(Table 3), it was desirable to perform a multivariate analysis to clarify whether both IgE and ulcerative colitis were independent factors for the development of biliary carcinoma. This analysis was impossible because there was no occurrence of biliary carcinoma in the high IgE group, which meant that the odds ratio or hazard ratio could not be calculated. However, considering no significant relation between high IgE level and ulcerative colitis ($P = .661$, χ^2 test with Yates' correction) (Table 1), IgE and ulcerative colitis seemed to be independent factors.

Despite the significantly different prevalence of biliary carcinoma between patients with and without increased IgE levels, there was no significant difference in the prevalence of end points including biliary carcinoma, liver transplantation, and death by other causes. This is because PSC with increased IgE level is still a progressive disease and may lead to eventual liver dysfunction. However, considering the different occurrence rates of biliary carcinoma, a significant difference might be found if our study populations were larger.

In summary, increased IgE level was related closely to older age at onset of PSC and reduced occurrence of biliary carcinoma. It has not yet been determined whether IgE is related directly to the pathogenesis of PSC or whether it increased reactively. In any event, it is certain that grouping PSC in terms of IgE should be attempted on a larger scale to validate our finding. Adequate grouping may contribute to the clarification of the pathogenesis of PSC, which remains poorly understood.

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Reprint requests

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Conflicts of interest

The authors disclose no conflicts.

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ORIGINAL ARTICLE

Clinical and histological changes associated with corticosteroid therapy in IgG4-related tubulointerstitial nephritis

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Abstract
Objectives This study aimed to investigate the clinicopathological changes induced by corticosteroid therapy in immunoglobulin (Ig)G4-related tubulointerstitial nephritis (TIN).
Methods We studied six IgG4-related TIN patients receiving renal biopsies before and after corticosteroid therapy. Their clinical data and histological findings were evaluated before and after therapy.
Results Elevated serum creatinine levels rapidly improved after corticosteroid therapy except for two patients, in whom it persisted. Abnormal radiological findings improved in all patients, although focal cortical atrophy persisted in three. Histologically, TIN-like dense lymphoplasmacytic infiltration, interstitial fibrosis, IgG4-positive plasma cell, CD4+CD25+ T cell, and Foxp3+ cell infiltration were characteristic before therapy. After therapy, the area with cell infiltration decreased and regional fibrosis became evident in the renal interstitium. The number of IgG4-positive plasma cells and Foxp3+ cells significantly diminished even in the early stage of therapy, whereas low to moderate numbers of CD4+ and CD8+ T cells still infiltrated where inflammation persisted in the later stage.
Conclusions Our study shows that persistent renal insufficiency associated with macroscopic atrophy and microscopic fibrosis is not so rare in IgG4-related TIN. Pathologically, the behavior of regulatory T cells during the clinical course is quite similar to that of IgG4-positive plasma cells, and the behavior pattern of those cells is distinctive.

Keywords IgG4-related disease · Tubulointerstitial nephritis · IgG4-positive plasma cell · Regulatory T cell · Corticosteroid therapy

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Introduction

Immunoglobulin (Ig)G4-related disease (IgG4-RD) is a recently recognized systemic inflammatory disease with multiorgan involvement [1–5], including the kidney. Since 2004, accumulated case reports and case series have defined the radiographic and histopathological characteristic findings of IgG4-related kidney disease [6–20]. Two large

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studies [21, 22] demonstrated clinicopathological features of IgG4-related tubulointerstitial nephritis (TIN) that included high levels of serum IgG4, IgG, and IgE; hypocomplementemia; and TIN with copious IgG4-positive plasma cell infiltration with fibrosis. However, the clinical course and histological changes occurring after corticosteroid therapy have not yet been well characterized. In addition, although IgG4-RD is thought to be steroid responsive, not all cases achieve recovery of normal renal function [23], with, for example, one case report describing a patient who required maintenance hemodialysis despite corticosteroid therapy [24]. Therefore, in order to establish the optimal treatment plan to prevent progressive kidney damage, renal re-biopsy seems to be necessary. We undertook this study to evaluate the influence of corticosteroid therapy on the clinical and histopathological findings in IgG4-related TIN.

Materials and methods

Patients and materials

Between 1 September 2005 and 31 August 2010, we identified 11 IgG4-RD patients with kidney involvement, including renal parenchymal and pelvic lesions. We diagnosed IgG4-RD according to the provisional diagnostic criteria for IgG4-RD of Masaki et al. [25]. Renal biopsies were performed in six patients in whom IgG4-related TIN was highly suspected because of renal dysfunction, elevation of renal tubular markers, or abnormal imaging finding, and we diagnosed them with IgG4-related TIN based on diagnostic criteria proposed by Kawano et al. [26] and Raissian et al. [22]. Two patients (patients 1 and 5 in Table 1) had been included in our earlier studies [27, 28], two (patient 1 and 6) in a report by Saeki et al. [21], and one (patient 1) in a report by Yamaguchi et al. [29].

All six patients underwent re-biopsy while receiving corticosteroid therapy, as re-evaluation of the extent of cell infiltration and fibrosis was necessary to assess the future dosage regimen of corticosteroid. One patient (patient 1 in Table 1) underwent re-biopsy 14 months after the start of therapy, one (patient 2) 7 months later, three (patients 3, 4, and 5) 4 months later, and one (patient 6) only 1 month later. These biopsy specimens were obtained randomly, not specifically from the mass lesions. We examined these 12 specimens histologically and immunohistochemically. With regard to the extent of the renal interstitial lesion, diffuse TIN was defined as being present when $\geq$80% of the renal interstitium in renal biopsy specimens was affected and focal TIN when <80% was affected. We also retrospectively evaluated the clinical and radiographic findings of these six patients. Serial laboratory data during the clinical course were analyzed, and the computed tomography (CT) findings of renal lesions were investigated before and after corticosteroid therapy in all patients. This study received institutional ethics board approval, and informed consent for all data and samples was obtained from each patient. The research was in compliance with the Declaration of Helsinki.

Single immunostaining

Bouin's fluid- or formalin-fixed and paraffin-embedded renal specimens of six patients with IgG4-related TIN were used for the immunostaining of IgG4, CD138, CD4, CD8, and Foxp3. The immunostaining was performed using a monoclonal antibody against human IgG4 (Zymed Laboratory, San Francisco, CA, USA), CD138 (AbD Serotec, Oxford, UK), CD4 (Nichirei, Tokyo, Japan), CD8 (Nichirei), and Foxp3 (AbD Serotec). The deparaffinized sections were microwaved in citrate buffer (pH 6.0) for 15 min. Cells positive for IgG4, CD138, CD4, CD8, or Foxp3 were counted in five different high-power fields (HPF: 10× eyepiece and 40× lens) with intense cell infiltration.

Dual fluorescent immunostaining of CD4 and CD25

All Bouin's fluid-fixed and paraffin-embedded renal specimens were used for dual fluorescent immunostaining of CD4 and CD25. The deparaffinized sections were microwaved in citrate buffer (pH 6.0) for 20 min and incubated with normal donkey serum for protein blocking for 30 min. The specimens were incubated with a mouse monoclonal antibody to CD25 (Leica Microsystems, Wetzlar, Germany) and a rabbit monoclonal antibody to CD4 (Spring Bioscience, CA, USA) overnight at 4°C. Then, the specimens were incubated for 1 h at room temperature with Alexa Fluor 488-labeled donkey anti-mouse IgG antibodies and Alexa Fluor 594-labeled donkey anti-rabbit IgG antibodies (Molecular Probes, Carlsbad, CA, USA) and observed under a laser microscope and digitally merged. No positive staining was observed when the primary antibodies were replaced with normal donkey serum in the negative control of the staining procedures.

Statistical analysis

Statistical analysis was performed using the Wilcoxon signed rank test for continuous non-normally distributed data. Significant differences were defined as $P < 0.05$.

Results

Patient profiles

We analyzed six patients [four men and two women; average age 71.0 (range 59–79) years] with IgG4-related

Table 1 Clinicopathological characteristics of six patients with immunoglobulin (Ig)G4-related tubulointerstitial nephritis

No.	Age/sex	Cr at pre-Tx (*N* 0.6–1)	IgG (*N* 870–1,600)	IgG4 (*N* < 105)	IgE (*N* < 250)	CH50 (*N* 32–47)	C3 (*N* 65–135)	C4 (*N* 13–35)	ANA	RF	U-β2MG (*N* < 250)
1	59/M	1.15	2,850	1,470	456	34	81	16	(−)	(−)	335
2	79/M	0.54	4,756	409	457	8	41	3	(+)	(−)	496
3	77/W	0.59	2,256	984	292	60	110	27	(+)	(−)	78
4	68/M	1.90	3,830	736	242	3	33	1	(+)	(+)	7,375
5	75/W	2.55	3,695	486	1,226	2	18	2	(+)	(−)	35,490
6	68/M	7.26	4,661	1,120	335	5	10	7	(+)	(+)	3,240

No.	Age/sex	U-NAG (*N* < 10)	Eosinophilia	Allergy	U-Pr	CT findings	Initial renal biopsy findings	Initial Tx of post-biopsy	Cr at re-biopsy (*N* 0.6–1)	Period between initial biopsy and re-biopsy (months)
1	59/M	1.6	(+)	(−)	(−)	LDLs	fTIN	PSL 30	1.06	14
2	79/M	19	(−)	(−)	(−)	(−)	fTIN + EndPGN	PSL 20	0.51	7
3	77/W	0.4	(−)	Rhinitis	(−)	LDLs + P	fTIN	PSL 20	0.63	4
4	68/M	8.8	(−)	BA	(−)	LDLs	fTIN + IgAGN	PSL 30	1.20	4
5	75/W	5.9	(−)	(−)	(+)	S	dTIN + HSPN	PSL 30	1.05	4
6	68/M	16.9	(−)	(−)	(+)	S	dTIN + EndPGN	PSL 30	1.45	1

Conversion factor for Cr: mg/dl to μmol/l, ×88.4

ANA antinuclear antibody, *BA* bronchial asthma, *CH50* serum CH50 at initial renal biopsy (U/ml), *Cr* serum creatinine (mg/dl), *CT* computed tomography, *C3* serum C3 at initial renal biopsy (mg/dl), *C4* serum C4 at initial renal biopsy (mg/dl), *dTIN* diffuse tubulointerstitial nephritis, *EndPGN* endocapillary proliferative glomerulonephritis *fTIN* focal tubulointerstitial nephritis, *HSPN* Henoch–Schönlein purpura nephritis, *IgAGN* IgA nephropathy, *IgG* serum immunoglobulin G at initial renal biopsy (mg/dl), *IgG4* serum immunoglobulin G4 at initial renal biopsy (mg/dl), *IgE* serum immunoglobulin E at initial renal biopsy (IU/ml), *LDLs* multiple low-density lesions of the renal parenchyma, *P* diffuse thickening of the renal pelvic wall, *post-biopsy* post-renal biopsy, *PSL* prednisolone mg/day, *re-biopsy* renal re-biopsy, *RF* rheumatoid factor, *S* diffuse bilateral renal swelling, *Tx* treatment, *U-β2MG* urine β2-microglobulin at initial renal biopsy (μg/l), *U-NAG* urine *N*-acetyl-β-D-glucosaminidase at initial renal biopsy (U/l), *U-Pr* proteinuria

TIN. None had been treated with corticosteroid or any other immunosuppressants before the diagnosis. All patients had other organ involvement associated with IgG4-RD: four (66.7%) dacryoadenitis and/or sialoadenitis, six (100%) multiple lymphadenopathy, and three (50.0%) lung lesions. Pancreatic lesion was detected in only one patient (16.7%). Prostate, liver, and joint lesion was detected in one patient each. All patients were treated with prednisolone at an initial dose of 20–30 mg/day after the initial renal biopsy (Table 1).

Laboratory findings and changes associated with corticosteroid therapy

At presentation, all patients showed elevated levels of serum IgG4 (average 867.5 mg/dl, range 409–1,470 mg/dl, normal range <105 mg/dl) and IgG (average 3,675 mg/dl, range 2,256–4,756 mg/dl, normal range 870–1,700 mg/dl). Five patients (83.3%) showed elevated serum IgE levels (average 501.3 IU/ml, range 242–1,226 IU/ml, normal range <250 IU/ml), and only one patient had eosinophilia (eosinophils >5%). Four patients (66.7%) had hypocomplementemia. Antinuclear antibodies were positive in five patients (83.3%) and rheumatoid factor in two (33.3%). Serum creatinine concentration was elevated in four patients (66.7%). Urine β2-microglobulin (β2-MG) excretion was elevated in five patients (83.3%) and urine *N*-acetyl-β-D-glucosaminidase (NAG) level in two (33.3%), whereas two patients (33.3%) had proteinuria (Table 1). In three patients who showed severe renal dysfunction (serum creatinine >1.5 mg/dl) before corticosteroid therapy (patients 4, 5, and 6 in Table 1), serum creatinine rapidly declined within 1 month after the start of therapy. However, some degree of renal dysfunction persisted in two of them (Fig. 1a). Four patients had hypocomplementemia before corticosteroid therapy (patients 2, 4, 5, and 6 in Table 1), and corticosteroid therapy promptly improved serum complement levels within 2 months in three of them; recovery of serum complement level was very slow in one other (Fig. 1b). On the other hand, urine β2-MG and NAG concentrations fluctuated despite the corticosteroid therapy (data not shown).

Computed tomography findings and changes associated with corticosteroid therapy

Computed tomography revealed some radiologic findings of the renal parenchymal lesions in five patients. Among them, multiple low-density lesions on enhanced CT (Fig. 2a, c, e) were the most frequent findings and were observed in three patients (patients 1, 3, and 4 in Table 1), one of whom also had diffuse thickening of the renal pelvic wall (Fig. 2c). On the other hand, the renal parenchymal

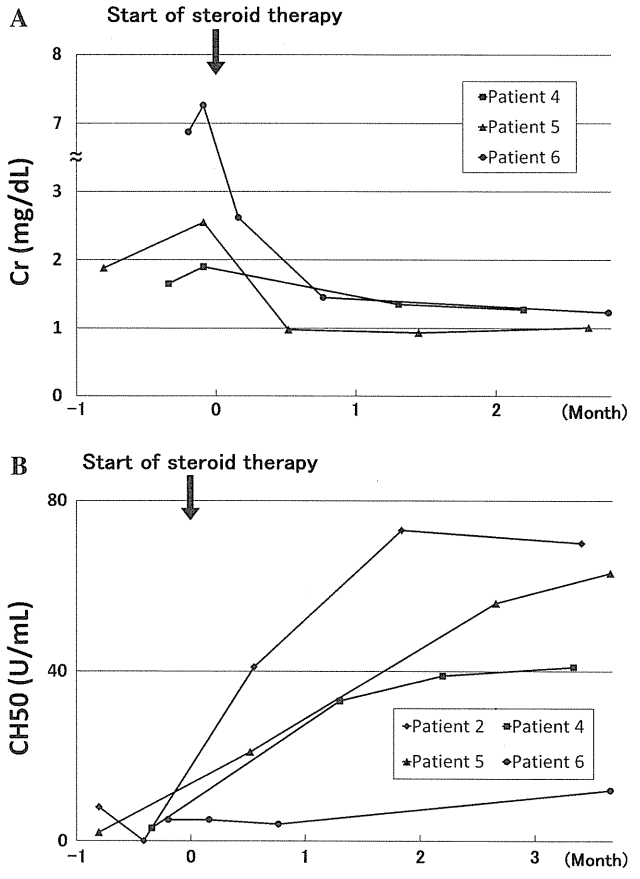

Fig. 1 Patient's clinical course. Serum creatinine concentrations rapidly declined within 1 month after corticosteroid therapy in patients 4, 5, and 6, who showed severe renal dysfunction before therapy (**a**). Hypocomplementemia was improved by corticosteroid therapy within 2 months in patients 2, 4, and 5, but response to therapy was insufficient in patient 6 (**b**)

lesion observed in two patients (patients 5 and 6 in Table 1) who underwent only plain CT because of severe renal dysfunction before corticosteroid therapy was diffuse bilateral renal swelling (Fig. 2g, i). No radiological abnormal findings were apparent in only one patient (patient 2 in Table 1).

On the whole, corticosteroid therapy quickly ameliorated renal lesions. Three patients with multiple low-density lesions on enhanced CT showed recovery of contrast enhancement of the renal cortex after therapy. However, scar-like focal cortical atrophy persisted in two of them (Fig. 2b, f). Two patients with diffuse bilateral renal swelling showed apparent improvement of the renal swelling after therapy (Fig. 2h, j). A few small scar-like focal cortical atrophy persisted in one of them (Fig. 2h), whereas relatively uniform contrast enhancement was observed in the other (Fig. 2j). Diffuse thickening of the renal pelvic wall became thinner after therapy in patient 3 (Fig. 2d).

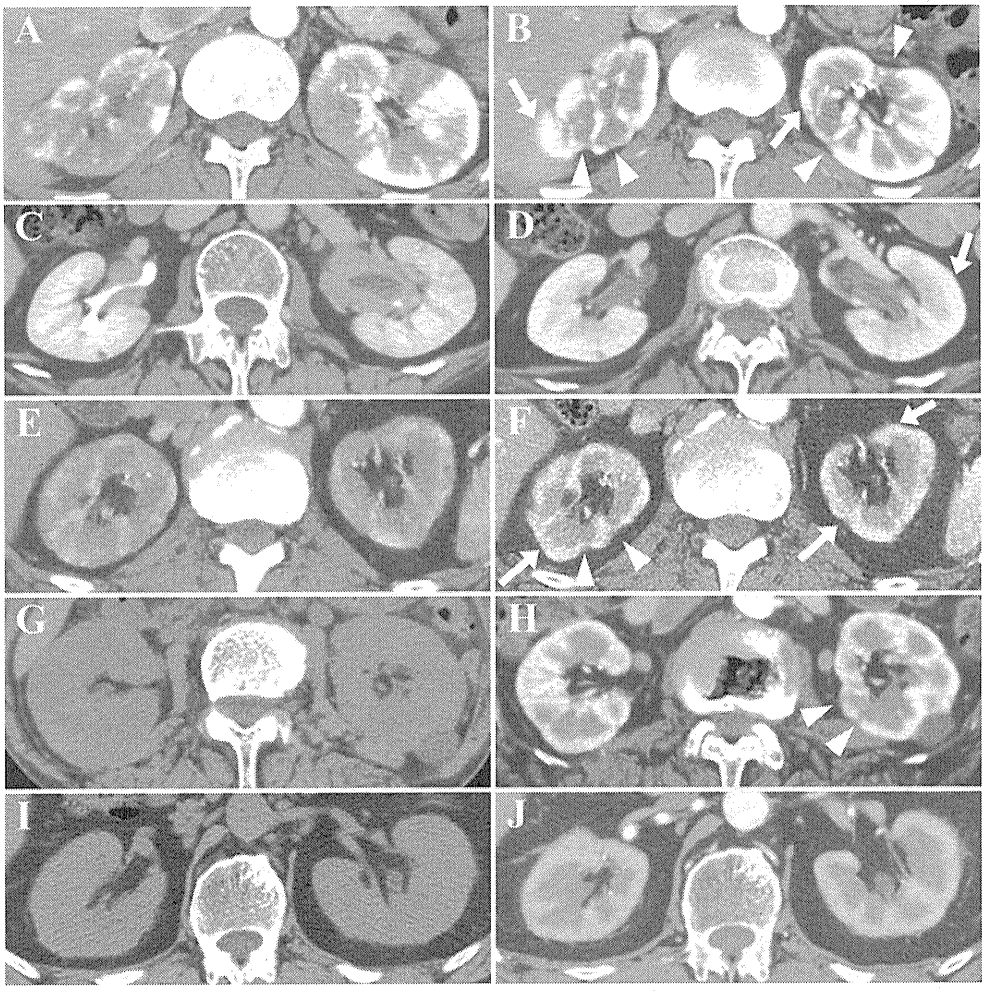

Fig. 2 Contrast-enhanced (**a–f**, **h**, **j**) or plain (**g**, **i**) computed tomography (CT) findings of immunoglobulin (Ig)G4-related renal lesions and changes induced by corticosteroid therapy. Multiple low-density lesions on enhanced CT (**a** patient 1, **c** patient 3, **e** patient 4), diffuse bilateral renal swelling on plain CT (**g** patient 5, **i** patient 6), and diffuse thickening of the renal pelvic wall (**c**) were observed before therapy. Contrast enhancement of the renal cortex recovered after therapy in all patients with multiple low-density lesions (**b**, **d**, **f** *arrows* show recovering lesions), though some atrophic areas of decreased enhancement remained (**b**, **f** *arrowheads* show atrophic lesions). Diffuse bilateral renal swelling was improved with small areas of decreased enhancement 4 months after therapy in patient 5 (**h** *arrowheads* show lesions with decreased enhancement) and with relatively uniform contrast enhancement 1 month after therapy in patient 6 (**j**). Diffuse thickening of the renal pelvic wall became less marked 4 months after therapy in patient 3 (**d**)

Histological and immunohistochemical findings and changes associated with corticosteroid therapy

Histological findings of six renal specimens before corticosteroid therapy were as follows: Dense lymphoplasmacytic infiltration and sparse distribution of atrophic renal tubules in the renal interstitium were present (Fig. 3a–c). The findings of renal tubulitis were mild (Fig. 3a, e, f). The interstitial lesions were often localized, and the border of the lesion was fairly clear (Fig. 3b). On the other hand, the interstitial lesion was diffuse in two patients (patients 5 and 6 in Table 1) with severe renal dysfunction whose plain CT images before therapy showed diffuse bilateral renal swelling (Fig. 3c). In one case (patient 6), inflammation extended beyond the renal capsule (Fig. 3d). Fibrosis with occasional infiltrating cells was observed (Fig. 3e, f). Lymph follicles were not evident in any patient. Glomerular lesions were observed in four: one showed Henoch–Schönlein purpura nephritis [28], another IgA nephropathy, and the other two focal endocapillary proliferation. Immunohistochemically, in addition to IgG4-positive plasma cells (Fig. 5a), Foxp3+ cells (Fig. 5b) and CD4+CD25+ T cells (Fig. 5c) were detected in the interstitial lesions of IgG4-related TIN. CD4+ T cells (Fig. 6a) and CD8+ T cells (Fig. 6b) were also found there.

The re-biopsy specimen obtained 1 month after corticosteroid therapy from patient 6 showed that areas with intense cell infiltration remained (Fig. 4a), but infiltration of IgG4-positive plasma cells (Fig. 5d), Foxp3+ cells (Fig. 5e), and CD4+CD25+ T cells (Fig. 5f) considerably decreased, and there was little obvious fibrosis of the renal

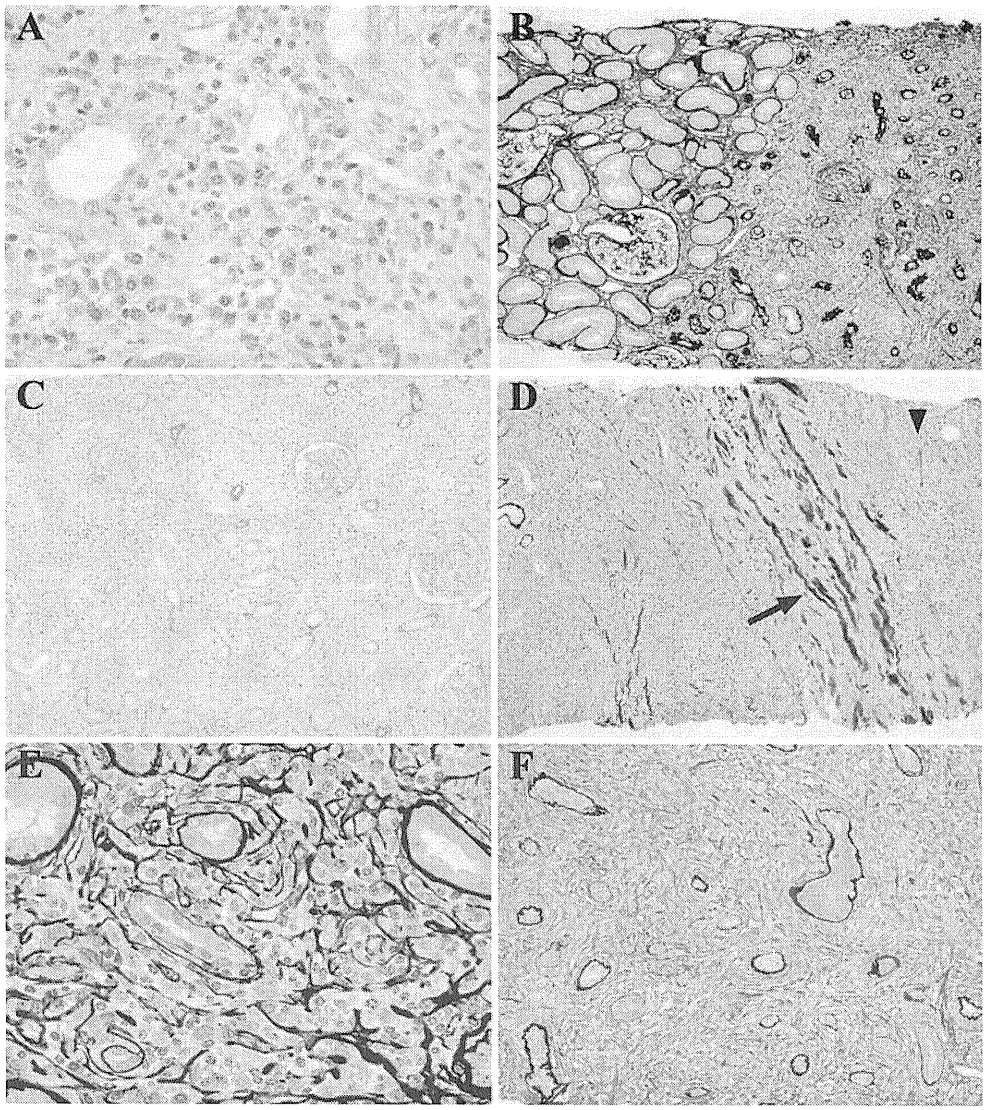

Fig. 3 Light microscopy findings of the renal interstitium before corticosteroid therapy. Severe lymphoplasmacytic infiltration with tubular atrophy was observed (**a** patient 6, **b** patient 4, **c** patient 6). Interstitial lesions were often focal, and the borderline between lesion and nonlesion was fairly clear (**b**). In two patients with severe renal dysfunction, interstitial lesion was diffuse (**c**). Inflammatory lesion beyond the renal capsule was detected (**d** patient 6, *arrow* shows the renal capsule, and *arrowhead* shows inflammation beyond the renal capsule). A characteristic fibrosis that appeared to surround infiltrating cells was observed (**e** patient 4, **f** patient 5) [**a** Hematoxylin and eosin (H&E) staining ×400; **b**, **d** periodic acid-methenamine-silver (PAM)-H&E staining ×100; **c** periodic acid-Schiff (PAS) staining ×100; **e**, **f** PAM-H&E staining ×400]

interstitium (Fig. 4a). The re-biopsy specimen 14 months after therapy from patient 1 demonstrated an obvious decrease of cell infiltration except for small localized infiltrations, whereas patchy marked fibrosis remained (Fig. 4b). In the re-biopsy specimens 4 months after therapy from patient 5 who showed severe renal dysfunction before therapy, we observed mixed lesions where cell infiltration was dense or fibrosis was predominant or neither of these features was noted (Fig. 4c, d, e, respectively). In the re-biopsy specimens 4 months after therapy from patient 3 who showed normal renal function before therapy, we observed the same three components as in patient 5, but fibrosis was not marked. In one patient with diffuse renal interstitial cell infiltration and sparse distribution of atrophic tubules (patient 6), tubular atrophy and sparseness of tubular distribution seemed to partially improve after therapy (Figs. 3c, 4f). In the immunostaining specimens, cell count of various infiltrating cells in the lesions with intense cell infiltration before and after corticosteroid therapy revealed that the characteristic infiltrating cells, such as IgG4-positive plasma cells and Foxp3+ cells, showed significant decreases after corticosteroid therapy (Figs. 5d, e, 7a–c). Other CD4+ T cells and CD8+ T cells were relatively persistent, even in the later stage of therapy

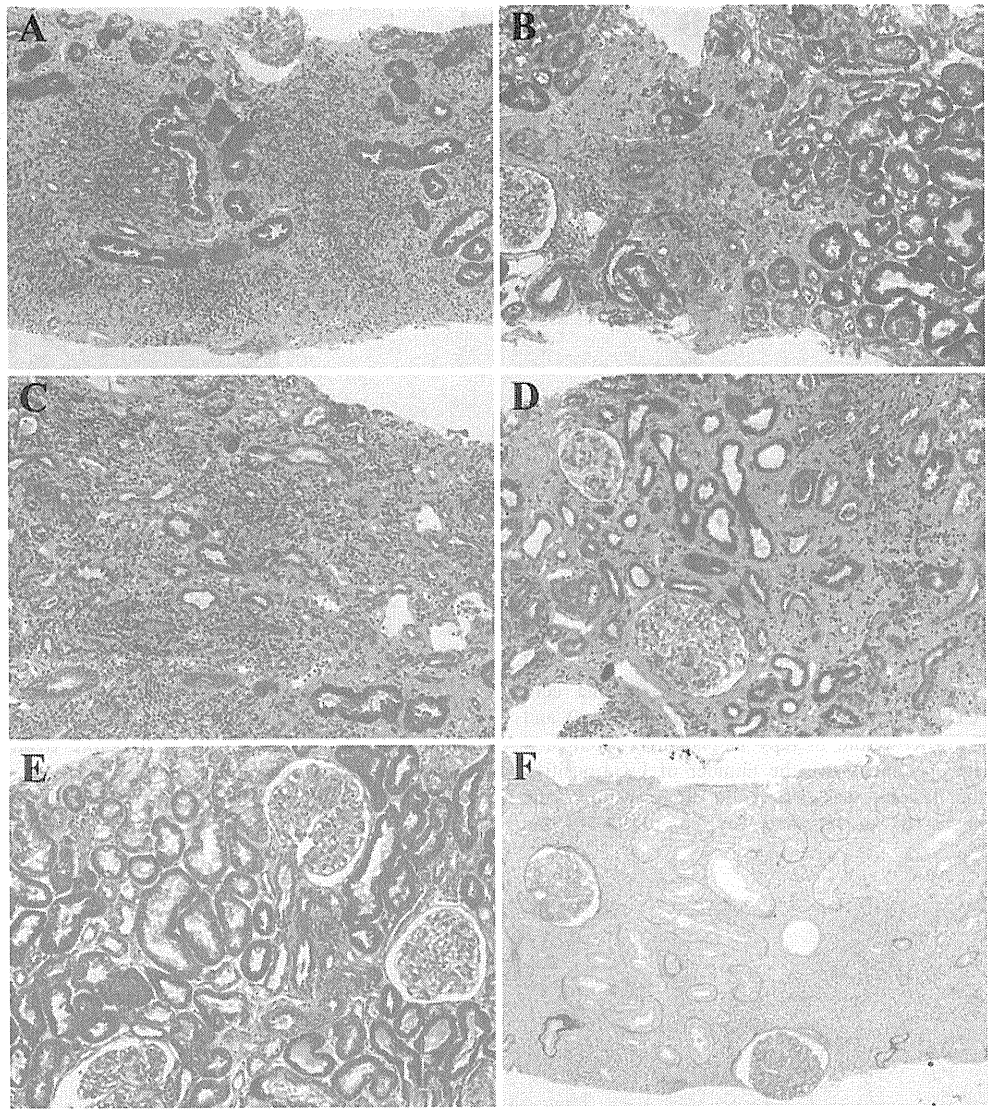

Fig. 4 Changes in light microscopy findings of the renal interstitium after corticosteroid therapy. Azan staining showed that interstitial fibrosis was not evident 1 month after corticosteroid therapy in patient 6 (**a**), whereas localized severe fibrosis was observed 14 months after therapy in patient 1 (**b**). There were mixed lesions where cell infiltration was dense (**c**) or fibrosis was predominant (**d**) or neither of them was noted (**e**) 4 months after therapy in patient 5. Interstitium in the posttreatment specimen of patient 6 seemed to show recovery of some renal tubules 1 month after corticosteroid therapy (**f**) [**a**–**e** Azan staining ×100, **f** periodic acid-Schiff (PAS) staining ×100]

(Figs. 6c, d, 7d, e). The average number of IgG4-positive plasma cells in five different HPF decreased from 156.4/HPF (range 102–210) to 2.0/HPF (range 0–3) in patient 1; 41.8/HPF (range 36–60) to 7.2/HPF (range 1–15) in patient 2; 50.2/HPF (range 25–77) to 12.2/HPF (range 5–36) in patient 3; 27.6/HPF (range 9–46) to 5.0/HPF (range 3–11) in patient 4; 62.2/HPF (range 45–80) to 4.8/HPF (range 3–7) in patient 5; and 73.2/HPF (range 50–108) to 20.8/HPF (range 19–22) in patient 6 (Fig. 7a). In the same way, the average number of Foxp3+ cells decreased from 35.4/HPF (range 26–52) to 8.4/HPF (range 1–16) in patient 1; 25.4/HPF (range 18–45) to 3.8/HPF (range 2–6) in patient 2; 18.0/HPF (range 10–23) to 1.2/HPF (range 0–3) in patient 3; 41.8/HPF (range 36–46) to 4.8/HPF (range 4–6) in patient 4; 33.2/HPF (range 30–35) to 3.0/HPF (range 0–5) in patient 5; and 23.0/HPF (range 19–30) to 3.2/HPF (range 1–5) in patient 6 (Fig. 7c).

Discussion

This study examined the clinicopathological features in patients with IgG4-related TIN before and after corticosteroid therapy. The characteristic laboratory findings before therapy were elevated serum IgE levels or eosinophilia, high incidence of hypocomplementemia, and hyper-β2

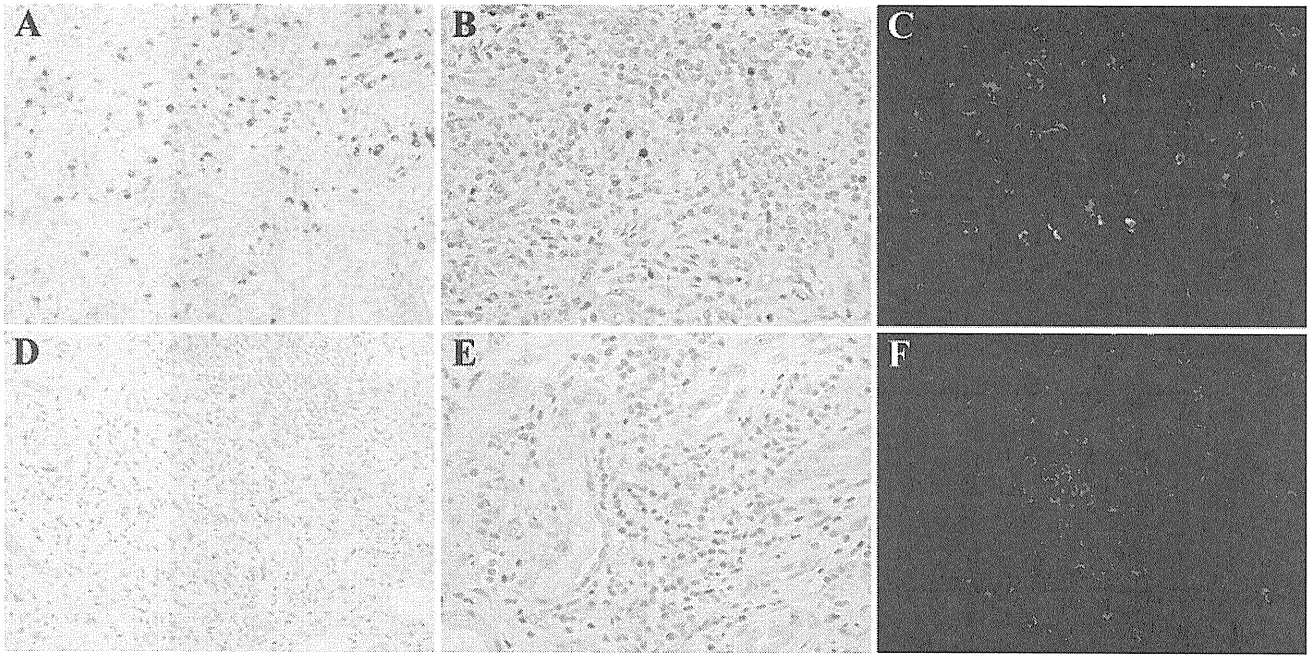

Fig. 5 Changes in immunoglobulin (Ig)G4-positive plasma cell, Foxp3+ cell, and CD4+CD25+ cell infiltration before (**a–c**) and 1 month after (**d–f**) corticosteroid therapy in patient 6. Compared with the pretreatment specimens (**a**), the number of IgG4-positive plasma cells in the lesions was markedly diminished in the posttreatment specimens (**d**). In the same way, compared with the pretreatment specimens (**b**), the number of Foxp3+ cells obviously decreased in the posttreatment specimens (**e**). Some CD4+CD25+ cells in the lesions were observed before treatment (**c**), whereas almost none were detected after therapy (**f**) [**a**, **d** IgG4 ×200, **b**, **e** Foxp3 ×400, **c**, **f** CD4 (*red*) and CD25 (*green*) ×400]

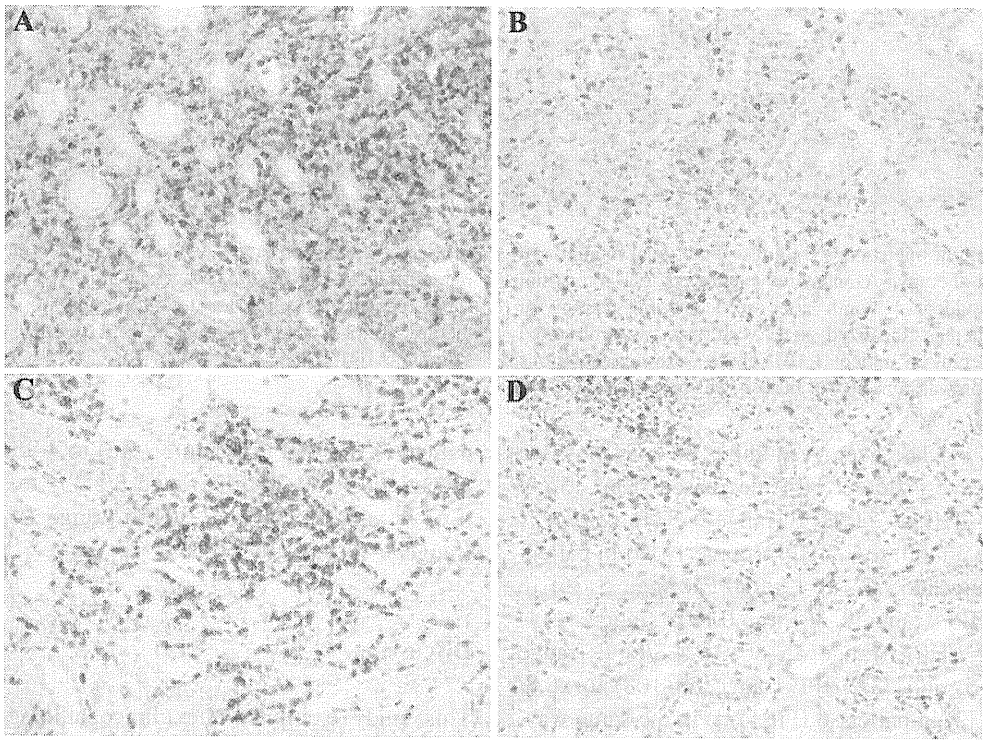

Fig. 6 Changes in CD4+ and CD8+ T cell infiltration before (**a**, **b**) and 4 months after (**c**, **d**) corticosteroid therapy in patient 5. The extent of CD4+ T cell (**a**, **c**) and CD8+ T cell (**b**, **d**) infiltration in the lesions remained largely constant, even after therapy (**a**, **c** CD4 ×100, **b**, **d** CD8 ×100)

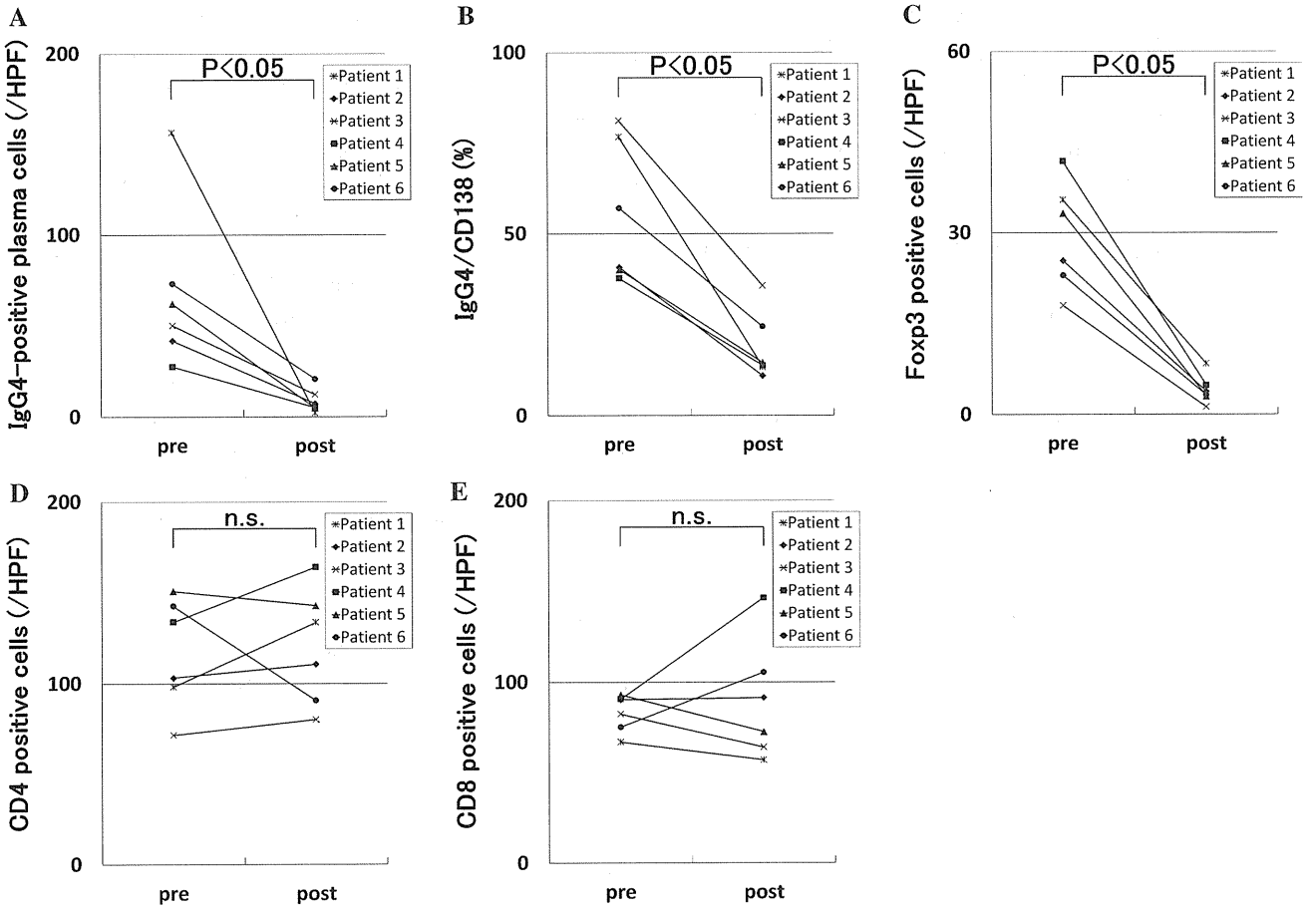

Fig. 7 Changes in numbers of infiltrating cells before and after corticosteroid therapy. In six IgG4-related TIN cases, the average of IgG4-positive plasma cell numbers and the ratio of IgG4-positive cells to CD138-positive cells in the lesions significantly decreased after compared with before corticosteroid therapy (**a**, **b**, respectively). Similarly, average of Foxp3+ cell numbers significantly decreased (**c**). However, average of CD4+ and CD8+ T cell numbers did not change (**d**, **e**, respectively)

microglobulinuria. Renal insufficiency was also frequent, and almost all patients with IgG4-related TIN had some radiographic abnormalities. Therapy with 20–30 mg/day of prednisolone rapidly improved renal function in three patients with severe renal dysfunction, whereas some renal insufficiency persisted in two. Radiographic abnormalities persisted in half of the patients. Histologically, infiltration of CD4+CD25+ T cells and Foxp3+ cells in addition to IgG4-positive plasma cells was characteristic in IgG4-related TIN. Notably, IgG4-positive plasma cells, Foxp3+ cells, and CD4+CD25+ cells had already decreased 1 month after corticosteroid therapy in one case (patient 6), whereas small to moderate numbers of CD4+ T cells and CD8+ T cells infiltrated where inflammation persisted in all patients. In the later stage after treatment, patchy or regional fibrosis remained to some extent.

In this study, radiographic findings of renal lesions were almost the same as those noted in past reports [21, 30]. Multiple low-density lesions on enhanced CT, diffuse bilateral renal swelling, and thickening of the renal pelvic wall were the major features. Past reports mentioned that corticosteroid therapy was effective in improving the appearance of renal lesions on imaging study, although cortical scar or renal capsular dimpling persisted in a small number of cases [30–33]. In our study, although some renal parenchymal lesions showed recovery of contract enhancement, others progressed to scar-like atrophy in three of six cases. In contrast to past studies [30–33], both imaging study and histological analysis were performed before and after corticosteroid therapy in all patients in this study. Our data might suggest that these atrophic lesions in imaging study correspond to the histological fibrotic lesions. In addition, recovery of contrast enhancement might relate to the partially normalized renal interstitium observed in the posttreatment specimens of patients 5 and 6, who showed diffuse TIN and renal tubular atrophy before therapy. In this way, IgG4-related TIN could leave macroscopic atrophy and microscopic fibrosis, which might explain why renal function did not totally recover after corticosteroid therapy in our patients with severe renal insufficiency. The possibility of these sequelae must be considered when determining the corticosteroid dose and optimal timing of the initiation of treatment.

The main histological findings before corticosteroid therapy were consistent with previously published histological features of IgG4-related TIN [21, 22, 34]. In addition, extension of inflammation beyond the renal capsule reported by Yamaguchi et al. [29] was also observed and seemed to correspond to the extension of lesions beyond the pancreatic capsule in autoimmune pancreatitis (AIP). Zen et al. [35] reported infiltration of CD4+CD25+ T cells and Foxp3+ cells in the lesions of IgG4-related pancreatitis and cholangitis and that those lesions had significantly increased levels of Th2 and regulatory T-cell cytokines. Nakashima et al. [36] also reported increased Th2 and regulatory cytokines in the lesions of IgG4-related TIN. In this study, we similarly confirmed the presence of CD4+CD25+ T and Foxp3+ cells in the renal interstitium where lymphocytes and IgG4-positive plasma cells infiltrated. Accordingly, the presence of CD4+CD25+ T or Foxp3+ cell infiltration might be another distinctive finding of IgG4-related TIN. As Houghton and Troxell [37] reported that an abundant infiltration of IgG4-positive plasma cell is not so specific for IgG4-related TIN, it will be necessary to survey the presence or absence of these regulatory cells in TIN diseases other than IgG4-related TIN.

We focused on histological and immunohistochemical changes of IgG4-related TIN during the clinical course of corticosteroid therapy. The area with infiltrating cells in the renal interstitium decreased with the passage of time after therapy was initiated. Conversely, more obvious regional fibrosis was observed in the re-biopsy specimens, although there was the possibility of sampling bias due to randomly performed biopsies. Reflecting these histological findings, the radiological lesions to some degree showed recovery following therapy, whereas in some parts, there was progression to scar formation. These findings suggest the need to search for ways to prevent fibrosis in addition to control of inflammation. Whether early initiation of corticosteroid therapy can prevent fibrosis remains to be verified, as there was no untreated control group in this study and so this point could not be concluded definitively. However, comparing posttreatment findings of patients 3 and 5, both of whom underwent re-evaluation 4 months after the start of therapy, macroscopic atrophy and microscopic fibrosis were more marked in patient 5 with severe renal dysfunction before therapy than in patient 3 with normal renal function then. This difference implies that early initiation of corticosteroid therapy prevents fibrosis to some extent. The possibility should also be considered that corticosteroid therapy alone is insufficient to prevent residual fibrosis despite early initiation. Other treatment options should be examined, as rituximab therapy, for example, has been reported to be effective in achieving clinical improvement in IgG4-RD [38, 39]. The characteristic cells, including CD4+CD25+ T cells, Foxp3+ cells, and IgG4-positive plasma cells, seemed to disappear quickly after corticosteroid initiation and did not reappear as long as corticosteroid was administered, whereas other CD4+ T and CD8+ T cells persisted for a long time in lesions where cell infiltration was still observed. This finding suggests that we cannot precisely evaluate IgG4-positive plasma cell infiltration in diagnosing IgG4-related TIN if corticosteroid therapy has already been initiated. The pathogenic significance of this finding remains unclear. More accumulation of clinical and histological data of IgG4-related TIN, including recurrences during corticosteroid tapering or after cessation of corticosteroid therapy and basic research based on those data, are essential to elucidate the roles of these cells in the pathogenesis of IgG4-related TIN.

Biomarkers that could be used as a goal for treating IgG4-related TIN have not yet been established. It was reported that in AIP, serum IgG4, IgG, and circulating immune complex decreased after corticosteroid therapy and increased at relapse, whereas serum C3 and C4 levels showed reciprocal changes [40]. Tabata et al. [41] suggested that the measurement of serial serum IgG4 levels was useful to determine disease activity of IgG4-RD. Nevertheless, whether normalization of serum IgG4 levels could be a treatment goal is unclear, as it is not always observed despite apparent clinical remission [41]. Cutoff values of these markers as a goal should be further examined. Nishi et al. [42] reported that elevated urine NAG and/or α1-microglobulin concentrations were useful markers for detecting renal abnormalities in their AIP patients with or without clinically detected renal involvement. However, neither urine β2-MG nor NAG entirely responded to corticosteroid therapy and fluctuated despite the continuation of treatment in our study. Thus, reliable markers showing a goal for treatment remain to be identified. Further studies are required to seek such markers, including novel candidates.

The small number of cases is a limitation of our study that relates to the rarity of this disease, and the inconsistent follow-up times of radiological and histological data in each patient is another limitation. However, with increasing worldwide awareness of IgG4-related TIN as a distinct disease entity [21, 22], we can expect larger studies that include many more cases and anticipate that, with more patients enrolled in each stage of corticosteroid therapy, especially in the very early and later stages, the dynamics of various infiltrating cells during the clinical course will be better clarified.

In conclusion, our investigations suggest that, clinically, persistent renal insufficiency associated with macroscopic atrophy and microscopic fibrosis is not so rare in IgG4-related TIN. Pathologically, the behavior of regulatory T cells during the clinical course of corticosteroid therapy is

quite similar to that of IgG4-positive plasma cells, and the distinctive behavior pattern of those cells may provide a clue to the mechanisms underlying this disease. Further studies are required to elucidate the total picture of this disease, including its clinical and pathogenic aspects.

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Conflict of interest None.

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Review Article

IgG4-Related Disease : A Novel Lymphoproliferative Disorder Discovered and Established in Japan in the 21st Century

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IgG4-related disease is a novel lymphoproliferative disorder that shows hyper-IgG4-γ-globulinemia and IgG4-producing plasma cell expansion in affected organs with fibrotic or sclerotic changes. Patients show systemic inflammatory conditions and various symptoms depending on the affected organ. Since the first report of patients with elevated serum IgG4 in sclerosing pancreatitis in 2001, various systemic disorders described by many names have been reported. Despite similarities in the organs involved in IgG4-related Mikulicz's disease and Sjögren's syndrome, there are marked clinical and pathological differences between these conditions. Most patients diagnosed with autoimmune pancreatitis in Japan have IgG4-related pancreatitis [Type 1 autoimmune pancreatitis (AIP), lymphoplasmacytic sclerosing pancreatitis (LPSP)], a disease distinct from some of the western type [Type 2 AIP, idiopathic duct-centric chronic pancreatitis (IDCP), autoimmune pancreatitis with granulocytic epithelial lesions (GEL)]. Diagnosis of IgG4-related disease is characterized by both elevated serum IgG4 (>135 mg/dL) and histopathological features including lymphocyte and $IgG4^+$ plasma cell infiltration ($IgG4^+$ plasma cells/IgG^+ plasma cells > 40%). Differential diagnosis from other distinct disorders, such as sarcoidosis, Castleman's disease, Wegener's granulomatosis, lymphoma, cancer, and other existing conditions associated with high serum IgG4 level or abundant IgG4-bearing plasma cells in tissues is necessary. We have begun a clinical prospective study to establish a treatment strategy (Phase II prospective treatment study for IgG4-multiorgan lymphoproliferative syndrome : UMIN R000002311). [*J Clin Exp Hematopathol 51(1) : 13-20, 2011*]

Keywords: Mikulicz's disease, Sjögren's syndrome, autoimmune pancreatitis, Castleman's disease, glucocorticoid

WHAT IS IgG4-RELATED DISEASE ?

IgG4-related disease is a lymphoproliferative disorder that shows hyper-IgG4-γ-globulinemia and IgG4-producing plasma cell expansion in affected organs with fibrotic or sclerotic changes. Patients show systemic inflammatory conditions and various symptoms depending on the affected organ. Although the lacrimal glands, salivary glands, and pancreas are the major affected organs, the involvement of various other organs has been reported, and it is questionable whether all of these represent the same conditions. Another feature of IgG4-related disease is particular glucocorticoid responsiveness. Furthermore, spontaneous regression without any treatment may occur. Thus, the most important purpose of diagnosis of IgG4-related disease is the definition of therapeutic strategy. There are a number of disorders with similar characteristics, and differential diagnosis must be made for diseases with poor responsiveness to glucocorticoid or different clinical courses.

We are now conducing multicentric cooperative research and continuing critical discussion regarding this condition, with financial support from Intractable Diseases, Health, and Labor Sciences Research Grants from the Ministry of Health, Labor, and Welfare to two groups led by Prof. Kazuichi Okazaki and by Prof. Hisanori Umehara.

There are many synonyms because IgG4-related disease is a systemic disease, such as IgG4-multiorgan lymphoproliferative syndrome ($IgG4^+$ MOLPS),[1] IgG4-related sclerosing disease,[2] systemic IgG4-related plasmacytic syndrome (SIPS),[3] *etc.* As the use of many different names for the same disease entity causes confusion and misunderstanding, the standardized official term "IgG4-related disease" was decided upon at the second meeting of the Umehara group on 11 Feb 2010.[4]

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DIAGNOSIS OF IgG4-RELATED DISEASE

Although IgG4-related disease is a newly defined clinical entity and is not yet well known, it is not an extremely rare condition. The incidence rate of new cases of IgG4-related disease calculated using the data for Ishikawa prefecture is 2.63-10.2 per 1 million people ; therefore, 336 to 1,300 new cases may develop every year in Japan (reported by Suzuki R, *et al.*).[4] Even if there are some differences in local distribution of incidence, several new cases may be encountered at main hospitals, such as university hospitals.

Similar to other diseases, it is not possible for physicians to make a correct diagnosis if they do not suspect a particular clinical entity and if there is no established diagnostic approach for IgG4-related disease. Although it is not so difficult for physicians to suspect IgG4-related disease if they have some experience with typical cases, it is difficult to make a diagnosis on first encountering this disease. Therefore, we proposed diagnostic criteria for IgG4$^+$ MOLPS and prepared diagnostic guidelines (Table 1).[5]

Diagnosis of IgG4-related Mikulicz's disease

Mikulicz's disease (MD) is a clinical condition that shows bilateral symmetrical dacryoadenitis (swelling of the lacrimal glands) and sialadenitis (swelling of the parotid glands and submandibular glands). Since Morgan *et al.* reported that MD is not a distinct clinical and pathological disease but is merely one manifestation of a more generalized symptom complex known as Sjögren's syndrome (SS),[6] MD has attracted very little interest in western countries. However, MD has attracted attention and been reported in Japan. Yamamoto *et al.* reported that MD is also a subtype of IgG4-related disease,[7] and an IgG4$^+$ MOLPS/MD research group was organized in September 2004 to perform a retrospective national study. The results of this study revealed many differences between MD and SS[1,8] : 1) male SS patients are very rare, but almost half of MD patients are male ; 2) swelling of glands (lacrimal, parotid, and submandibular) is remarkable, but symptoms of dryness (xerostomia, xerophthalmia) are unobtrusive in patients with IgG4$^+$ MD ; 3) the incidence of autoantibodies is lower in patients with IgG4$^+$ MD than in SS (the incidence of rheumatoid factor and anti-nuclear antibodies in IgG4$^+$ MD is almost one quarter that in SS, and most cases of IgG4$^+$ MD are negative for anti-SSA antibodies and anti-SSB antibodies) ; 4) serum IgG4 level is high and IgG4$^+$ plasma cell concentration is high in IgG4$^+$ MD ; and 5) rates of allergic rhinitis and bronchial asthma, serum IgE concentrations, and eosinophil count among white blood cells are higher in IgG4$^+$ MD than in SS, suggesting the involvement

Table 1. Proposed diagnostic criteria for systemic IgG4-related diseases : IgG4$^+$ MOLPS (the grant from Intractable Diseases, Health and Labor Sciences Research Grants from the Ministry of Health, Labor and Welfare. Entitled the research for establishing a novel disorder, IgG4-related multiorgan lymphoproliferative syndrome ; IgG4$^+$ MOLPS ; Umehara's group). Diagnosis of IgG4$^+$ MOLPS is defined with both 1) and 2)

1) Elevated serum IgG4 (>135 mg/dL)
AND
2) Histopathological features including lymphocyte and IgG4$^+$ plasma cell infiltration (IgG4$^+$ plasma cells/IgG$^+$ plasma cells>40%) with typical tissue fibrosis or sclerosis.

Note :
- It is necessary to distinguish IgG4$^+$ MOLPS from other distinct disorders, including sarcoidosis, Castleman's disease, Wegener's granulomatosis, lymphoma, and cancer.
- Patients fulfilling only one of the above criteria are classified as "suspected IgG4$^+$ MOLPS."
- Patient fulfilling both (1) and (2) and having other distinct disorders (designated as "XX"), are classified as having "XX disease with suspected association with IgG4$^+$ MOLPS."
- Patients diagnosed with IgG4$^+$ MOLPS, but refractory to glucocorticoid treatment, should be re-diagnosed.

Diagnostic guideline ; Suspicious of IgG4$^+$ MOLPS

1. Presence of only one can be enough the suspicious IgG4$^+$ MOLPS lesion.
   1) Symmetrical swelling of one of the lacrimal, parotid or submandibular glands
   2) Autoimmune pancreatitis
   3) Inflammatory pseudotumor
   4) Retroperitoneal fibrosis
   5) Histopathological findings are similar to lymphoplasmacytosis or suspected Castleman's disease.
2. Presence of at least two would be sufficient for suspected IgG4$^+$ MOLPS.
   1) unilateral swelling of one of the lacrimal, parotid, or submandibular glands ; 2) orbital tumorous lesion, 3) autoimmune hepatitis, 4) sclerosing cholangitis, 5) prostatitis, 6) patchy meningitis, 7) interstitial pneumonitis, 8) interstitial nephritis, 9) mediastinal fibrosis, 10) thyroiditis or hypothyroidism, 11) hypophysitis, 12) inflammatory aneurysm.
3. Common findings in patients with IgG4$^+$ MOLPS.
   1) polyclonal hyper-IgG-gammopathy, 2) elevation of serum IgE or eosinophilia, 3) hypocomplementemia or presence of immune complex in serum, 4) tumorous lesion or lymphadenopathy with strong accumulation in ^{67}Ga-scan or ^{18}FDG-PET-scan

of allergic factors in this disease.

The majority of MD patients suffer IgG4-related dacryoadenitis and sialadenitis, but other conditions, such as SS, sarcoidosis, and lymphoma (especially, mucosa-associated lymphoid tissue, MALT lymphoma) may present symmetrical swelling of lacrimal and salivary glands. Thus, the clinical definition of IgG4-negative MD is still a contentious issue.

We have proposed diagnostic criteria for $IgG4^+$ MD as part of the $IgG4^+$ MOLPS/MD research group, which were approved by the Japanese Sjögren's Syndrome Society at the meeting in September 2008 (Table 2).[5] The therapeutic effect of glucocorticoid treatment in SS is insufficient, and use of glucocorticoid is not generally recommended due to its adverse effects. In contrast, glucocorticoid therapy can reduce $IgG4^+$ MD patients' symptoms dramatically, so we strongly recommend its use. This is therefore an important criterion because it is related to therapeutic strategy.

For diagnosis, high serum concentration of IgG4 (>135 mg/dL) or histopathological findings of $IgG4^+$ plasma cell infiltration in swollen lacrimal, parotid, or submandibular glands ($IgG4^+$ plasma cells/IgG^+ plasma cells > 40%, in 5 high-power fields) is required. As biopsy may be invasive and may cause some complications, informed consent is required following extensive discussion with an ophthalmologist and/or otorhinolaryngologist. Minor salivary gland biopsy may sometimes be substituted when biopsy of major salivary glands is difficult. The sensitivity of detection of $IgG4^+$ plasma cells is relatively low (although this is sometimes sufficient for diagnosis), and sclerosis/fibrosis is unremarkable in minor salivary gland specimens.

Diagnosis of type 1 autoimmune pancreatitis

Autoimmune pancreatitis (AIP) is a pancreatitis that is suspected autoimmune mechanism with symptoms similar to those of pancreatic cancer ; therefore, differential diagnosis between these conditions is critical.

This pathology was named lymphoplasmacytic sclerosing pancreatitis (LPSP) by Kawaguchi in 1991,[9] and is characterized by massive lymphocyte and plasma cell infiltration, fibrosis that focally gives rise to a swirling pattern (storiform fibrosis), focal destruction of pancreatic acini, and replacement with fibrosis. The same inflammatory process is observed around the main and interlobular ducts, leaving the duct epithelium and lumen intact. Veins are obliterated by the same inflammatory process (obliterative phlebitis).

In 1995, Yoshida *et al.* proposed the concept of AIP because these patients had hyper-γ-globulinemia, various autoantibodies, lymphocytic infiltration into pancreatic tissue, complication with other autoimmune diseases, and good glucocorticoid responsiveness, which fulfilled MacKay's criteria for autoimmune disease.[10] Diagnostic criteria for AIP were later proposed twice (in 2002 and 2006) by the Japan Pancreas Society (Table 3).[11]

As Hamano *et al.* reported high serum IgG4 concentration in AIP patients[12] and patients were shown to have IgG4-producing plasma cell infiltration in pancreatic tissue,[13] serum and tissue IgG4 became key markers for diagnosis of AIP.

Although AIP has also been reported in western countries, some cases of AIP, especially in Europe, appeared in younger patients and were sometimes complicated with inflammatory bowel diseases ; therefore, at least some of these cases appear to represent a different disorder from AIP in Japan. Histopathologically, some cases of AIP reported in western countries are "idiopathic duct-centric chronic pancreatitis (IDCP)"[14] or "autoimmune pancreatitis with granulocytic epithelial lesions (GEL),"[15] which are caused by neutrophilic granulocyte infiltration and are not related to IgG4. Chari *et*

Table 2. Diagnostic criteria of systemic IgG4-related Mikulicz's disease (Japanese Society of Sjögren's Syndrome, Sep 2008)

1) Persistent (>3 months), symmetrical swelling of the lacrimal, parotid, and submandibular glands, involving at least two pairs.
2) Serologically high levels of immunoglobulin (Ig) G4 (≥135 mg/L).
3) Marked IgG4-positive plasmacyte infiltration (≥40% IgG4-positive/IgG-positive cells in five high-power fields) into lacrimal and salivary gland tissues.

In terms of diagnosis, IgG4-related Mikulicz's disease is defined as satisfying Item 1 and either Item 2 and/or 3. This form of systemic IgG4-related disease is often accompanied by lesions in multiple organs. Sarcoidosis, Castleman's disease, Wegener's granulomatosis, and malignant lymphoma need to be considered as differential diagnoses.

Table 3. Clinical diagnostic criteria for autoimmune pancreatitis 2006 (The research group of refractory pancreatic disease, grant from the Ministry of Health, Labor and Welfare. Japan Pancreas Society)[11]

1) Diffuse or segmental narrowing of the main pancreatic duct with irregular wall and diffuse or localized enlargement of the pancreas by imaging studies, such as abdominal ultrasonography (US), computed tomography (CT), and magnetic resonance imaging (MRI).
2) High serum γ-globulin, IgG or IgG4, or the presence of autoantibodies, such as antinuclear antibodies and rheumatoid factor.
3) Marked interlobular fibrosis and prominent infiltration of lymphocytes and plasma cells in the periductal area, occasionally with lymphoid follicles in the pancreas.

For diagnosis, criterion 1 must be present, together with criterion 2 and/or criterion 3. Diagnosis of autoimmune pancreatitis is established when criterion 1, together with criterion 2 and/or criterion 3, are fulfilled. However, it is necessary to exclude malignant diseases, such as pancreatic or biliary cancers.

al. referred to IgG4-related AIP (LPSP) as type 1 and neutrophilic granulocyte lesions of AIP (IDCP, GEL) as type 2 (Table 4).[16] Although these two entities are similar in their good glucocorticoid responsiveness, they are completely different disorders so using the same disease category seems to be inappropriate.

Several international meetings have been held to determine diagnostic criteria for AIP,[17] but a final decision has yet to be made. The consensus regarding type 1 AIP is swelling of the pancreas, hyper-IgG4-γ-globulinemia, pathological features of LPSP, including fibrosis, obliterative phlebitis, and $IgG4^+$ plasma cell infiltration, and good glucocorticoid responsiveness. However, the pancreas is an organ from which sufficient biopsy specimens are difficult to obtain using standard procedures, except open laparotomy. Many Japanese physicians and researchers seem to exclude addition of glucocorticoid responsiveness to the diagnostic criteria of type 1 AIP without sufficient imaging examination, including endoscopic retrograde cholangiopancreatoscopy (ERCP). In contrast, because invasive examinations including ERCP are rarely performed in western countries, many researchers and physicians have proposed adding glucocorticoid responsiveness to the criteria. This controversy makes it difficult to reach a consensus.

IgG4-related disease and other organ involvement

Reports of IgG4-related disease in type 1 AIP are followed by cholangitis, cholecystitis, dacryoadenitis,[1,2,8,18] sialadenitis,[1,2,8] retroperitoneal fibrosis, mediastinal fibrosis, tubulointerstitial nephritis,[19] pulmonary lesions such as interstitial pneumonitis, inflammatory pseudotumor of the lung, liver, or breast, lymphadenopathy,[18] hypophysitis, pachymeningitis, arthritis, skin lesions, inflammatory aortic aneurysm, tumorous lesion of coronary artery, some types of autoimmune hepatitis, thyroiditis, prostatitis, gastritis, major duodenal papilla lesions, colitis or colon polyps, pouchitis, *etc.*

IgG4-related disease occurs in various systemic organs, and as the difficulty and invasiveness of biopsy procedures differ among organs, the diagnostic criteria may also differ for each organ. As a systemic disorder, both serum and histopathological findings (Fig. 1) should be present, such as our criteria for $IgG4^+$ MOLPS (Table 1).[5]

With regard to the diagnostic criteria of $IgG4^+$ MD, the involved organs, *i.* e., the lacrimal and/or salivary glands, are located relatively close to the body surface. Therefore, histopathological findings are important for diagnosis. However, both clinical disease distribution and serum data can also be used for diagnosis of $IgG4^+$ MD.

For diagnosis of AIP, however, it is extremely difficult to obtain biopsy tissue samples from the pancreas. Therefore, diagnosis is centered around serum data, pathological findings, imaging examination, and/or glucocorticoid responsiveness, as mentioned above.

For diagnosis in other organs, although it is important to obtain biopsy specimens, it may be difficult to perform biopsy of deep lesions, such as those in the retroperitoneum, aorta, hypophysis, or dura mater in addition to the pancreas. Therefore, serum data and imaging findings must be considered. Occasionally, patients are diagnosed with a solitary lesion, but the majority of cases have multiple organ involvement. Therefore, it may be possible to obtain biopsy specimens from organs that can be reached more easily and in a less invasive manner, and to examine the distribution of lesions by 2-deoxy-2-(^{18}F) fluoro-D-glucose-positron emission tomography (^{18}FDG-PET) scan, ^{67}Garium-scan, *etc.*, and finally to estimate glucocorticoid responsiveness.

Related groups are working to develop diagnostic criteria and guidelines for IgG4-related nephropathy and IgG4-related lung disease, as subsidiaries of the Umehara group of Health and Labor Sciences Research Grants from Ministry of Health, Labor and Welfare. These groups are also collaborating with the Japanese Society of Nephrology and the Japanese Respiratory Society, respectively.

An international conference of IgG4-related disease will be held in Boston, USA, in October 2011. Before this conference, selected members of Japanese researchers will meet and

Table 4. Differences between Type 1 (LPSP) and Type 2 (IDCP, GEL) histopathological presentations of the autoimmune pancreatitis.

	Type 1	Type 2
Age	Elderly	Young adult
Sex	Male dominant	No gender bias
Distribution	Whole world	Western countries
Serum IgG4	Elevated	Not elevated
Histopathology	LPSP	IDCP, GEL
Infiltrating cells	$IgG4^+$ plasma cells	Granulocytes
Complication	Various general disorders	Inflammatory bowel disease

LPSP, lymphoplasmacytic sclerosing pancreatitis ; IDCP, idiopathic duct-centric chronic pancreatitis ; GEL, autoimmune pancreatitis with granulocytic epithelial lesion

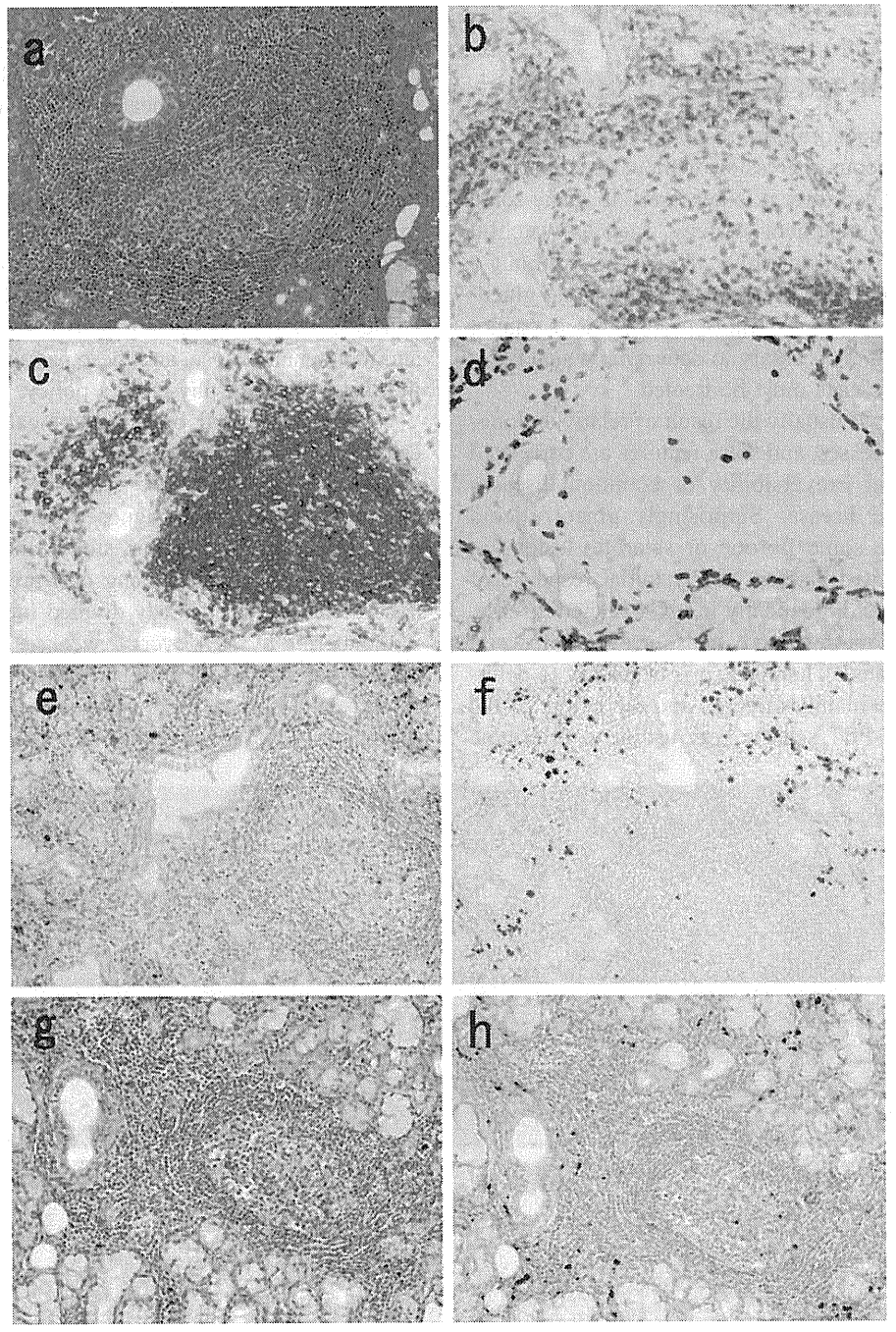

Fig. 1. Histopathological findings of labial minor salivary gland biopsy in IgG4-multiorgan lymphoproliferative syndrome (IgG4$^+$ MOLPS)/Mikulicz's disease (*1a-1h*). (***1a***) Hematoxylin & eosin staining ; (***1b***) CD3 ; (***1c***) CD20 ; (***1d***) CD38 ; (***1e***) IgG ; (***1f***) IgG4 immunostaining. (***1g***) κ and (***1h***) λ-*in situ* hybridization. Massive lymphocyte and plasmacyte infiltration and lymphoid follicle formation were seen in IgG4$^+$ MOLPS. The ducts remained clear without lymphocytic infiltration. CD20$^+$ B cells remained in the follicle, and CD3$^+$ T cells were seen around the follicle. CD38$^+$ plasma cells, IgG$^+$ cells, and IgG4$^+$ plasma cells were scattered in the periphery of the follicle. The ratio of IgG4$^+$ plasma cells/IgG$^+$ plasma cells was $>$40%. There was no remarkable monoclonality between κ and λ-positive B cells (λ showed clearer staining but the differences were small). Revised figure from Masaki *et al.*[1]

publish the Japanese criteria for IgG4-related disease.

TREATMENT OF IgG4-RELATED DISAESE

Not only good glucocorticoid responsiveness, but also cases showing spontaneous regression without any treatment have been reported. Therefore, it is necessary to make some choices with regard to treatment strategy, such as watchful waiting or surgical resection only. However, without a randomized control study among groups treated using glucocorticoid *vs.* watchful waiting, definitive conclusions cannot be made. In addition, it is necessary to determine which types of IgG4-related disease case must be treated.

Fibrosis or sclerosis is usually the result of relatively long-term inflammatory processes, and these features are correlated with refractoriness and irreversibility of common diseases other than IgG4-related disease. Surprisingly, glucocorticoid treatment can improve some fibrotic or sclerotic lesions in patients with IgG4-related disease. Early initial response of glucocorticoid is usually dramatically in IgG4-related disease, however more longer ignorance may cause irreversibility and function failure of organs. Therefore, it is necessary to determine which organs are mainly affected and the extent of the disease spread. ^{18}FDG-PET scan is very useful to determine the distribution of IgG4-related disease, and therefore this technique is highly recommended to determine treatment indications and strategy ; unfortunately, however, ^{18}FDG-PET scan is not covered by health insurance in Japan at present. ^{67}Garium-scan may serve as an alternative if ^{18}FDG-PET is not available. Irreversible functional failure of the pancreas, kidney, lung, or liver will adversely affect the patient's quality of life and result in poor prognosis. Therefore, glucocorticoid treatment should be applied. Although glucocorticoid treatment is effective in IgG4-related disease, there is no consensus regarding starting dose, period of use, how to taper, and maintenance dose, and these parameters are dependent on the institution and physician's policy.

We planned and began a clinical prospective study to establish optimal treatment strategy (Phase II prospective treatment study for IgG4$^+$ MOLPS : UMIN R000002311). We enrolled patients diagnosed according to our tentative diagnostic criteria into this study, and glucocorticoid treatment was implemented using oral prednisolone at an initial dose of 0.6 mg/kg per day divided into three doses per day, with tapering by 10% every 2 weeks. A maintenance dose of 10 mg per day was continued for at least 3 months, and a further daily dose of prednisolone was left up to the attending physician. Final maintenance dose will be decided with refer-

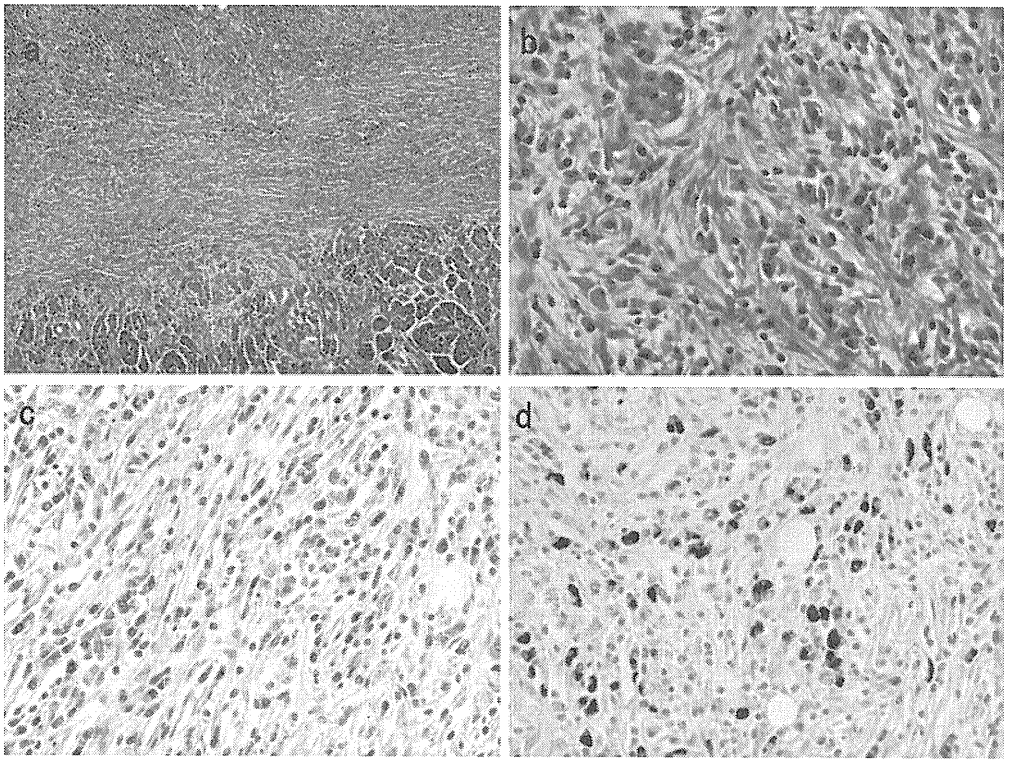

Fig. 2. Histopathological findings of Type 1 autoimmune pancreatitis (AIP). (***2a***, ***2b***) Hematoxylin & eosin staining ; (***2c***) IgG and (***2d***) IgG4 immunostaining. Lymphoplasmacytic infiltration and fibrosis giving rise to storiform fibrosis. Numerous IgG4-positive plasma cells were identified, and the ratio of IgG4$^+$ plasma cells (*2d*)/IgG$^+$ plasma cells (*2c*) was $>$40%.

ence to symptoms and clinical data in each case. In this study, we verified that the majority of patients require 5-10 mg per day of prednisolone as a maintenance dose, because 30%-40% relapse rates have been reported after discontinuation of glucocorticoid.

In typical cases of IgG4-related disease, glucocorticoid response can be confirmed after several days. Although the palpable organs, such as the lacrimal, parotid, and submandibular glands, and lymph nodes, can be confirmed by physical examination, the deep organs, such as the pancreas, should be confirmed by imaging examination (computed tomography) 2 weeks after commencement of glucocorticoid treatment. If the response is not sufficient at 2 weeks, differential diagnosis from other diseases, such as cancer, lymphoma, Castleman's disease, sarcoidosis, *etc.*, should be performed again.

Not only AIP patients, but also those with other types of IgG4-related disease without particular pancreatic lesions, may have glucose intolerance. Thus, glucocorticoid therapy would worsen glucose intolerance, and some patients would require insulin therapy. Informed consent is therefore also important in such cases.

Little evidence of treatment for relapsed and refractory cases have been established. Another course of glucocorticoid is usually effective, but other immunosuppressants, such as azathiopurin,[20] cyclophosphamide, methotrexate, and mizoribine,[21] have also been tried. Furthermore, rituximab[22,23] or bortezomib[24] were reported to show good response rates in studies performed in western countries. However, as mentioned above, it is possible that glucocorticoid refractory cases may be incorrectly diagnosed. It is therefore necessary to establish treatment strategy in a step by step manner, and new agents should be examined in clinical trials.

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