



Retroperitoneal fibrosis in the era of immunoglobulin G4-related disease

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Background: Retroperitoneal fibrosis (RPF) is a rare disease characterized by fibroinflammatory tissue in the periaortic or periliac retroperitoneum, where it frequently encases ureters. There is emerging evidence that a subset of this disease is part of a spectrum of multisystemic autoimmune diseases collectively referred to as “immunoglobulin G4 (IgG4)-related disease”.

Methods: We retrospectively analyzed 27 idiopathic RPF patients and identified a subset as IgG4-related RPF, which we categorized according to recently published comprehensive diagnostic criteria. We compared clinical and laboratory characteristics and response to treatment between the two groups.

Results: Of 27 total patients, 16 (59.3%) were diagnosed as having IgG4-related RPF, and these were predominantly male. They were also significantly older and more likely to have other organ involvement, hydronephrosis, and postrenal acute kidney injury (AKI) compared to those with idiopathic RPF. However, there was no difference in response rate to systemic steroid treatment.

Conclusion: IgG4-related RPF accounts for a substantial portion of RPF cases previously identified as “idiopathic RPF” in Korea. Clinical and laboratory characteristics of IgG4-related RPF are similar to those of idiopathic RPF except for a striking male predominance, older age, and higher incidence of postrenal AKI in IgG4-related RPF. More comprehensive, prospective studies are needed to clearly distinguish IgG4-related RPF from idiopathic RPF based on clinical manifestation and to further assess treatment response and long-term prognosis.

Keywords: Acute kidney injury, Immunoglobulin G4, Retroperitoneal fibrosis, Steroid treatment

Introduction

Retroperitoneal fibrosis (RPF) is an uncommon disorder characterized by the presence of fibroinflammatory tissue in the peri-aortic and peri-iliac retroperitoneum [1]. It frequently spreads into adjacent structures and causes ureteral obstruction with postrenal acute kidney injury (AKI) as one of the most common complications [1,2]. RPF has long been considered to be idiopathic in a majority of cases, except some cases resulting from malignancy, autoimmune diseases, drugs, radiation therapy, aortic aneurysm, or certain infections [3]. However, emerging evidence suggests that many cases of idiopathic RPF are a clinical manifestation of a multisys-

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temic autoimmune disease called “IgG4-related disease (IgG4-RD)” [4]. IgG4-RD is an increasingly recognized lymphoproliferative disorder characterized by increased serum IgG4 concentration, pathologic tissue infiltration of IgG4-positive plasma cells, and “storiform” fibrosis in affected organs [5]. Since the first published description of patients with sclerosing pancreatitis in 2001 [6], various systemic disorders affecting one or multiple organs have been attributed to IgG4-RD. An elevated serum IgG4 concentration is found in a majority of these patients, but approximately 30% of patients have normal serum IgG4 concentration [7]. Therefore, a definitive diagnosis of IgG4-RD can only be made based on histologic assessment of biopsies.

Glucocorticoids are often initiated to treat idiopathic RPF, but there is no universally accepted treatment regimen. For cases of ureteral obstruction, nephrostomy or stenting is required to relieve the obstruction and preserve kidney function [8].

The purpose of this study was to assess the prevalence of IgG4-related RPF among patients diagnosed with idiopathic RPF and to compare clinical and laboratory features of IgG4-related RPF with those of idiopathic RPF. We also compared the severity of disease, the initial response to glucocorticoid therapy, and relapse after discontinuation of therapy between the two groups.

Methods

Patients and study design

This was a retrospective, two-center study of 52 patients diagnosed with RPF between January 2006 and July 2017 at Korea University Anam Hospital and Korea University Kuro Hospital. The Institutional Review Board of each hospital approved this study (2017AN0366, 2018GR0026).

Diagnosis of RPF was based on radiologist reports of computed tomography (CT) or magnetic resonance imaging (MRI). Among 52 patients with RPF in our cohort, we identified those in whom the serum IgG4 concentration was measured or a biopsy was performed at the time of diagnosis. Patients with no serum IgG4 evaluation at the time of diagnosis were excluded. Patients with RPF due to secondary causes such as malignancy, drugs, radiation therapy, aortic aneurysm, or certain infections were also excluded.

We divided the patients into two groups, IgG4-related RPF and idiopathic RPF, based on the comprehensive diagnostic criteria published by Umehara et al [9] in 2011. According to this criteria, presence of a retroperitoneal mass, elevated IgG4 concentration (≥ 135 mg/dL), and representative tissue pathology (marked lymphocyte/plasmacyte infiltration and fibrosis, $\geq 40\%$ ratio of IgG4/IgG-positive cells, and ≥ 10 IgG4-positive plasma cells/high powered field [HPF]) received a “definite” diagnosis of IgG4-related RPF. The presence of a mass with either pathologic findings or elevated serum IgG4 concentration suggested “probable” or “possible” IgG4-related RPF, respectively.

We reviewed patients’ electronic hospital records to evaluate characteristics of age, sex, body mass index (BMI), and associated comorbidities (hypertension, diabetes mellitus, and hydronephrosis). The following laboratory data were also recorded: erythrocyte sedimentation rate (ESR), C-reactive protein (CRP), white blood cell count, eosinophil count, BUN, creatinine, C3, C4, and urine analysis. AKI was diagnosed and classified based on Kidney Disease: Improving Global Outcomes (KDIGO) clinical practice guidelines for AKI (i.e., ≥ 0.3 mg/dL or $\geq 50\%$ increase in serum creatinine level from baseline within 48 hours or urine output < 0.5 mL/kg/hour over 6 hours) [10].

The extent of organ involvement was estimated by imaging modalities of CT, MRI, or positron emission tomography CT. In patients with periaortic lesions, we measured the thickness and longitudinal length of the periaortic cuff. Maximal periaortic mass thickness, not including the aortic wall, was measured using a CT axial scan. The treatment modalities, radiologic response rate, and recurrence rate were also evaluated. Because there is no universally accepted definition of remission or recurrence in IgG4-related RPF [11], we defined remission as reduction and recurrence of size increase of the retroperitoneal mass on imaging modalities.

Statistical analysis

SPSS software, version 24 (IBM Corp., Armonk, NY, USA) was used for statistical analyses. Data summaries for quantitative data are shown as the mean \pm standard deviation or the median and range (minimum–maximum). In contrast, categorical data summaries are

expressed as frequencies and percentages. Comparisons between the two groups were performed using Student's *t* test or Mann–Whitney *U* test for quantitative data and the chi-square test or Fisher exact test for categorical data.

Results

Clinical features of the overall cohort

Out of 52 patients with RPF, 18 with no serum IgG4 evaluation at the time of diagnosis and 7 with secondary causes (5 malignancy, 1 radiation therapy, and 1 infection) were excluded. Twenty-seven patients were finally included in the analysis. The mean age was 59.7 ± 13.6 years, and 62% (17 patients) were male. Aorta involvement occurred in 23 cases (85.2%), 20 (87.0%) of which were detected around the infrarenal portion of the abdominal aorta and 17 of which simultaneously affected the iliac arteries. Hydronephrosis was present in 16 cases (59.3%), 14 of which were initially diagnosed as postrenal AKI. Percutaneous nephrostomy or double J ureteral catheter was transiently inserted in 12 patients.

Clinical and laboratory features

According to comprehensive diagnostic criteria [9], 16 of 27 RPF patients (59.3%) were classified as having IgG4-related RPF based on the biopsy findings and/or elevated serum IgG4 concentration (≥ 135 mg/dL) at the time of diagnosis (3 definite, 3 probable, 10 possible), and 11 patients were classified as having idiopathic RPF. The median serum IgG4 levels of the two groups were 264.8 mg/dL (range, 21–3,660 mg/dL) and 30.5 mg/dL (range, 14.0–97.3 mg/dL), and serum IgG4/IgG ratio was

0.194 and 0.024 for IgG4-related RPF and idiopathic RPF, respectively. A biopsy procedure was performed in nine patients, and all specimens were collected from retroperitoneal mass by surgical procedure. Six of nine (66.7%) cases showed representative biopsy findings (marked lymphocyte/plasmacyte infiltration and fibrosis, $> 40\%$ ratio of IgG4/IgG-positive cells, and > 10 IgG4-positive plasma cells/HPF) (Fig. 1) and were classified as IgG4-related RPF, and the three patients were classified as idiopathic RPF.

The clinical features of both groups are summarized in Table 1. The IgG4-related RPF group was significantly older (66.1 ± 10.48 years vs. 50.5 ± 12.69 years, $P = 0.002$) and contained significantly more males (81% vs. 36%, $P = 0.040$) compared to the idiopathic RPF group. However, there was no statistically significant difference in BMI or underlying comorbid conditions such as hypertension, diabetes mellitus, or heart failure.

Although 8 of 16 patients (50.0%) with IgG4-related RPF had other organ involvements, with the pancreas being the most commonly affected organ, there were no patients in the idiopathic RPF group with other organ involvement ($P = 0.008$). However, there was no significant difference in incidence of aorta involvement, thickness or length of paraaortic cuff, or location of mass.

Hydronephrosis was more frequently found in IgG4-related RPF (75.0% vs. 36.4%, $P = 0.045$), and the incidence of postrenal AKI was also significantly higher in this group (68.8% vs. 27.3%, $P = 0.034$) compared to the idiopathic RPF group. The median creatinine level was 1.37 mg/dL (range, 0.69–5.42) and 0.77 mg/dL (range, 0.52–7.18) in IgG4-related RPF and idiopathic RPF, respectively. Percutaneous nephrostomy or double J catheter was inserted in 9 of 16 patients with IgG4-related RPF and 3 of 11 patients with idiopathic RPF. All patients recovered

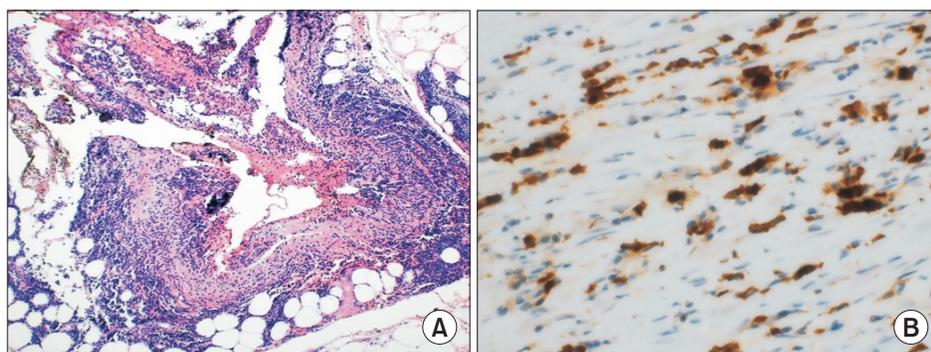


Figure 1. Histology of immunoglobulin G4 (IgG4)-related retroperitoneal fibrosis. (A) Low power view (40 \times) of hematoxylin & eosin staining of laparoscopically removed retroperitoneal tissue shows massive infiltration of lymphocytes. (B) High power view (200 \times) shows IgG4-positive plasma cells exceeding 10/high powered field.

Table 1. Clinical characteristics of patients with IgG4-related RPF and idiopathic RPF

Characteristic	IgG4-related RPF (n = 16)	Idiopathic RPF (n = 11)	P value
Age (yr)	66.1 ± 10.5	50.5 ± 12.7	0.002*
Sex, male	13 (81.3)	4 (36.4)	0.040*
BMI (kg/m ²)	24.7 ± 2.9	24.9 ± 2.1	0.755
DM	3 (18.8)	2 (18.2)	1.000
HTN	7 (43.8)	4 (36.4)	1.000
AKI	11 (68.8)	3 (27.3)	0.034*
AKI stage 1	5	1	
AKI stage 2	0	0	
AKI stage 3	6	2	
Hydronephrosis	12 (75.0)	4 (36.4)	0.045*
Thickness of paraaortic cuff (mm)	17.3 ± 6.5	16.0 ± 8.4	0.661
Length of paraaortic cuff (mm)	92.7 ± 48.8	64.8 ± 38.0	0.150
Aorta involvement	13 (81.3)	10 (90.9)	0.624
Iliac artery involvement	11 (68.8)	6 (54.5)	0.687
Other organ involvement	8 (50.0)	0 (0)	0.008*

Data are presented as mean ± standard deviation, number (%), or number only.

AKI, acute kidney injury; BMI, body mass index; DM, diabetes mellitus; HTN, hypertension; IgG, immunoglobulin G; RPF, retroperitoneal fibrosis.

**P* < 0.05.

Table 2. Laboratory characteristics of patients with IgG4-related RPF and idiopathic RPF

Characteristic	IgG4-related RPF (n = 16)	Idiopathic RPF (n = 11)	P value
Initial Hb (g/dL)	12.2 ± 1.7	12.3 ± 1.7	0.859
Platelets, ×1,000	214 (107–557)	286 (119–387)	0.084
WBC (/μL)	6,445 (3,400–14,740)	7,730 (4,060–13,000)	0.537
Eosinophil count (/μL)	272 (27–872)	121 (8.1–299)	0.034*
BUN (mg/dL)	20.2 (8.2–60.3)	15.0 (6.6–59.8)	0.199
Creatinine (mg/dL)	1.37 (0.69–5.42)	0.77 (0.52–7.18)	0.032*
ESR (mm/hr)	47.1 ± 23.520	48.0 ± 35.380	0.944
CRP (mg/L)	5.7 (0.34–152)	13.5 (1.72–109)	0.598
Albumin (g/dL)	3.95 (3.3–4.2)	4.1 (3.3–5.0)	0.342
Uric acid (mg/dL)	6.6 (3.2–10.8)	5.0 (3.5–12.9)	0.302
Total calcium (mg/dL)	8.90 ± 0.241	8.97 ± 0.561	0.823
P (mg/dL)	3.83 ± 1.290	4.10 ± 1.243	0.655
C3 (mg/dL)	105.1 ± 24.045	129.0 ± 42.569	0.228
C4 (mg/dL)	24.7 (9.5–35)	31.9 (18.2–76)	0.199
Serum IgG4 (mg/L)	264.8 (21–3,660)	30.5 (14–97.3)	0.000*
Serum IgG (mg/L)	1,560 (867–3,729)	1,391 (831–1,566)	0.120
Serum IgG4/IgG ratio	0.194 (0.096–1.649)	0.024 (0.011–0.112)	0.000*

Data are presented as mean ± standard deviation or median (range).

BUN, blood urea nitrogen; CRP, C-reactive protein; ESR, erythrocyte sedimentation rate; Hb, hemoglobin; IgG, immunoglobulin G; P, phosphorus; RPF, retroperitoneal fibrosis; WBC, white cell count.

**P* < 0.05.

renal function to baseline level, and no patients showed progressive CKD during a median follow-up period of 18 months.

Hemoglobin, platelet, ESR, and CRP levels, as well as white blood cell counts, were comparable between the two groups, with the exception of eosinophil count,

Table 3. Treatment response and prognosis of patients with IgG4-related RPF and idiopathic RPF

Parameter	IgG4-related RPF (n = 16)	Idiopathic RPF (n = 11)
Patients initially treated with steroid	13	5
Response +	13 (100)	5 (100)
Discontinuation	4	3
Relapse	4 (100)	0 (0)

Data are presented as number only or number (%).

IgG, immunoglobulin G; RPF, retroperitoneal fibrosis.

which was significantly increased in IgG4-related RPF (Table 2).

Treatment and prognosis

Treatment response and prognosis of patients in both groups are reported in Table 3. As shown, 13 of 16 patients (81.3%) with IgG4-related RPF and 5 of 11 patients (45.5%) with idiopathic RPF were initially treated with systemic steroid (0.5–1.0 mg/kg) or steroid/methotrexate. Median duration of treatment was 26 months, and all patients from both groups showed partial or complete radiological response (100%). There were 8 patients who underwent follow-up serum IgG4 level assessment after 1 year of systemic steroid treatment, demonstrating significantly decreased serum IgG4 concentration (264 [range, 53–577] mg/dL vs. 82 [range, 15–205] mg/dL, $P = 0.012$).

All four patients who discontinued systemic steroid treatment in the IgG4-related RPF group relapsed, with a median relapse time of 14 months, and required retreatment with systemic steroid. However, the three patients who discontinued steroid in the idiopathic RPF group maintained remission for up to 48 months.

Discussion

To the best of our knowledge, this is the first report in Korea demonstrating that a substantial portion of idiopathic RPF can be classified within the spectrum of IgG4-RD. RPF, previously called “Ormond’s disease,” is a rare clinicopathological condition characterized by fibroinflammatory tissue that surrounds the abdominal aorta and iliac arteries, frequently extending into the retroperitoneal space and encasing the ureters [1,2]. Clinical manifestations include nonspecific constitutional

symptoms such as fatigue, anorexia, and weight loss. Localized symptoms also occur and include renal colic or peripheral edema caused by compression of the ureters or lymphatics, respectively [1,8,12]. Until recently, most RPF cases have been considered idiopathic, except some cases resulting from secondary causes such as malignancies, radiation, or drugs [3]. However, emerging evidence suggests that a significant proportion of idiopathic RPF can be classified as part of the autoimmune disease IgG4-RD [4].

IgG4-RD is an increasingly recognized fibroinflammatory disease affecting multiple organs including the pancreas, aorta, lacrimal gland, and retroperitoneum [13]. Typical pathological findings include IgG4⁺ lymphoplasmacytic infiltration, storiform fibrosis, and obliterative phlebitis [13,14]. Definite diagnosis can be made in patients with 1) fibrous tissue in the periaortic or periiliac retroperitoneum; 2) plasma IgG4 level, ≥ 135 mg/dL; and 3) lymphoplasmacytic infiltration with more than 10 IgG4⁺ cells/HPF or IgG4/IgG ratio > 0.4 [9,15]. Although we used comprehensive diagnostic criteria including patients without tissue pathology [9], we found that IgG4-related RPF accounted for approximately 60% of the RPF cases examined.

In our study, patients with IgG4-related RPF were significantly older and showed greater male predominance compared to patients with idiopathic RPF. This is comparable to several other previously published studies [16–19], while Khosroshahi et al [4] reported similar male predominance between the two groups in a study in the United States of America. Similar to other reports [4,18,20], other organ involvement was much more frequent in IgG4-related RPF (50% vs. 0%), and the pancreas was the most commonly affected organ.

The prevalence of hydronephrosis in IgG4-related RPF has been estimated to be 33% to 66% [4,21]. Interestingly, 12 of 16 patients (75.0%) with IgG4-related RPF demonstrated hydronephrosis in our study, and 11 patients were diagnosed with postrenal AKI, 9 of whom needed temporary percutaneous nephrostomy or ureteral catheter placement. These complications were significantly more frequent than those observed in idiopathic RPF cases (hydronephrosis, 4 of 11; postrenal AKI, 3 of 11; relief of obstruction, 3 of 11), which contrasts with previous reports demonstrating a comparable frequency of hydronephrosis between the two groups [4,21]. Further study is needed

to identify the reasons underlying this discrepancy and should emphasize the volume, extent, or duration of the retroperitoneal mass within the two groups.

Systemic steroids were given as the initial treatment modality for both groups, and all patients in both groups showed a 100% response rate. No patients in our cohort experienced B cell depletion. Due to the absence of a widely accepted definition of remission or recurrence, we used a reduction or increase of retroperitoneal mass measured via imaging modalities to define them.

All four patients with IgG4-related RPF that discontinued systemic steroids relapsed, with a median relapse time of 14 months, and required maintenance immunosuppressive drugs. In contrast, all three patients in the idiopathic group who stopped systemic steroids maintained remission for at least 48 months without other immunosuppression. However, conclusions regarding prognosis and relapse cannot be made due to the small sample size in our study, and randomized, controlled studies enrolling a larger number of patients are needed to determine universally-accepted and objective response criteria for IgG4-related RPF.

In addition to the small sample size, that only 3 of 16 patients fulfilled the full diagnostic criteria of IgG4-related RPF and were categorized as “definite” is also a major limitation of this study. However, this report raises awareness of this rare systemic autoimmune disease and demonstrates that a substantial proportion of idiopathic RPF could be classified within the spectrum of rare systemic autoimmune diseases, presenting with different clinical and prognostic behaviors and requiring different treatment strategies than idiopathic RPF.

In conclusion, our two-center retrospective study demonstrated that a substantial proportion of idiopathic RPF can be classified within the spectrum of the multisystemic autoimmune disease “IgG4-RD,” suggesting that clinicians should consider this possibility and examine the presence of other organ involvement. More studies are necessary to better understand the pathophysiology, treatment response, and ultimate prognosis of this rare disease.

Conflicts of interest

All authors have no conflicts of interest to declare.

References

- [1] Vaglio A, Salvarani C, Buzio C. Retroperitoneal fibrosis. *Lancet* 2006;367:241-251.
- [2] Vaglio A, Maritati F. Idiopathic retroperitoneal fibrosis. *J Am Soc Nephrol* 2016;27:1880-1889.
- [3] Vaglio A, Palmisano A, Corradi D, Salvarani C, Buzio C. Retroperitoneal fibrosis: evolving concepts. *Rheum Dis Clin North Am* 2007;33:803-817, vi-vii.
- [4] Khosroshahi A, Carruthers MN, Stone JH, et al. Rethinking Ormond’s disease: “idiopathic” retroperitoneal fibrosis in the era of IgG4-related disease. *Medicine (Baltimore)* 2013;92:82-91.
- [5] Masaki Y, Kurose N, Umehara H. IgG4-related disease: a novel lymphoproliferative disorder discovered and established in Japan in the 21st century. *J Clin Exp Hematop* 2011;51:13-20.
- [6] Hamano H, Kawa S, Horiuchi A, et al. High serum IgG4 concentrations in patients with sclerosing pancreatitis. *N Engl J Med* 2001;344:732-738.
- [7] Sah RP, Chari ST. Serologic issues in IgG4-related systemic disease and autoimmune pancreatitis. *Curr Opin Rheumatol* 2011;23:108-113.
- [8] Pipitone N, Vaglio A, Salvarani C. Retroperitoneal fibrosis. *Best Pract Res Clin Rheumatol* 2012;26:439-448.
- [9] Umehara H, Okazaki K, Masaki Y, et al. Comprehensive diagnostic criteria for IgG4-related disease (IgG4-RD), 2011. *Mod Rheumatol* 2012;22:21-30.
- [10] Khwaja A. KDIGO clinical practice guidelines for acute kidney injury. *Nephron Clin Pract* 2012;120:c179-c184.
- [11] Cristian S, Cristian M, Cristian P, et al. Management of idiopathic retroperitoneal fibrosis from the urologist’s perspective. *Ther Adv Urol* 2015;7:85-99.
- [12] Kermani TA, Crowson CS, Achenbach SJ, Luthra HS. Idiopathic retroperitoneal fibrosis: a retrospective review of clinical presentation, treatment, and outcomes. *Mayo Clin Proc* 2011;86:297-303.
- [13] Stone JH, Zen Y, Deshpande V. IgG4-related disease. *N Engl J Med* 2012;366:539-551.
- [14] Deshpande V, Gupta R, Sainani N, et al. Subclassification of autoimmune pancreatitis: a histologic classification with clinical significance. *Am J Surg Pathol* 2011;35:26-35.
- [15] Umehara H, Okazaki K, Nakamura T, et al. Current approach to the diagnosis of IgG4-related disease - combination of comprehensive diagnostic and organ-specific criteria. *Mod Rheumatol* 2017;27:381-391.

- [16] Zen Y, Onodera M, Inoue D, et al. Retroperitoneal fibrosis: a clinicopathologic study with respect to immunoglobulin G4. *Am J Surg Pathol* 2009;33:1833-1839.
- [17] Uchida K, Masamune A, Shimosegawa T, Okazaki K. Prevalence of IgG4-related disease in Japan based on nationwide survey in 2009. *Int J Rheumatol* 2012;2012:358371.
- [18] Castelein T, Coudyzer W, Blockmans D. IgG4-related peri-aortitis vs idiopathic periaortitis: is there a role for atherosclerotic plaque in the pathogenesis of IgG4-related peri-aortitis? *Rheumatology (Oxford)* 2015;54:1250-1256.
- [19] Stone JR. Aortitis, periaortitis, and retroperitoneal fibrosis, as manifestations of IgG4-related systemic disease. *Curr Opin Rheumatol* 2011;23:88-94.
- [20] Yamashita K, Haga H, Mikami Y, et al. Degree of IgG4+ plasma cell infiltration in retroperitoneal fibrosis with or without multifocal fibrosclerosis. *Histopathology* 2008;52:404-409.
- [21] Koo BS, Koh YW, Hong S, et al. Clinicopathologic characteristics of IgG4-related retroperitoneal fibrosis among patients initially diagnosed as having idiopathic retroperitoneal fibrosis. *Mod Rheumatol* 2015;25:194-198.