)	Sirolimus (N=41)	P Value†	Placebo (N=43)	Sirolimus (N=46)	P Value‡
Pulmonary function								
FEV_1 (ml)	1272±414	1383±394	−134±182§	19±124	<0.001	−12±2¶	1±2	<0.001
FVC (ml)	2843±668	2780±735	−129±233§	97±260	0.001	−11±3¶	8±3¶	<0.001
Total lung capacity (ml)	5464±1217	4944±982	−7±650	94±504	0.65	−2±7	8±7	0.34
Residual volume (ml)	2502±969	2112±617	−16 ±514	38±538	0.61	−3±7	4±7	0.46
Functional residual capacity (ml)	3260±968	2912±660	−123±521	53±335	0.43	−11±6	6±6	0.049
DL_{CO} (ml/mm Hg/min)	9.61±4.06	9.62±3.92	−0.62±2.89§	−0.06±1.50	0.38	−0.06±0.03¶	−0.01±0.02	0.17
6-Minute walk distance (m)	418±107	431±104	26±51§	24±59§	0.99	1.47±0.87	1.65±0.81¶	0.88
Score on EuroQOL visual-analogue scale for quality of life‖	65.60±18.47**	73.71±18.03	−2.34±15.77	6.10±16.96	0.02	−0.21±0.20	0.39±0.19¶	0.03
Total score on Functional Performance Inventory††	2.33±0.47	2.35±0.49	−0.05±0.24	0.10±0.38	0.08	−0.009±0.004¶	0.005±0.004	0.03
Serum VEGF-D (pg/ml)	2444±3862**	862±540	−14.81±1113	−1032±1301§	0.001	−2.42±17.23	−88.01±16.61¶	0.001

* Plus–minus values are means ±SD. DL_{CO} denotes diffusing capacity for carbon monoxide, FEV_1 forced expiratory volume in 1 second, FVC forced vital capacity, and VEGF-D vascular endothelial growth factor D.

† Two-sided P values for the comparison between the placebo and sirolimus groups of the mean change from baseline were calculated with the use of a general linear model, with adjustment for baseline levels of variables.

‡ Two-sided P values for the comparison between the placebo and sirolimus groups of the rate of change per month were calculated with the use of a linear mixed-effects model.

‡§ The two-sided P value for the change from baseline within the placebo or sirolimus group, as calculated with the use of the Wilcoxon signed-rank test, was less than 0.05 for FVC and 6-minute walk distance, less than 0.01 for DL_{CO}, and less than 0.001 for FEV_1 and VEGF-D level.

¶ The two-sided P value for the rate of change per month within the placebo or sirolimus group, as calculated with the use of a linear mixed-effects model, was less than 0.05 for the total score of the Functional Performance Inventory, the DL_{CO}, and the EuroQOL visual-analogue scale, less than 0.01 for the FVC, and less than 0.001 for VEGF-D level and FEV_1.

‖ The EuroQOL visual-analogue scale measures self-reported ratings of health status. Scores range from 0 to 100, with lower scores indicating worse functioning.

** The two-sided P value for the difference between the placebo and sirolimus groups at 12 months was less than 0.05, as calculated with the use of the Wilcoxon rank-sum test.

†† Scores on the Functional Performance Inventory range from 1 to 4, with lower scores indicating lower health status.

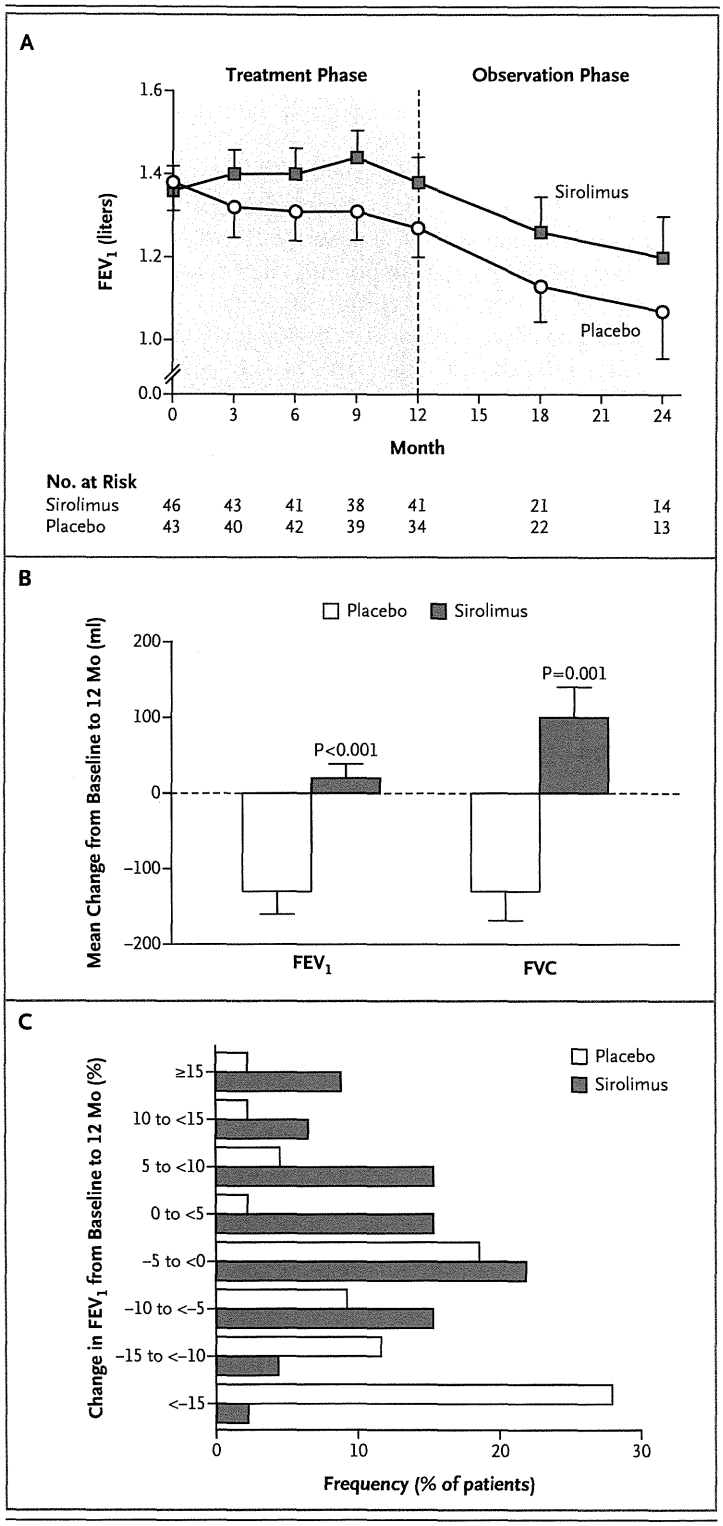

Figure 2. **Change in Lung Function during the Treatment and Observation Phases of the Trial.**

Panel A shows the mean forced expiratory volume in 1 second (FEV_1) at baseline and at each follow-up visit in the placebo and sirolimus groups. The number of patients for whom FEV_1 data were available at each time point is also shown. Panel B shows the mean changes from baseline to 12 months in FEV_1 and forced vital capacity (FVC) in the 34 patients in the placebo group and the 41 patients in the sirolimus group for whom 12-month data were available. Panel C shows the frequency of FEV_1 changes, in increments or decrements of 5% of the baseline value, from baseline to 12 months. The percentage of patients who had any improvement in FEV_1 was significantly greater in the sirolimus group than in the placebo group (46% vs. 12%, P<0.001). Conversely, a significantly greater percentage of patients in the placebo group than in the sirolimus group had some worsening of FEV_1 (67% vs. 44%). In Panels A and B, T bars indicate standard errors.

the treatment phase was also significant (P=0.049), but the differences in the slopes for total lung capacity, residual volume, diffusing capacity for carbon monoxide, and distance covered on a 6-minute walk test were not significant (Table 2).

There were significant differences favoring sirolimus in the change from baseline to 12 months in the score on the EuroQOL visual-analogue scale for quality of life and in the total score on the Functional Performance Inventory. The changes in other measures of health-related symptoms did not differ significantly between the two groups (Table 2). Mean VEGF-D levels were similar in the two groups at baseline but were significantly lower in the sirolimus group than in the placebo group at 6 and 12 months (Tables 1 and 2).

ANALYSES OF DATA FROM THE OBSERVATION YEAR

FEV_1 declined in both groups during the observation year (a decline of 8±2 ml per month in the placebo group and of 14±3 ml per month in the sirolimus group) (Fig. 2A). Although these slopes were both less than zero (P=0.005 and P<0.001, respectively), the difference between them did not reach significance (P=0.08). The mean change in FEV_1 from baseline to 24 months did not differ significantly between the two groups (−180±100 ml in the placebo group and −150±170 ml in the sirolimus group). Similarly, with respect to FVC, there were no significant between-group differences in the observation-year slopes or the mean changes from baseline to 24 months. The mean serum VEGF-D levels at 24 months remained elevated in the placebo group (2107±2146 pg per milliliter in the 13 patients for whom data were

available at 24 months) and depressed in the sirolimus group (930±461 pg per milliliter in the 14 patients for whom data were available at 24 months). There were no significant differences in the slopes from 12 to 24 months or in the mean change from baseline to 24 months in any other variables measured, including lung volumes, diffusing capacity for carbon monoxide, distance covered on a 6-minute walk test, and symptoms.

ADVERSE EVENTS

The most common adverse events during the treatment period were mucositis, diarrhea, nausea, hypercholesterolemia, acneiform rash, and swelling in the lower extremities (Table 3). The excess adverse events in the sirolimus group occurred mainly in eight categories: blood or bone marrow events, gastrointestinal events, dermatologic problems (1.9 events per patient in the placebo group vs. 3.0 events per patient in the sirolimus group, P<0.001), metabolic disturbances or abnormal laboratory results (2.2 events per patient in the placebo group vs. 2.8 events per patient in the sirolimus group, P=0.04), musculoskeletal or soft-tissue events, pain, neurologic events, and ocular or visual problems. Serious adverse cardiac events occurred only in the sirolimus group and included pericarditis, atrial arrhythmia (2 events in 1 patient), and tachycardia and fluid overload after embolization of an angiomyolipoma (2 events in 1 patient). Serious adverse pulmonary or upper respiratory events occurred only during the treatment period and were reported more frequently among patients receiving placebo than among those receiving sirolimus (P<0.001 by Fisher's exact test). During the observation year, considerably fewer adverse events (both overall and per patient) occurred in both groups, but serious adverse events occurred more frequently in the placebo group than in the sirolimus group.

DISCUSSION

Our study shows that treatment with sirolimus for 1 year has beneficial effects in patients with LAM, including the stabilization of FEV_1 and improvement in FVC, quality of life, and some functional performance measures. No effects were observed on the diffusing capacity of the lung for carbon monoxide or on exercise tolerance, and the positive effects on airflow waned after sirolimus was discontinued. Sirolimus was associated with an increased frequency of adverse events, as compared with placebo, though the rates of serious adverse events were similar in the two study groups.

Although the minimum clinically important differences for measures of lung function have not been established in patients with LAM, the minimum clinically important difference in FEV_1 in patients with chronic obstructive lung disease has been estimated to be 100 to 140 ml,[22] which is a change that patients can perceive, that is a typical response after use of a bronchodilator, and that predicts a relapse after an exacerbation.[22] The absolute mean between-group change of 153 ml in the FEV_1 over the course of 1 year in our study compares favorably with this estimate. In the placebo group, the observed annual mean decline of 134 ml per year in the FEV_1 from the baseline level of 1.38 liters indicates that lung function is lost at the rate of almost 10% per year in patients with moderately severe LAM. Therapies that stabilize lung function could potentially delay the need for lung transplantation, with its associated risks. The importance of the observed changes in lung function is further supported by the positive correlation with the scores on the Functional Performance Inventory and the EuroQOL visual-analogue scale for quality of life, both of which have been validated in patients with other lung diseases.[23,24]

Sirolimus therapy positively affected lung function only during the treatment period. After discontinuation of sirolimus, the FEV_1 in patients in the sirolimus group declined in parallel with the decline in the placebo group, and the mean change from baseline to 24 months did not differ significantly between the two groups. Thus, sirolimus therapy for 1 year did not appear to accelerate the subsequent rate of decline in lung function — a theoretical concern raised by the apparent rebound increase in growth rates of angiomyolipomas after withdrawal of sirolimus.[18] These data should be interpreted with caution, given the high withdrawal rate in the observation period and the early termination of the second trial year for some patients. Other study limitations include the possibility that the treatment assignments may have been inadvertently revealed owing to cholesterol elevations and the development of mouth ulcers and rashes in some patients in the sirolimus group.

Table 3. **Total Adverse Events and Serious Adverse Events in the Two Study Groups.***

Category	Total Adverse Events				Serious Adverse Events			
	Treatment Period		Observation Period		Treatment Period		Observation Period	
	Placebo	*Sirolimus*	*Placebo*	*Sirolimus*	*Placebo*	*Sirolimus*	*Placebo*	*Sirolimus*
	number of events							
Allergy or immunologic event	13	11	1	1				
Auditory or ear-related event	2	4	0	1				
Blood or bone-marrow event	4	12	1	0	0	1	0	0
Cardiac arrhythmia	0	2	0	2				
Cardiac event, general	12	15	3	2	0	5	0	0
Constitutional symptom	35	46	7	7				
Death not related to an event	0	0	1	0	0	0	1	0
Dermatologic event	41	106	8	11				
Endocrinologic event	3	2	2	0	0	0	1	0
Gastrointestinal event	181	275	12	20	1	3	0	0
Hemorrhage or bleeding event	14	17	3	1	0	0	1†	0
Hepatobiliary or pancreatic event	1	0	1	0	0	0	1	0
Infection	74	78	20	24	3	2	1	0
Lymphatic event	8	15	6	0				
Metabolic event or abnormal laboratory result	26	56	1	6	0	1	0	0
Musculoskeletal or soft-tissue event	21	35	2	4	0	1	0	1
Neurologic event	27	33	4	7				
Ocular or visual problem	3	8	1	2				
Pain	115	130	9	13	1	7	0	0
Pulmonary or upper respiratory event	121	97	17	32	13	2	0	0
Renal or genitourinary event	8	11	0	2				
Sexual or reproductive problem	8	5	0	0				
Vascular event	1	1	0	0	0	1	0	0
Total adverse events	718	959	99	135				
Total serious adverse events					18	23	5	1
Definitely not related to study drug					1	11	2	1
Probably not related to study drug					8	7	0	0
Possibly related to study drug					5	3	3	0
Probably related to study drug					4	2	0	0

* Adverse events were assessed with the use of the Common Terminology Criteria for Adverse Events (version 3.0). Details of the statistical analysis plan for adverse events are provided in the Supplementary Appendix.

† A level 5 serious adverse event due to intracranial hemorrhage resulted in the death of one patient in the 18th month after the beginning of the study (the 6th month of the observation period).

Sirolimus was associated with an improvement in FVC; the absolute mean change in FVC (230 ml) was similar in magnitude to the minimum clinically important difference of 250 ml proposed for scleroderma-related lung disease.[25] The increase in FVC with sirolimus was accompanied by an increase in the slope for functional residual capacity and trends toward increases in the slopes for total lung capacity and residual volume, suggesting that a reduction in restrictive impairment is a potential mechanism for higher airflow. The reduction in residual volume noted

in a previous open-label trial, which suggested a reduction in gas trapping, was not seen in our trial.[18]

Serious adverse respiratory events occurred less frequently in the sirolimus group than in the placebo group during the treatment period, a finding that is consistent with a potential beneficial effect of the drug on other manifestations of the disease. The between-group difference of 8.44 from baseline to 12 months in the quality of life scores on the EuroQOL visual-analogue scale (on which scores range from 0 to 100, with lower scores indicating worse functioning) was greater than half the standard deviation, an accepted threshold for clinical significance.[26] The lack of a significant between-group difference in the distance covered on a 6-minute walk test suggests that improvement in lung function may not be accompanied by an increase in exercise capacity, though a treatment effect might have been obscured by the relatively high baseline exercise tolerance of the patients or limitations in the performance characteristics of the test.

Levels of serum VEGF-D, a lymphangiogenic growth factor implicated in the pathophysiology of LAM,[11,12] were reduced in response to sirolimus. The persistent depression in mean VEGF-D levels after discontinuation of the drug may be consistent with a durable treatment effect in some patients, but this finding is difficult to interpret, given the attrition that occurred during the observation year. Further study is needed to determine whether it is possible to predict which patients will benefit from sirolimus treatment and whether the VEGF-D level can serve as a biomarker of disease severity, disease progression, or treatment response.

These results indicate that sirolimus may be useful in treating patients with moderately severe LAM-related lung disease. LAM typically progresses slowly, and given the risks of sirolimus therapy, treatment decisions should be made on an individual basis. The mean FEV_1 for patients in this trial (48% of the predicted value) was lower than the population-based mean among patients in the LAM Registry of the NIH[27] (70% of the predicted value), making it difficult to generalize the results from the patients in this study to patients with milder or more severe lung disease due to LAM. Since the stabilization of lung function appears to require continuous exposure to sirolimus, additional trials are needed to determine which patients will benefit from treatment and the optimal dose and duration of treatment. Given the side-effect profile of the drug during a 1-year period, future studies must carefully evaluate the long-term safety of sirolimus.

Supported by grants from the NIH Office of Rare Disease Research, administered by the National Center for Research Resources (RR019498, to Drs. Trapnell and McCormack; and RR019259, to Drs. Lee and Krischer), the Food and Drug Administration (FD003362, to Dr. McCormack), Canadian Institutes of Health Research (to Drs. Downey, Cohen, and Singer), Pfizer (formerly Wyeth) Pharmaceuticals (to Dr. McCormack), the Japanese Ministry of Health, Labor, and Welfare (H 19 Rinshoshiken 008, to Drs. Nakata and Inoue), the LAM Foundation (to Dr. McCormack), the Tuberous Sclerosis Alliance (Rothberg Courage Award, to Dr. McCormack), Cincinnati Children's Hospital Medical Center (Institutional Clinical and Translational Science Award 1UL1RR026314 01, to Drs. Young and McCormack, and Translational Research Initiative Award, to Drs. McCormack and Trapnell), Vi and John Adler, and the Adler Foundation. Drs. Moss and Taveira-DaSilva were supported by the Division of Intramural Research, National Heart, Lung, and Blood Institute. Pfizer provided the study drug and money for study visit costs.

Disclosure forms provided by the authors are available with the full text of this article at NEJM.org.

We thank John Bissler, M.D., for his assistance in providing the Cine MRI of a woman with tuberous sclerosis and lymphangioleiomyomatosis.

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