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ORIGINAL ARTICLE

Long-term prognosis of AL and AA renal amyloidosis: a Japanese single-center experience

Masatoyo Ozawa[1,2] · Atsushi Komatsuda[1] · Hiroshi Ohtani[3] · Mizuho Nara[1] · Ryuta Sato[1,2] · Masaru Togashi[1] · Naoto Takahashi[1] · Hideki Wakui[1,4]

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Abstract

Background Few studies have been conducted on the long-term prognosis of patients with amyloid light chain (AL) and amyloid A (AA) renal amyloidosis in the same cohort.

Methods We retrospectively examined 68 patients with biopsy-proven renal amyloidosis (38 AL and 30 AA). Clinicopathological findings at the diagnosis and follow-up data were evaluated in each patient. We analyzed the relationship between clinicopathological parameters and survival data.

Results Significant differences were observed in several clinicopathological features, such as proteinuria levels, between the AL and AA groups. Among all patients, 84.2 % of the AL group and 93.3 % of the AA group received treatments for the underlying diseases of amyloidosis. During the follow-up period (median 18 months in AL and 61 months in AA), 36.8 % of the AL group and 36.7 % of the AA group developed end-stage renal failure requiring dialysis, while 71.1 % of the AL group and 56.7 % of the AA group died. Patient and renal survivals were significantly longer in the AA group than in the AL group. eGFR of >60 mL/min/1.73 m^2 at biopsy and an early histological stage of glomerular amyloid deposition were identified as low-risk factors. A multivariate analysis showed that cardiac amyloidosis and steroid therapy significantly influenced patient and renal survivals.

Conclusions Our results showed that heart involvement was the major predictor of poor outcomes in renal amyloidosis, and that the prognosis of AA renal amyloidosis was markedly better than that in previously reported cohorts. Therapeutic advances in inflammatory diseases are expected to improve the prognosis of AA amyloidosis.

Keywords Amyloid A · Amyloid light chain · Japanese single-center cohort · Long-term prognosis · Renal amyloidosis

Introduction

The amyloidoses are an uncommon group of disorders that are characterized by the extracellular deposition of insoluble fibrils as a result of the abnormal folding of precursor proteins. More than 25 structurally unrelated proteins are known to cause amyloidosis [1]. The kidney is the organ most commonly involved in systemic amyloidosis. Ongoing amyloid deposition in the kidney has been associated with the progressive deterioration of renal function. The two main types of renal amyloidosis are immunoglobulin light chain (AL) amyloidosis, which is associated with plasma cell dyscrasia, and amyloid A (AA) amyloidosis secondary to chronic inflammatory conditions [1]. AL amyloidosis accounts for the largest number of cases, followed by AA amyloidosis in most studies published in Western countries. Other forms are only reported

✉ Atsushi Komatsuda
komatsud@med.akita-u.ac.jp

1 Department of Hematology, Nephrology, and Rheumatology, Akita University Graduate School of Medicine, 1-1-1 Hondo, Akita, Akita 010-8543, Japan

2 Department of Nephrology, Akita Red Cross Hospital, Akita, Japan

3 Department of Nephrology, Akita Kousei Medical Center, Akita, Japan

4 Department of Life Science, Akita University Graduate School of Engineering Science, Akita, Japan

infrequently [2, 3]. The current treatment approach for AL amyloidosis is to eradicate the clonal plasma cells producing the amyloidogenic light chain, while that for AA amyloidosis is to treat the underlying inflammatory disease, thereby reducing the production of the serum AA protein [1].

The prognosis of patients with renal amyloidosis is expected to improve due to recent advances in treatments for AL and AA amyloidosis [1, 4]. However, data on the prognosis of patients with amyloidosis and renal involvement are commonly limited to patients with either the AL or AA type [5–7]. Few studies from Western countries have compared the survival and renal outcomes of patients with AL and AA renal amyloidosis in the same cohort [8]. Therefore, we herein performed a retrospective study with a focus on long-term outcomes and prognostic factors in our cohort of Japanese patients with AL and AA renal amyloidosis, and compared the survival data in our cohort with those in previous studies from other Japanese institutes [9].

Patients and methods

Patients and clinicopathological analysis

This study was based on the renal histological records (between January 1980 and September 2013) of 5693 patients (excluding pediatric and transplant patients) studied at Akita University Hospital and its affiliated hospitals. Renal biopsies were performed on all patients after written informed consent had been obtained. Among 5693 patients, 97 Japanese patients were diagnosed with renal amyloidosis, giving a prevalence rate of 1.7 % among all renal biopsies.

Renal biopsy samples were processed using standard techniques for light and immunofluorescence microscopies. Formalin-fixed, paraffin-embedded sections were stained with hematoxylin and eosin, methenamine silver, periodic acid-Schiff, and Azan/Masson trichrome. Based on the findings of tubular atrophy, interstitial fibrosis, small round cell infiltration, and protein cast formation, tubulo-interstitial damage was categorized as mild (<25 %), moderate (25–50 %), or severe (>50 %). Amyloid staining was performed with Congo-red, Dylon, or Direct Fast Scarlet 4BS. Amyloid distribution patterns were assessed according to the predominant site of amyloid deposition, as described by Hopfer et al. [10] and by an image analysis as described below. The glomerular early type was defined when amyloid deposits were predominantly located in glomeruli in a segmental or mild global manner (Fig. 1a). In the early global phase, the glomerular capillaries were still easily recognizable with mild to moderate mesangial widening. The glomerular late type was defined when amyloid deposits were predominantly located in glomeruli in a moderate to severe global manner (Fig. 1b). In the late global phase, the glomerular capillaries became narrow and were less recognizable with severe mesangial amyloid deposits. In the series by Hopfer et al. [10], most cases had vascular involvement once glomerular involvement became diffuse. The vascular type was defined when the most obvious amyloid deposits were observed within the arteries or arterioles (Fig. 1c). In the series by Hopfer et al. [10], vascular-type cases also showed glomerular amyloid deposits, particularly at the vascular pore. The interstitial type was defined when amyloid deposits were predominantly located in the tubulo-interstitial area. Cryostat sections for immunofluorescence microscopy were stained with fluorescein isothiocyanate-conjugated rabbit antibodies to the human γ heavy chain, α heavy chain, μ heavy chain, κ light chain, λ light chain, C3, and C1q (DakoCytomation, Glostrup, Denmark).

AA amyloidosis was diagnosed based on the finding of positive immunohistochemical staining for AA. Indirect immunoperoxidase staining was performed using a mouse monoclonal antibody against human AA (Quartett, Berlin, Germany), peroxidase-labeled anti-mouse IgG (Dako, Carpentaria, CA, USA), and the Liquid DAB Substrate-Chromogen System (Dako). The diagnosis of AL amyloidosis was based on the findings of negative immunohistochemical staining for AA and predominantly positive immunofluorescence staining for the κ light chain or λ light chain. In the case of inconclusive immunofluorescence findings, indirect immunoperoxidase staining was performed using a rabbit antibody against the human κ light chain (GeneTex, San Antonio, TX, USA), rabbit antibodies against the human λ light chain (GeneTex and DakoCytomation), peroxidase-labeled anti-rabbit IgG (Dako), and the Liquid DAB Substrate-Chromogen System (Dako). Two cases were further examined by indirect immunoperoxidase staining using a set of rabbit antibodies against synthetic peptides corresponding to the human immunoglobulin light chain constant and variable regions [11, 12] at Yamaguchi University, Japan.

Clinical data were collected from medical records for age, sex, urinalysis, serum albumin, serum creatinine (Cr), and monoclonal proteins in the serum and/or urine at the time of renal biopsy. The estimated glomerular filtration rate (eGFR) was determined using the formula for Japanese patients [13]. In patients with AL amyloidosis, the myeloma-associated type was regarded according to the updated diagnostic criteria proposed by the International Myeloma Working Group [14] when bone marrow plasma cells were ≥10 % in the presence of myeloma-defining events (hypercalcemia, renal failure, anemia, and bone lesions). Cardiac amyloidosis was confirmed by autopsy

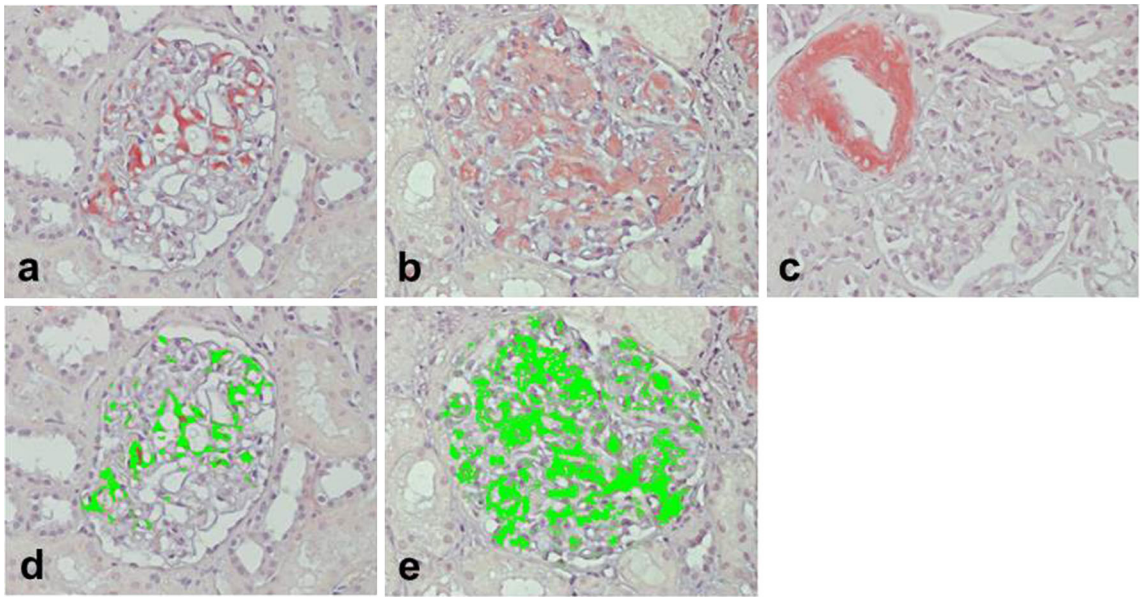

Fig. 1 Histological patterns of amyloid distribution in renal biopsy specimens. Typical figures of the glomerular early type (**a**), glomerular late type (**b**), and vascular type (**c**) are shown (Amyloid staining, ×400). Digital images of the glomerular early type (**d**) and glomerular late type (**e**) are shown with *green* corresponding to the area of amyloid-positive staining. Glomerular-positive area ratios in (**d**) and (**e**) are 8.1 and 24.8 %, respectively (color figure online)

findings, or was suspected based on the findings of electrocardiography, ultrasound echocardiography, or myocardial scintigraphy. Treatment information and follow-up data were collected via a questionnaire in order to determine the outcome of each patient after biopsy.

Among the 97 patients with renal amyloidosis, those meeting any of the following criteria were excluded from the study: (1) insufficient renal biopsy specimens for histological re-evaluation (4 cases); (2) follow-up period <1 year (lost to the follow-up due to untraceable medical records in 10 cases, transferred out in 5 cases, and loss of faith in the treatment in 1 case) (patients who died or developed end-stage renal disease (ESRD) requiring dialysis within a year were included in this study); (3) an inconclusive diagnosis for the typing of renal amyloidosis (2 cases); and (4) insufficient clinical data of urinary protein levels (7 cases). Based on these criteria, 68 Japanese patients with renal amyloidosis were retrospectively enrolled in this study. The study protocol was approved by the Ethics Committee of Akita University Hospital (Approval number 1026).

Digital pathological image analysis

The glomerular amyloid-positive area ratio (GAPAR) on amyloid-stained specimens was quantitatively evaluated using WinRoof version 5.5.0 software (Mitani Co., Fukui, Japan). In 51 out of 53 patients with the glomerular type deposition, the mean ratio was calculated from data on 2–3 glomeruli. In the remaining 2 patients, the specimens contained one glomerulus for evaluation. We determined optimized GAPAR cut-off values to predict patient survival using a receiver operating characteristic curve analysis; the best GAPAR cut-off value was 14.4 %. Based on this result, the glomerular early type was defined when GAPAR was <15 % (Fig. 1d), and the glomerular late type was defined when GAPAR was ≥15 % (Fig. 1e).

The interstitial fibrosis area ratio (IFAR) on Masson trichrome-stained specimens from all enrolled patients was also quantitatively evaluated using WinRoof version 5.5.0 software.

Statistical analysis

Data are presented as the mean ± standard deviation, median with a range, or counts and percentages. Differences between groups were evaluated using the Student's *t* test (for normally distributed continuous variables), Mann–Whitney *U* test (for non-normally distributed continuous variables), or χ^2 test (for categorical variables). The relationship between IFAR and renal function at biopsy was analyzed using Spearman's rank correlation coefficient. Patient survival and renal survival (censored for death) curves according to the typing of amyloidosis, eGFR, and histological patterns were constructed using the Kaplan–Meier method. Differences between groups were assessed using the log-rank test. Furthermore, a multivariate Cox regression analysis was performed in order to

determine the variables affecting patient and renal survival, such as the types of amyloidosis (AL/AA), age, proteinuria, renal function, heart involvement, and treatments, as analyzed in the previous study by Bergesio et al. [8], as well as histological patterns. All analyses were performed using SPSS Statistics Version 21 software (IBM Japan, Tokyo, Japan). P values <0.05 were considered significant in all analyses.

Results

Patient characteristics

The main clinicopathological findings at the diagnosis, and treatments and outcomes during the follow-up in each patient with AL and AA amyloidosis are shown in Tables 1 and 2, respectively.

The differences observed in these clinicopathological features between the two groups are summarized in Table 3. In our cohort of patients, AL amyloidosis was more common than AA amyloidosis. The median age of patients was higher in the AL group (68 years) than in the AA group (59 years) ($P = 0.001$). There were no significant gender differences between the two groups. Proteinuria levels were significantly higher in the AL group (median 4.0 g/gCr or g/day) than in the AA group (median 2.1 g/gCr or g/day) ($P = 0.028$), while serum albumin levels were similar between the two groups. No significant differences were observed in serum Cr levels or eGFR between the two groups. There were no significant differences in amyloid distribution types between the two groups. Simultaneous vascular amyloid deposition was observed in all patients with the glomerular late type. Simultaneous glomerular amyloid deposition, particularly at the vascular pore, was observed in half of the patients with the vascular type. These results were consistent with previous findings obtained in the series by Hopfer et al. [10]. Regarding tubulo-interstitial alterations, the prevalence of moderate to severe tubulo-interstitial damage was slightly higher in the AA group (63.3 %) than in the AL group (39.5 %) ($P = 0.051$).

Laboratory data in all patients were also assessed according to amyloid distribution patterns and tubulo-interstitial damage categories. Proteinuria levels were significantly lower in the vascular type group (median 1.1 g/gCr or g/day) than in the glomerular early type group (median 3.0 g/gCr or g/day) ($P = 0.007$) and glomerular late type group (median 4.7 g/gCr or g/day) ($P = 0.004$) (Fig. 2a). eGFR was significantly lower in the glomerular late type group (median 34.0 mL/min/1.73 m^2) than in the glomerular early type group (median: 52.8 mL/min/1.73 m^2) ($P = 0.008$) or in the vascular type group (median 45.9 mL/min/1.73 m^2 ($P = 0.030$) (Fig. 2b). eGFR was significantly lower in the moderate to severe tubulo-interstitial damage group (median: 28.1 mL/min/1.73 m^2) than in the mild tubulo-interstitial damage group (median 59.1 mL/min/1.73 m^2) ($P < 0.001$) (Fig. 2c). IFAR was significantly higher in the AA group (median 10.0 %) than in the AL group (median 7.2 %) ($P = 0.040$) (Fig. 2d). IFAR showed a negative correlation with eGFR ($R = -0.555$, $P < 0.001$) (Fig. 2e).

In patients with AL amyloidosis, serum monoclonal proteins were identified in 26 patients (68.4 %), with high prevalences of IgG-λ (38.5 %), IgA-λ (30.8 %), and the isolated λ light chain (11.5 %). Urinary monoclonal proteins were identified in 19 patients (50.0 %), with a high prevalence of the λ light chain (84.2 %). Amyloid typing on renal biopsy specimens also showed a high prevalence of the λ light chain subtype (84.2 %).

Underlying diseases in patients with AL and AA amyloidosis are shown in Table 4. In AL amyloidosis, the primary type was the most frequent (68.4 %), followed by the myeloma-associated type (13.2 %) and IgM paraproteinemia-associated type (5.3 %). An extremely rare case, in which AL amyloidosis developed during the long-term follow-up of heavy chain deposition disease, was reported previously [15]. In AA amyloidosis, rheumatoid arthritis (RA) was the most frequent underlying disease (63.6 %), followed by Crohn's disease (10.0 %) and various chronic inflammatory diseases. Rare cases associated with polyangiitis overlap syndrome, sarcoidosis, and indeterminate inflammatory bowel disease complicating extracapillary glomerulonephritis that developed after the initiation of biologics were reported previously [16–18].

Treatments

The median observation period was significantly longer in the AA group (61 months) than in the AA group (18 months) ($P = 0.003$) (Table 3). The treatments for each patient in both groups are summarized in Table 3.

In the AL group, 32 patients (84.2 %) underwent steroid-based or new agent-based therapies. Among the treated patients, melphalan/prednisolone (PSL)-based therapy (52.6 %) was the most frequently used therapy, followed by PSL monotherapy (13.1 %), new agent-based therapy (10.6 %), and others.

In the AA group, 28 patients (93.3 %) underwent steroid-based or biologics-based therapies. Steroid monotherapy was generally selected for old cases of RA and cases of other diseases such as inflammatory bowel diseases. Combination therapies were selected for unsuccessfully controlled cases with steroids only. Combination therapies including biologics were frequently selected for most of the recent survival cases. Among the treated patients, PSL monotherapy

Table 1 Clinicopathological characteristics of each patient with AL amyloidosis

Case no.	Age/ sex	RB (year)	UP (g/gCr or g/day)	S-Alb (g/dl)	S-Cr (mg/ dl)	eGFR (ml/ min)	MM	Serum IEP	Urine IEP	Amy subtype	Pathological classification	TI-D	Therapy	Follow-up (month)	RS (month)	Outcome	Cause of death
1	55/F	1981	3.0	2.1	4.5	8.8				λ	Glo early (vd+)	Severe	PSL	2		D	Heart failure[a]
2	59/M	1986	6.8	3.3	3.4	15.8		IgG-λ	–	λ	Glo early (vd+)	Mod	PSL	46	HD (22)	A	
3	65/M	1987	7.2	2.5	2.5	21.5			–	κ	Glo late (vd+)	Mod	None	2		D	DIC
4	72/M	1987	1.8	4.0	1.2	46.6		IgA-λ		λ	Glo early (vd+)	Mild	MP	18		D	Heart failure[a]
5	51/F	1988	4.9	4.5	1.3	34.8	+	IgA-κ		κ	Glo early (vd−)	Mild	MP	25	HD (25)	D	Renal failure
6	60/F	1988	7.0	3.2	0.7	65.4			κ	κ	Glo early (vd+)	Mild	MP	1		D	Sudden death
7	50/M	1989	2.0	2.7	9.2	5.6		IgA-λ		λ	Glo late (vd+)	Severe	PSL	5	HD (0)	A	
8	66/F	1994	4.0	2.5	0.4	117.4		IgG-κ		κ	Vascular (gd−)	Mild	PSL	106	HD (79)	D	MDS
9	60/M	1995	8.8	3.0	1	59.9		–	λ	λ	Glo early (vd+)	Mild	MEVP	11	HD (9)	D	Hepatic failure
10	65/M	1995	5.8	1.5	1.1	52.7	+	IgA-λ	λ	λ	Glo late (vd+)	Mild	MP	82	HD (71)	D	Heart failure[a]
11	75/M	1996	3.9	2.8	2.7	19.0		IgG-λ	–	λ	Glo late (vd+)	Mod	None	2	HD (0.25)	D	Sudden death
12	49/F	1998	2.6	3.3	0.6	82.0		IgG-λ	–	λ	Glo early (vd−)	Mild	PSL, CPM	200	HD (191)	A	
13	68/M	1999	4.3	2.6	0.9	64.9			λ	λ	Glo early (vd+)	Mild	MP	179		A	
14	68/F	1999	10.0	1.6	1	42.7		IgG-λ		λ	Glo late (vd+)	Mild	MP, CPM	3		D	Sepsis
15	44/F	1999	0.1	3.2	0.9	54.3		IgG-λ	λ	λ	Vascular (gd−)	Mild	MP	17		D	Heart failure[a]
16	76/M	2000	1.6	3.4	1.2	45.9		IgA-λ		λ	Vascular (gd+)	Mod	MEVP	14		D	Heart failure[a]
17	65/F	2001	3.3	2.6	0.8	55.2		–	λ	λ	Glo early (vd+)	Mild	MP	82		D	MDS
18	76/F	2002	5.0	2.3	0.8	52.8		–		λ	Glo early (vd−)	Mild	None	10	CAPD (0.5)	D	GI bleeding
19	72/F	2003	8.0	2.0	1	42.0		IgM-λ	λ	λ	Glo late (vd+)	Mod	MP	41	HD (21)	D	Heart failure[a]
20	67/F	2003	4.4	2.6	0.8	54.7		–	κ	κ	Glo late (vd+)	Mild	MP	1		D	Heart failure
21	61/M	2004	1.9	3.5	0.9	66.9		IgG-λ		λ	Glo early (vd−)	Mild	MP	104	HD (102)	D	Heart failure
22	75/F	2004	0.7	2.8	0.8	53.0		λ	λ	λ	Glo early (vd+)	Mod	MP	2		D	Sudden death
23	69/F	2005	3.5	1.6	0.8	54.3		IgG-λ		λ	Glo late (vd+)	Mild	MP	85	HD (36)	D	Sudden death
24	68/F	2006	3.0	2.5	0.6	74.7		–	–	λ	Glo early (vd+)	Mild	MP	68		D	Heart failure
25	78/F	2006	3.6	2.3	0.6	71.8		IgA-λ	–	λ	Glo early (vd+)	Mild	MP	105		A	
26	75/M	2006	0.7	3.7	1.6	33.6	+	IgG-κ	κ	κ	Glo early (vd+)	Mod	MP	19		D	Sudden death
27	78/M	2007	3.0	3.6	0.8	70.9			–	λ	Glo early (vd+)	Mild	None	30	HD (30)	D	Renal failure
28	77/F	2007	4.7	2.4	2.2	17.4		λ	λ	λ	Glo early (vd+)	Mod	None	2	HD (2)	D	Heart failure[a]
29	68/F	2008	1.0	3.5	0.7	63.1		IgA-λ	λ	λ	Vascular (gd+)	Mild	MP	78		A	
30	57/F	2008	10.0	1.8	0.6	78.6		–	λ	λ	Glo early (vd+)	Mild	MP, VAD	16		D	Sepsis
31	64/F	2008	7.3	2.8	0.5	92.8		IgG-λ	λ	λ	Glo early (vd−)	Mild	PSL	69		A	
32	71/F	2009	5.6	1.6	0.5	90.0		IgA-λ	λ	λ	Glo early (vd−)	Mod	MP, BOR	57		A	
33	58/M	2010	10.7	3.2	1.7	33.9		IgA-λ	–	λ	Glo late (vd+)	Severe	BOR	52		A	
34	63/F	2011	5.0	2.1	4.0	9.6		IgG-λ	λ	λ	Glo late (vd+)	Mod	None	0.25		D	Heart failure

Table 1 continued

Case no.	Age/ sex	RB (year)	UP (g/gCr or g/day)	S-Alb (g/dl)	S-Cr (mg/dl)	eGFR (ml/min)	MM	Serum IEP	Urine IEP	Amy subtype	Pathological classification	TI-D	Therapy	Follow-up (month)	RS (month)	Outcome	Cause of death
35	84/M	2012	5.0	2.0	1.2	44.6		λ	λ	λ	Glo early (vd+)	Mod	MP	26		A	
36	81/F	2012	2.2	3.3	1.2	33.3		IgM-λ	λ	λ	Glo early (vd+)	Mod	MP	13		A	
37	69/F	2012	2.6	4.4	0.8	54.3	+		λ	λ	Vascular (gd−)	Mild	Dex, THAL	4		D	Heart failure[a]
38	63/F	2013	2.8	3.8	1.0	43.7	+	IgG-λ	λ	λ	Vascular (gd−)	Mild	LEN	7		D	Heart failure[a]

A alive, *Alb* albumin, *Amy* amyloidosis, *BOR* bortezomib, *CAPD* continuous ambulatory peritoneal dialysis, *CPM* cyclophosphamide, *Cr* creatinine, *D* dead, *Dex* dexamethasone, *DIC* disseminated intravascular coagulation, *eGFR* estimated glomerular filtration rate, *F* female, *gd* simultaneous glomerular deposition, *GI* gastrointestinal, *glo* glomerular, *HD* hemodialysis, *IEP* immunoelectrophoresis, *LEN* lenalidomide, *M* male, *MDS* myelodysplastic syndrome, *MEVP* melphalan + cyclophosphamide + vincristine + prednisolone, *mod* moderate, *MP* melphalan + prednisolone, *NA* not available, *PSL* prednisolone, *R-CHOP* rituximab + cyclophosphamide + doxorubicin + vincristine + prednisolone, *RB* renal biopsy, *RS* renal survival, *S* serum, *THAL* thalidomide, *TI-D* tubulo-interstitial damage, *UP* urinary protein, *VAD* vincristine + doxorubicin + dexamethasone, *vd* simultaneous vascular deposition

[a] Cardiac amyloidosis was confirmed by autopsy findings, or was suspected based on the findings of electrocardiography, ultrasound echocardiography, or myocardial scintigraphy

was the most frequently used therapy (26.6 %), followed by PSL with methotrexate (MTX) (with or without mizoribine or azathioprine) (23.3 %), PSL with gold, salazosulphapyridine, or bucillamine (16.7 %), PSL with MTX and biologics (with or without tacrolimus) (13.3 %), and others. In most of the long-term survival cases, markedly decreased serum CRP levels were confirmed after the treatments (Table 2). A case of AA amyloidosis secondary to RA, in which nephrotic syndrome was successfully treated with PSL and MTX, was reported previously [19].

Patient survival

During the follow-up period, 27 patients with AL amyloidosis (71.1 %) and 17 patients with AA amyloidosis (56.7 %) died (Table 3). The causes of death in these patients are shown in Table 5. All known causes were considered to be directly or indirectly related to the disease. In AL amyloidosis, heart failure was the most frequent cause (48.2 %) (autopsy-proven or suspected cardiac amyloidosis was 33.3 %), followed by sudden death (18.5 %), and others. In AA amyloidosis, heart failure was the most frequent cause (23.5 %) (autopsy-proven or suspected cardiac amyloidosis was 11.8 %), followed by renal failure (17.5 %) and others. The frequency of cardiac amyloidosis confirmed by autopsy or suspected by a cardiac work-up as the cause of death was significantly higher in the AL group than in the AA group ($P = 0.048$).

Patient survival curves were analyzed in the two groups of AL and AA amyloidosis, three groups classified according to eGFR at the time of renal biopsy, and three groups classified according to amyloid distribution patterns and two groups classified according to tubulo-interstitial alterations on renal biopsy specimens. Patient survival curves in AL and AA amyloidosis showed significantly longer survival in the AA group (median 89 months) than in the AL group (median 25 months) ($P = 0.018$) (Fig. 3a). Patient survival curves in groups classified according to eGFR showed significantly longer survival in patients with eGFR of >60 mL/min/1.73 m^2 (median 106 months) than in those with eGFR of 30–60 mL/min/1.73 m^2 (median 25 months) ($P = 0.012$) and eGFR of <30 mL/min/1.73 m^2 (median 42 months) ($P = 0.035$) (Fig. 3b). Patient survival curves in groups classified according to amyloid distribution patterns showed significantly longer survival in patients with the glomerular early type (median 82 months) than in those with the glomerular late type (median 20 months) ($P = 0.021$) (Fig. 3c). After separating AL and AA amyloidosis, patient survival curves in groups classified according to amyloid distribution patterns were also analyzed. In the AL group, no significant differences were observed among the glomerular early type (median 30 months), glomerular late type (median

Table 2 Clinicopathological characteristics of each patient with AA amyloidosis

Case no.	Age/sex	RB (year)	UP (g/d or g/g Cr)	S-Alb (g/dl)	S-Cr (mg/dl)	eGFR (ml/min)	Underlying disease	Pathological classification	TI-D	Therapy (Tx)	Follow-up (month)	RS (month)	Outcome	CRP (mg/dl) at RB/after Tx (months)	Cause of death
1	54/F	1984	12.9	4.2	8.3	4.5	RA	Glo early (vd+)	Severe	PSL, Gold	116	HD (2)	A	NA/NA	
2	34/F	1986	0	3.1	0.6	91.1	BD	Vascular (gd−)	Mild	PSL	339		A	6 +/0.6 (339)	
3	25/F	1994	6.0	2.2	0.8	72.7	JRA	Glo early (vd+)	Mod	PSL	61	HD (31)	D	1.9/3.8 (31)	GI bleeding
4	21/M	1994	15.5	3.2	0.5	172.8	CD	Glo early (vd+)	Mild	PSL, SASP	166	HD (141)	D	0.3/0.5 (141)	Peritnitis
5	35/F	1994	0.6	4.0	1.0	51.7	AS	Glo early (vd+)	Mild	PSL, CPM	223	HD (151)	A	4 +/0.8 (151)	
6	46/F	1995	6.2	2.3	0.9	53.6	RA	Glo late (vd+)	Mild	PSL, MTX	207	HD (150)	A	3.0/< 0.2 (148)	
7	75/F	1996	2.0	2.4	1.2	34.0	RA	Glo late (vd+)	Mod	PSL, MTX	43		D	0.6/1.0 (25)	Unknown
8	36/M	1997	1.1	4.7	2.3	27.9	KS	Vascular (gd−)	Mild	none	78	HD (68)	D	2.1/NA	Pneumonia
9	71/F	1997	1.0	3.2	1.1	38.0	RA	Glo early (vd+)	Mod	PSL	51		D	0.9/NA	Pneumonia
10	72/M	1997	6.1	1.3	1.2	46.6	unknown	Vascular (gd+)	Mod	PSL	11		D	0.8/NA	GI amyloidosis
11	65/F	1998	1.1	3.5	1.2	35.4	RA	Glo early (vd+)	Mod	PSL, MTX, MIZ	22		D	7.8/NA	Renal failure
12	61/F	2000	1.1	3.4	1.1	39.7	RA	Glo late (vd+)	Mod	PSL	20		D	2.0/NA	IP
13	25/F	2000	5.0	3.0	0.6	98.4	POS	Glo early (vd+)	Mild	PSL, CPM, CyA	173		A	6.0/0.2 (173)	
14	64/F	2000	1.9	3.4	1.9	21.5	RA	Glo early (vd+)	Mod	PSL, BUC	34		A	3.6/NA	
15	60/M	2000	1.2	2.6	1.6	35.8	RA	Glo early (vd+)	Mod	PSL, BUC	20		D	3.0/NA	Heart failure
16	77/F	2000	5.8	2.9	7.2	4.8	RA	Glo late (vd+)	Severe	none	6	HD (0)	D	0.7/NA	Renal failure
17	67/F	2000	2.8	2.9	1.8	22.5	RA	Glo early (vd+)	Mod	PSL, MTX, MIZ	110		D	2.7/0.0 (87)	Renal failure
18	53/M	2001	0.7	3.0	2.3	25.0	SAR	Vascular (gd+)	Mod	PSL	7	HD (5)	D	1.0/NA	Sepsis
19	61/F	2003	0	NA	2.1	19.6	RA	Vascular (gd−)	Mod	PSL, MTX	42		D	0.3/0.2 (38)	IP

Table 2 continued

Case no.	Age/sex	RB (year)	UP (g/d or g/g Cr)	S-Alb (g/dl)	S-Cr (mg/dl)	eGFR (ml/min)	Underlying disease	Pathological classification	TI-D	Therapy (Tx)	Follow-up (month)	RS (month)	Outcome	CRP (mg/dl) at RB/after Tx (months)	Cause of death
20	63/F	2003	2.8	2.6	2.5	16.0	RA	Glo early (vd+)	Mod	PSL, AZP, MTX	139		A	0.5/<0.2 (139)	
21	58/F	2003	1.3	3.1	2.6	15.7	RA	Glo late (vd+)	Mod	PSL, BUC	48	HD (32)	D	1.6/0.4 (31)	Heart failure[a]
22	54/F	2004	6.2	2.5	0.8	58.2	RA	Glo early (vd+)	Mild	PSL, MTX, TAC, BIO	122	HD (31)	A	0.3/0.1 (29)	
23	64/F	2004	0	4.4	1.0	43.5	RA	Vascular (gd+)	Mild	PSL, SASP, MTX, BIO	123		A	3.4/0.0 (123)	
24	64/F	2004	0.4	3.3	0.5	92.8	RA	Glo early (vd+)	Mild	PSL, MTX, BIO	89		D	3.1/0.1 (88)	Heart failure
25	47/F	2005	10.0	2.1	0.8	60.6	RA	Vascular (gd+)	Mild	PSL, MTX	88		A	3.1/0.2 (88)	
26	70/F	2007	2.4	2.6	1.6	25.3	IIBD	Vascular (gd+)	Mod	PSL	75		A	1.5/0.1 (75)	
27	64/M	2007	3.5	3.1	3.0	17.7	RA	Glo late (vd+)	Mod	PSL	1		D	2.4/NA	Heart failure[a]
28	35/M	2008	2.2	3.0	1.7	39.1	CD	Glo early (vd+)	Mod	MSL, BIO	76		A	0.1/0.1 (72)	
29	38/F	2008	2.7	1.5	1.1	45.5	CD	Glo early (vd+)	Mild	PSL, MSL, BIO	37	HD (22)	D	4.7/0.9 (36)	Sepsis
30	59/F	2010	0.1	3.3	1.4	30.8	RA	Vascular (gd+)	Severe	PSL, MTX, BIO	46		A	4.8/0.1 (46)	

A alive, *Alb* albumin, *AS* aortitis syndrome, *AZP* azathioprine, *BD* Behçet's disease, *BIO* biologics, *BUC* bucillamine, *CD* Crohn's disease, *CPM* cyclophosphamide, *Cr* creatinine, *CyA* cyclosporine A, *D* dead, *eGFR* estimated glomerular filtration rate, *F* female, *GI* gastrointestinal, *gd* simultaneous glomerular deposition, *glo* glomerular, HD hemodialysis, *IIBD* indeterminate inflammatory bowel disease, *IP* interstitial pneumonia, *JRA* juvenile rheumatoid arthritis, *KS* Kartagener's syndrome, *M* male, *MIZ* mizoribine, *mod* moderate, *MSL* mesalazine, *MTX* methotrexate, *NA* not available, *POS* polyangiitis overlap syndrome, *PSL* prednisolone, *RA* rheumatoid arthritis, *RB* renal biopsy, *RS* renal survival, *S* serum, *SAR* sarcoidosis, *SASP* salazosulphapyridine, *TAC* tacrolimus, *TI-D* tubulo-interstitial damage, *Tx* therapy, *UP* urinary protein, *vd* simultaneous vascular deposition

[a] Cardiac amyloidosis was suspected based on the findings of electrocardiography or ultrasound echocardiography

Table 3 Characteristics at renal biopsy, treatments, and outcomes of patients with AL and AA amyloidosis

	AL	AA	*P* value
Number of patients (%)	38 (55.9)	30 (44.1)	
Characteristics at renal biopsy			
Age (years) [median (range)]	68 (44–84)	59 (21–77)	0.001**
Gender (male/female)	14/24	7/23	0.231***
Proteinuria (g/gCr or g/day) [median (range)]	4.0 (0.1–10.7)	2.1 (0–15.5)	0.028**
Serum albumin (g/dL) [mean ± SD (range)]	2.8 ± 0.8 (1.5–4.5)	3.0 ± 0.8 (1.3–4.7)	0.327*
Serum Cr (mg/dL) [median (range)]	1.0 (0.4–9.2)	1.2 (0.5–8.3)	0.086**
eGFR (mL/min/1.73 m^2) [median (range)]	52.9 (5.6–117.4)	36.9 (4.5–172.8)	0.099**
>60 [*n* (%)]	12 (31.6 %)	6 (20.0 %)	0.283***
30–60 [*n* (%)]	19 (50.0 %)	13 (43.3 %)	0.584***
30 [*n* (%)]	7 (18.4 %)	11 (36.7 %)	0.090***
Amyloid distribution [*n* (%)]			
Glomerular early type	22 (57.9 %)	15 (55.0 %)	0.516***
Glomerular late type	10 (26.3 %)	6 (20.0 %)	0.542***
Vascular type	6 (15.8 %)	9 (30.0 %)	0.161***
Interstitial type	0	0	
Tubulo-interstitial damage			
Mild	23 (60.5 %)	11 (36.7 %)	0.051***
Moderate	12 (31.6 %)	16 (53.3 %)	0.660***
Severe	3 (7.9 %)	3 (10.0 %)	0.761***
Moderate + severe	15 (39.5 %)	19 (63.3 %)	0.051***
Monoclonal proteins in AL amyloidosis			
Positive serum monoclonal proteins [*n* (%)]	26 (68.4 %)		
IgG-λ	10 (38.5 %)		
IgA-λ	8 (30.8 %)		
λ	3 (11.5 %)		
IgG-κ	2 (7.7 %)		
IgM-λ	2 (7.7 %)		
IgA-κ	1 (3.8 %)		
Positive urinary monoclonal proteins [*n* (%)]	19 (50.0 %)		
λ	16 (84.2 %)		
κ	3 (15.8 %)		
Amyloid typing on biopsy specimens [*n* (%)]			
λ	32 (84.2 %)		
κ	6 (15.8 %)		
Treatments and outcomes			
Observation period (months) [median (range)]	18 (0.25–200)	61 (1–339)	0.003**
Treatments [*n* (%)]			
Steroids	31 (81.6 %)	27 (90.0 %)	0.330***
MP-based	20 (52.6 %)		
PSL only	5 (13.1 %)	8 (26.6 %)	
PSL + MTX (±MIZ or AZP)		7 (23.3 %)	
PSL + Gold or SASP or BUC		5 (16.7 %)	
PSL + MTX + BIO (±SASP or TAC)		4 (13.3 %)	
THAL (+Dex)/LEN	2 (5.3 %)		
BOR-based	2 (5.3 %)		
MEVP	2 (5.3 %)		
MSL + BIO (±PSL)		2 (6.7 %)	

Table 3 continued

	AL	AA	*P* value
PSL + CPM (±CyA)	1 (2.6 %)	2 (6.7 %)	
None	6 (15.8 %)	2 (6.7 %)	
ESRD requiring dialysis [*n* (%)]	14 (36.8 %)	11 (36.7 %)	0.988***
Death [*n* (%)]	27 (71.1 %)	17 (56.7 %)	0.218***

AZP azathioprine, *BIO* biologics, *BOR* bortezomib, *BUC* bucillamine, *CPM* cyclophosphamide, *Cr* creatinine, *CyA* cyclosporine A, *Dex* dexamethasone, *eGFR* estimated glomerular filtration rate, *ESRD* end-stage renal disease, *LEN* lenalidomide, *MEVP* melphalan + cyclophosphamide + vincristine + prednisolone, *MIZ* mizoribine, *MP* melphalan + prednisolone, *MSL* mesalazine, *MTX* methotrexate, *n* number, *PSL* prednisolone, *R-CHOP* rituximab + cyclophosphamide + doxorubicin + vincristine + prednisolone, *SASP* salazosulphapyridine, *SD* standard deviation, *TAC* tacrolimus, *THA*L thalidomide

* The Student's *t* test, ** Mann–Whitney *U* test, *** χ^2 test

3 months), and vascular type (median 14 months) (Fig. 3d). In the AA group, survival was slightly longer in the glomerular early type (median 110 months) than in the glomerular late type (median 20 months) ($P = 0.053$) (Fig. 3e). Patient survival curves in groups classified according to tubulo-interstitial damage showed slightly longer survival in patients with mild damage (median 82 months) than in those with moderate to severe damage (median 42 months) ($P = 0.084$) (Fig. 3f).

A multivariate Cox regression analysis of factors influencing patient survival in the overall population of patients showed that cardiac amyloidosis [hazard ratio (HR) 2.56, $P = 0.024$] and steroid therapy (HR 0.40, $P = 0.042$) significantly influenced survival, while the types of amyloidosis (AL/AA), age, proteinuria, eGFR, amyloid distribution, and tubulo-interstitial damage did not (Table 6).

Renal survival

Renal outcomes were calculated using the time interval between the diagnosis and beginning of dialysis, and all patients who died before reaching ESRD were regularly censored, as in the study by Bergesio et al. [8].

During the follow-up period, 14 patients with AL amyloidosis (36.8 %) and 11 patients with AA amyloidosis (36.7 %) developed ESRD requiring dialysis therapy (Table 3). All patients were on hemodialysis, except for 1 patient with AL amyloidosis who was on peritoneal dialysis (Tables 1, 2). In 2 patients (1 AL and 1 AA), dialysis was started before renal biopsy (Tables 1, 2). These 2 patients were excluded from the renal outcome analysis.

Renal survival (censored for death) curves were analyzed in two groups of AL and AA amyloidosis, three groups classified according to eGFR at the time of renal biopsy, and three groups classified according to amyloid distribution patterns and two groups classified according to tubulo-interstitial alterations on renal biopsy specimens. Renal survival curves in AL and AA amyloidosis showed significantly longer survival in the AA group (median 166 months) than in the AL group (median 85 months) ($P = 0.025$) (Fig. 4a). Renal survival curves in groups classified according to eGFR showed significantly longer survival in patients with eGFR of >60 mL/min/1.73 m^2 (median 200 months) than in those with eGFR of <30 mL/min/1.73 m^2 (median 78 months) ($P = 0.030$) (Fig. 4b). Renal survival curves in groups classified according to amyloid distribution patterns showed no significant differences among the glomerular early type (median 122 months), glomerular late type (median 82 months), and vascular type (median: undetermined) (Fig. 4c). After separating AL and AA amyloidosis, renal survival curves in groups classified according to amyloid distribution patterns were also analyzed. In the AL group, no significant differences were observed among the glomerular early type (median 104 months), glomerular late type (82 months), and vascular type (106 months) (Fig. 4d). In the AA group, no significant differences were noted among the glomerular early type (median 166 months), glomerular late type (median 48 months), and vascular type (median: undetermined) (Fig. 4e). Renal survival curves in groups classified according to tubulo-interstitial damage showed no significant difference between patients with mild damage (median 122 months) and those with moderate to severe damage (median 116 months) (Fig. 4f).

A multivariate Cox regression analysis of factors influencing renal survival in the overall population of patients showed that cardiac amyloidosis (HR 4.18, $P = 0.046$) and steroid therapy (HR 0.18, $P = 0.012$) significantly influenced survival, while the types of amyloidosis (AL/AA), age, proteinuria, eGFR, amyloid distribution, and tubulo-interstitial damage did not (Table 7).

Discussion

In the present study, the long-term prognosis of 68 cases of biopsy-proven renal amyloidosis (38 AL and 30 AA) diagnosed at a single Japanese center was analyzed. The

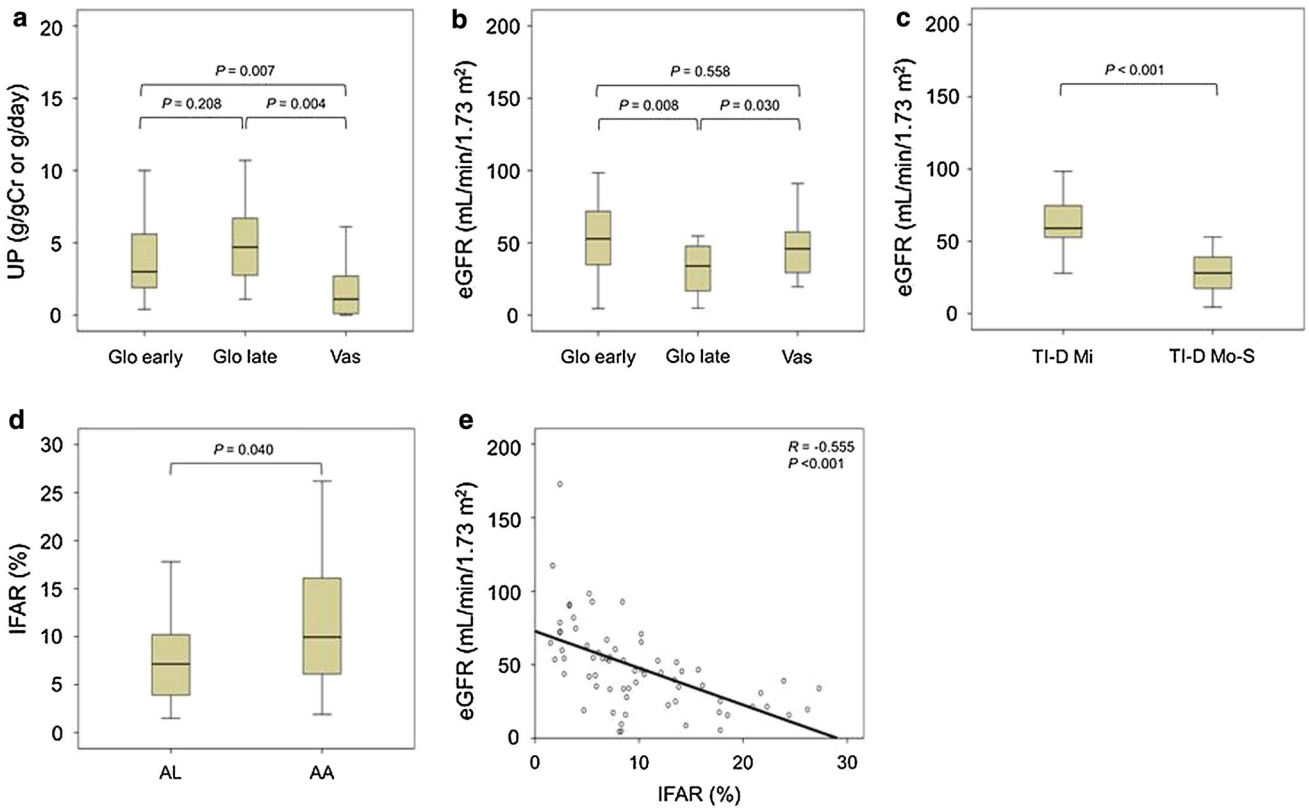

Fig. 2 **a** Urinary protein (UP) levels according to amyloid distribution patterns: the glomerular (glo) early, glomerular (glo) late, and vascular (vas) types. *Box* and *whisker plots* are shown. The *boxes* represent the 25–75th percentiles, whereas the *horizontal lines* within each *box* represent the median values. The *whiskers* represent the lowest and highest values in the 25th percentile minus the 1.5 interquartile range and 75 percentile plus 1.5 interquartile range regions, respectively. Glo early vs. vascular, $P = 0.007$; glo late vs. vascular, $P = 0.004$. **b** eGFR according to amyloid distribution patterns: the glomerular (glo) early, glomerular (glo) late, and vascular (vas) types. *Box* and *whisker plots* are shown. The *boxes* represent the 25–75th percentiles, whereas the *horizontal lines* within each *box* represent the median values. The *whiskers* represent the lowest and highest values in the 25th percentile minus 1.5 interquartile range and 75 percentile plus 1.5 interquartile range regions, respectively. Glo early vs. glo late, $P = 0.008$; Glo late vs. vascular, $P = 0.030$. **c** eGFR according to tubulo-interstitial damage (TI-D) categories: mild (Mi) and moderate to severe (Mo-S). *Box* and *whisker plots* are shown. The *boxes* represent the 25–75th percentiles, whereas the *horizontal lines* within each *box* represent the median values. The *whiskers* represent the lowest and highest values in the 25th percentile minus 1.5 interquartile range and 75 percentile plus 1.5 interquartile range regions, respectively. Mi vs. Mo-S, $P < 0.001$. **d** The interstitial fibrosis area ratio (IFAR) according to the type of amyloidosis. *Box* and *whisker plots* are shown. The *boxes* represent the 25–75th percentiles, whereas the *horizontal lines* within each *box* represent the median values. The *whiskers* represent the lowest and highest values in the 25th percentile minus 1.5 interquartile range and 75 percentile plus 1.5 interquartile range regions, respectively. AL vs. AA, $P = 0.040$. **e** Relationship between IFAR and eGFR. $R = -0.555$, $P < 0.001$

most important feature in our cohort was that most patients received treatments for the underlying diseases of amyloidosis. Our results showed significantly longer survival in the AA group than in the AL group, and also identified eGFR of >60 mL/min/1.73 m^2 at renal biopsy and an early histological stage of glomerular amyloid deposition as low-risk factors. In addition, the extent of tubulo-interstitial damage including interstitial fibrosis correlated with renal function at biopsy. Our multivariate analysis showed that cardiac amyloidosis and steroid therapy significantly influenced patient and renal survival in the overall population, whereas the different types of amyloidosis (AL/AA), age, proteinuria, and eGFR did not. On the other hand, in the cohort of Bergesio et al. [8], their multivariate analysis of factors affecting patient survival showed that age, heart involvement, serum Cr, and specific therapy correlated with survival in the overall population or in the AL group.

Establishing the type of amyloidosis in renal biopsies is essential for initiating an appropriate treatment. The recent recommendation is that AA immunostaining needs to be routinely performed, unless there is strong and selective staining of amyloid deposits for the same light chain found in the monoclonal spike in the serum and urine [20]. In the present study, the typing of amyloidosis in stored biopsy specimens was performed according to this

recommendation. In cases of suspected AL amyloidosis, the typing of renal amyloidosis was performed using direct immunofluorescence on frozen tissue and immunohistochemistry on paraffin-embedded tissue with commercially available antibodies against the κ and λ light chains. In some inconclusive cases, immunohistochemical studies were performed with useful antibodies for the typing of AL amyloidosis [11, 12].

The rates of the primary type and myeloma-associated type as underlying diseases in AL amyloidosis in our cohort were similar to those in a previous study from Japanese institutes [9]. On the other hand, the rate of RA as an underlying disease in AA amyloidosis in our cohort (63.6 %) was lower than those (91.2–97.1 %) in previous studies from Japanese institutes [4, 9], and was similar to that (50.6 % in cases excluding unknown cases) in a previous study from Italian institutes [3]. This may be due to referral bias and/or a recent change in the spectrum of underlying diseases leading to AA amyloidosis in Japan.

In our cohort, significant differences were observed in several clinicopathological features in the AL and AA groups. Median age was higher in the AL group than in the AA group. Proteinuria levels were higher in the AL group than in the AA group, while there was no significant difference in eGFR between the two groups. In addition, we found that tubulo-interstitial damage was slightly more severe in the AA group than in the AL group in our cohort. IFAR associated with chronic tubulo-interstitial injury was significantly higher in the AA group than in the AL group. This may partly be due to the use of non-steroidal anti-inflammatory drugs for patients with chronic inflammatory diseases in the AA group.

Few studies have compared patients with AL and AA renal amyloidosis in the same cohort. Bergesio et al. [8] reported the survival data of 290 cases of renal amyloidosis in an Italian collaborative study cohort in 2008. In their cohort, 97 out of 167 patients with AL amyloidosis (58.1 %) and 51 out of 86 patients with AA amyloidosis (59.3 %) did not receive specific treatments for the disease. The median survivals of patients with AL and AA amyloidosis were 37 and 79 months, respectively. The median renal survivals (censored for death) of patients with AL and AA amyloidosis were 45 and 33 months, respectively. Regarding the prognosis of the Japanese cohort of AL and AA renal amyloidosis, Sasatomi et al. [9] published the survival data of 97 patients (52 AL and 45 AA) in 2007; however, detailed information on treatments was not described. Their analysis on patient survival and renal survival (mean duration of the follow-up after renal biopsy: approximately 25 months) revealed no significant differences between the AL and AA groups, and the survival rates in both groups were poor.

In our cohort, 32 out of 38 patients with AL amyloidosis (84.2 %) and 28 out of 30 patients with AA amyloidosis (93.3 %) received treatments for the underlying diseases of amyloidosis. In patients with AL amyloidosis, melphalan/PSL-based therapy was the most frequently used therapy, followed by PSL monotherapy and new agent-based

Table 4 Underlying diseases in patients with AL and AA amyloidosis

	Number of patients (%)
AL amyloidosis	38
Primary amyloidosis	26 (68.4 %)
Myeloma-associated	5 (13.2 %)
IgM paraproteinemia-associated	2 (5.3 %)
Heavy chain deposition disease	1 (2.6 %)
Unknown	4 (10.5 %)
AA amyloidosis	30
Rheumatoid arthritis	19 (63.6 %)
Crohn's disease	3 (10.0 %)
Juvenile rheumatoid arthritis	1 (3.3 %)
Behçet's disease	1 (3.3 %)
Kartagener's syndrome	1 (3.3 %)
Aortitis syndrome	1 (3.3 %)
Polyangiitis overlap syndrome	1 (3.3 %)
Sarcoidosis	1 (3.3 %)
Indeterminate inflammatory bowel disease	1 (3.3 %)
Unknown	1 (3.3 %)

therapy. In patients with AA amyloidosis (63.6 % of patients had RA), PSL monotherapy was the most frequently used therapy, followed by PSL with disease-modifying anti-rheumatic drugs (DMARDs) including MTX and biologics. In our study, the median survivals of patients with AL and AA amyloidosis were 25 and 89 months, respectively. The median renal survivals (censored for death) of patients with AL and AA amyloidosis were 85 and 166 months, respectively. The survival of patients with AA amyloidosis appeared to be markedly better in our study than in previously reported cohorts [7, 8, 21]. In our AA type cases, most of the unsuccessfully controlled patients with PSL monotherapy were treated with combination therapies including MTX and biologics. In most long-term survival cases, we confirmed that serum CRP levels markedly decreased after these treatments. Therefore, we suggest that the development of treatment strategies for underlying inflammatory diseases (particularly for RA and inflammatory bowel disease) in AA amyloidosis led to improved survival. We previously described a patient with AA renal amyloidosis accompanied by RA, in whom the remission of nephrotic syndrome was achieved by PSL and MTX therapy [19]. Ueno et al. [22] also suggested that intensive therapeutic interventions with DMARDs and biologics have the potential to change the histologically predicted prognosis of RA-associated renal AA amyloidosis.

In contrast, the prognosis of patients with AL amyloidosis in our cohort was as poor as in other cohorts [5, 6, 8]. Based on the findings of an Italian collaborative study, Bergesio et al. [8] emphasized that heart involvement and specific treatments were the main factors affecting survival in patients with AL amyloidosis, and that an early diagnosis together with the wider application of current therapies for AL amyloidosis is needed. In our cohort, heart failure due to cardiac amyloidosis was the main cause of death in the AL group. Our multivariate analysis also showed that heart involvement and steroid therapy significantly influenced patient and renal survivals in the overall population. A recent study on large cohorts of patients with AL amyloidosis and renal involvement by Palladini et al. [6] showed that the progression of renal dysfunction was actually predicted by baseline proteinuria and eGFR, and suggested the existence of an "early stage" of renal damage defined by proteinuria $\leq$5 g/day and eGFR $\geq$50 mL/min/1.73 m^2. They also emphasized the need for an early diagnosis to identify and treat patients with AL amyloidosis before organ damage ensued. Our analysis identified eGFR of >60 mL/min/1.73 m^2 at biopsy and an early histological stage of glomerular amyloid deposition as low risk factors. Thus, intensive therapeutic interventions need to be considered, particularly for these cases.

IgM-associated AL amyloidosis was recently proposed as a rare (approximately 7 %), but distinct subtype of AL amyloidosis, with less advanced organ dysfunction: treatments with melphalan/dexamethasone may be effective for this entity [23]. In our cohort of 38 patients with AL renal amyloidosis, 2 patients (5.3 %) had IgM paraproteinemia.

Table 5 Causes of death in patients with AL and AA amyloidosis

	AL [*n* (%)]	AA [*n* (%)]	*P* value (χ^2 test)
Death [*n*]	27 (71.7 %)	17 (56.7 %)	
Causes of death			
Heart failure/cardiac amyloidosis[a]	13 (48.2 %)/9 (33.3 %)	4 (23.5 %)/2 (11.8 %)	0.048/0.048
Sudden death	5 (18.5 %)		
Renal failure	2 (7.4 %)	3 (17.5 %)	
Myelodysplastic syndrome	2 (7.4 %)		
Sepsis	1 (3.7 %)	2 (11.8 %)	
Pneumonia		2 (11.8 %)	
Interstitial pneumonia		2 (11.8 %)	
Gastrointestinal disease	1 (3.7 %)	2 (11.8 %)	
Cerebral infarction	1 (3.7 %)		
Hepatic failure	1 (3.7 %)		
Disseminated intravascular coagulation	1 (3.7 %)		
Peritonitis		1 (5.9 %)	
Unknown (death in hospital)		1 (5.9 %)	

n number

[a] Confirmed by autopsy or suspected by a cardiac work-up

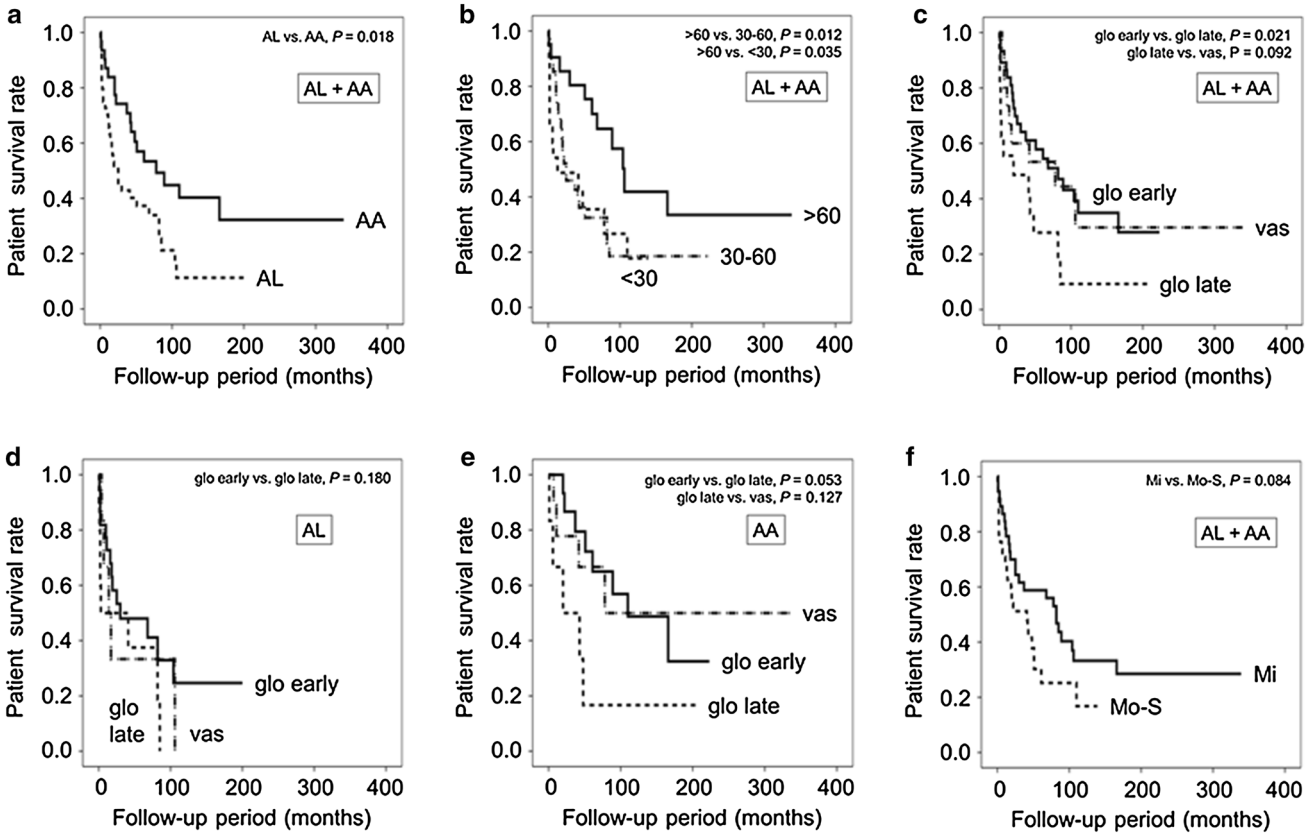

Fig. 3 **a** Patient survival of 38 patients with AL amyloidosis (median 25 months) and of 30 patients with AA amyloidosis (median 89 months). AL vs. AA, $P = 0.018$. **b** Patient survival of 18 patients with eGFR of >60 mL/min/1.73 m^2 (median 106 months), 32 patients with eGFR of 30–60 mL/min/1.73 m^2 (median 25 months), and 18 patients with eGFR of <30 mL/min/1.73 m^2 (median 42 months). >60 vs. 30–60, $P = 0.012$; >60 vs. <30, $P = 0.035$. **c** Patient survival of 37 patients with the glomerular (glo) early type (median 82 months), 16 patients with the glomerular (glo) late type (median 20 months), and 15 patients with the vascular (vas) type (median 78 months). Glo early vs. glo late, $P = 0.021$. **d** Patient survival of 38 patients with AL amyloidosis: 22 patients with the glomerular (glo) early type (median 30 months), 10 patients with the glomerular (glo) late type (median 3 months), and 6 patients with the vascular (vas) type (median 14 months). Glo early vs. glo late, $P = 0.180$. **e** Patient survival of 30 patients with AA amyloidosis: 15 patients with the glomerular (glo) early type (median 110 months), 6 patients with the glomerular (glo) late type (median 20 months), and 9 patients with the vascular (vas) type (median 78 months). Glo early vs. glo late, $P = 0.053$. **f** Patient survival of 34 patients with mild (Mi) tubulo-interstitial damage (median 82 months) and 34 patients with moderate to severe (Mo-S) tubulo-interstitial damage (median 42 months). Mi vs. Mo-S, $P = 0.084$

Table 6 Multivariate Cox regression analysis of factors affecting patient survival

	Overall population (AL + AA)	
	HR (95 % CI)	*P* value
Type (AL versus AA)	1.94 (0.77–4.88)	0.157
Age (10 years old)	1.14 (0.84–1.55)	0.403
Proteinuria (g/gCr or g/day)	0.94 (0.83–1.06)	0.330
eGFR (mL/min/1.73 m^2)	1.00 (0.98–1.01)	0.659
Amyloid distribution (glomerular late type)	1.40 (0.64–3.06)	0.405
Tubulo-interstitial damage (moderate + severe)	1.67 (0.69–4.07)	0.256
Cardiac amyloidosis[a]	2.56 (1.13–5.79)	0.024
Therapy (steroids)	0.40 (0.17–1.00)	0.042

CI confident interval, *Cr* creatinine, *eGFR* estimated glomerular filtration rate, *HR* hazard ratio

[a] Confirmed by autopsy or suspected by a cardiac work-up

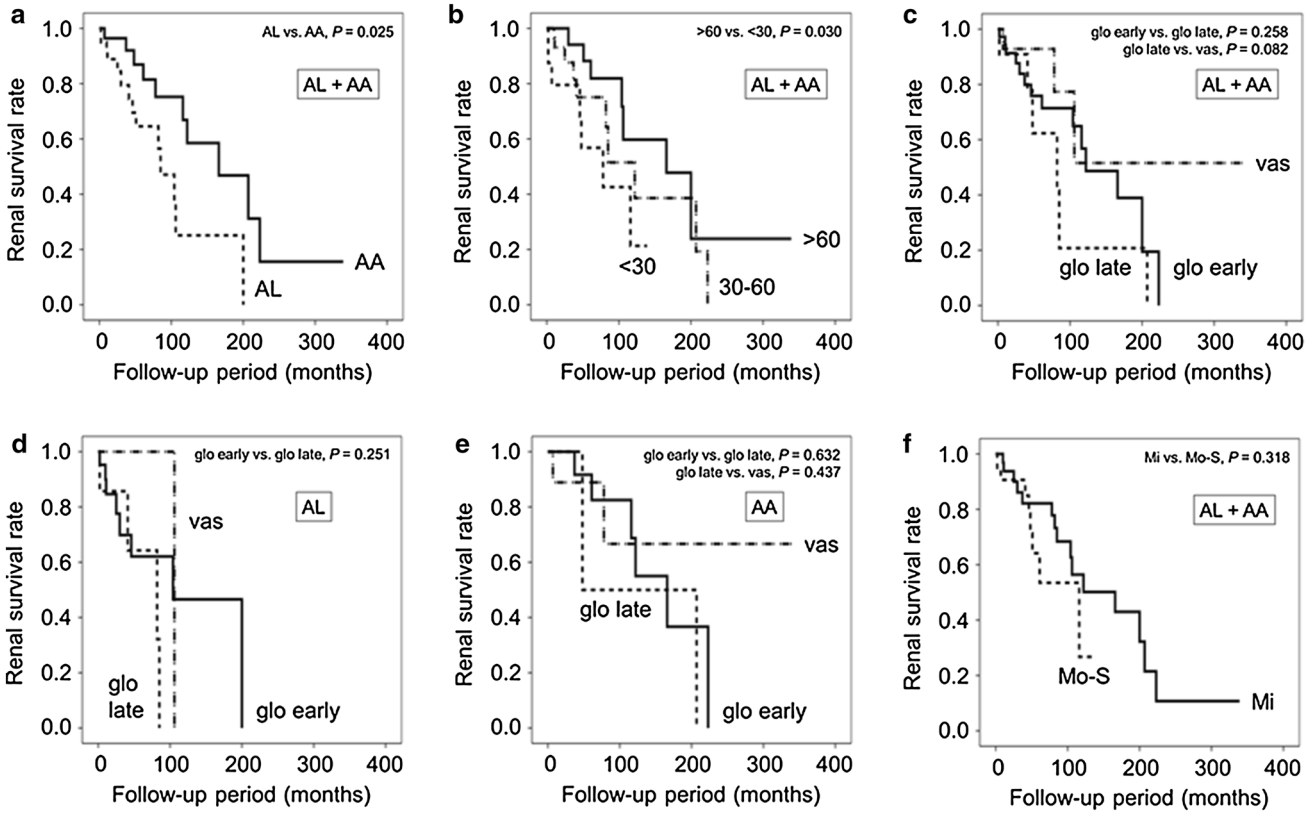

Fig. 4 **a** Renal survival (censored for death) of 37 patients with AL amyloidosis (median 85 months) and of 29 patients with AA amyloidosis (median 166 months). AL vs. AA, $P = 0.025$. **b** Renal survival (censored for death) of 18 patients with eGFR of >60 mL/min/1.73 m^2 (median 200 months), 32 patients with eGFR of 30–60 mL/min/1.73 m^2 (median 122 months), and 16 patients with eGFR of <30 mL/min/1.73 m^2 (median 78 months). >60 vs. <30, $P = 0.030$. **c** Renal survival (censored for death) of 37 patients with the glomerular (glo) early type (median 122 months), 14 patients with the glomerular (glo) late type (median 82 months), and 15 patients with the vascular (vas) type (median: undetermined). Glo early vs. glo late, $P = 0.258$. Glo late vs. vas, $P = 0.082$. **d** Renal survival (censored for death) of 37 patients with AL amyloidosis: 22 patients with the glomerular (glo) early type (median 104 months), 9 patients with the glomerular (glo) late type (median 82 months), and 6 patients with the vascular (vas) type (median 106). Glo early vs. glo late, $P = 0.251$. **e** Renal survival (censored for death) of 29 patients with AA amyloidosis: 15 patients with the glomerular (glo) early type (median 166 months), 5 patients with the glomerular (glo) late type (median 48 months), and 9 patients with the vascular (vas) type (median: undetermined). Glo early vs. glo late, $P = 0.632$. **f** Renal survival (censored for death) of 34 patients with mild (Mi) tubulo-interstitial damage (median 122 months) and 32 patients with moderate to severe (Mo-S) tubulo-interstitial damage (median 116 months). Mi vs. Mo-S, $P = 0.318$

Table 7 Multivariate Cox regression analysis of factors affecting renal survival

	Overall population (AL + AA)	
	HR (95 % CI)	*P* value
Type (AL versus AA)	3.21 (0.78–13.1)	0.105
Age (10 years old)	0.94 (0.58–1.54)	0.809
Proteinuria (g/gCr or g/day)	1.11 (0.97–1.27)	0.145
eGFR (mL/min/1.73 m^2)	0.99 (0.97–1.01)	0.222
Amyloid distribution (glomerular late type)	0.69 (0.20–2.33)	0.546
Tubulo-interstitial damage (moderate + severe)	0.99 (0.26–3.86)	0.990
Cardiac amyloidosis[a]	4.18 (1.03–17.04)	0.046
Therapy (steroids)	0.18 (0.05–0.68)	0.012

CI confident interval, *Cr* creatinine, *eGFR* estimated glomerular filtration rate, *HR* hazard ratio

[a] Confirmed by autopsy or suspected by a cardiac work-up

These patients were treated with melphalan/PSL, and 1 patient with the glomerular late type developed ESRD 21 months after the diagnosis.

There were several limitations in our study. This was a retrospective cohort study with a relatively small number of patients. Only seven clinicopathological variables (types of amyloidosis, age, proteinuria, eGFR, glomerular amyloid distribution, tubulo-interstitial damage, and cardiac amyloidosis) and one treatment (steroid therapy) were used as adjustments (covariates) in our multivariate Cox regression analysis. These adjustments were selected based on the results of the log-rank test (univariate analysis) in our study and the findings of the previous study by Bergesio et al. [8]. There were no standard treatment strategies for renal amyloidosis at the time of these studies. Prognostic factors for renal amyloidosis need to be determined by prospective cohort studies based on international therapeutic guidelines in the future.

In summary, our results point to an early diagnosis and treatments for underlying diseases as the main factors affecting the prognosis of patients with renal amyloidosis. The survival of patients with AA renal amyloidosis appears to have been greatly improved by therapeutic advances. Current treatment approaches for the eradication of clonal plasma cells are also expected to improve the prognosis of patients with AL renal amyloidosis.

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Compliance with ethical standards

Conflict of interest The authors declare that no conflict of interest exists.

References

1. Dember LM. Amyloidosis-associated kidney disease. J Am Soc Nephrol. 2006;17:3458–71.
2. Said SM, Sethi S, Valeri AM, Leung N, Cornell LD, Fidler ME, et al. Renal amyloidosis: origin and clinicopathologic correlations of 474 recent cases. Clin J Am Soc Nephrol. 2013;8:1515–23.
3. Bergesio F, Ciciani AM, Santostefano M, Brugnano R, Manganaro M, Palladini G, et al. Renal involvement in systemic amyloidosis—an Italian retrospective study on epidemiological and clinical data at diagnosis. Nephrol Dial Transplant. 2007;22:1608–18.
4. Nishi S, Alchi B, Imai N, Gejyo F. New advances in renal amyloidosis. Clin Exp Nephrol. 2008;12:93–101.
5. Pinney JH, Lachmann HJ, Bansi L, Wechalekar AD, Gilbertson JA, Rowczenio D, et al. Outcome in renal AL amyloidosis after chemotherapy. J Clin Oncol. 2011;29:674–81.
6. Palladini G, Hegenbart U, Milani P, Kimmich C, Foli A, Ho AD, et al. A staging system for renal outcome and early markers of renal response to chemotherapy in AL amyloidosis. Blood. 2014;124:2325–32.
7. Ahbap E, Kara E, Sahutoglu T, Basturk T, Koc Y, Sakaci T, et al. Outcome of 121 patients with renal amyloid A amyloidosis. J Res Med Sci. 2014;19:644–9.
8. Bergesio F, Ciciani AM, Manganaro M, Palladini G, Santostefano M, Brugnano R, et al. Renal involvement in systemic amyloidosis: an Italian collaborative study on survival and renal outcome. Nephrol Dial Transplant. 2008;23:941–51.
9. Sasatomi Y, Sato H, Chiba Y, Abe Y, Takeda S, Ogahara S, et al. Prognostic factors for renal amyloidosis: a clinicopathological study using cluster analysis. Intern Med. 2007;46:213–9.
10. Hopfer H, Wiech T, Mihatsch MJ. Renal amyloidosis revisited: amyloid distribution, dynamics and biochemical type. Nephrol Dial Transplant. 2011;26:2877–84.
11. Hoshii Y, Setoguchi M, Iwata T, Ueda J, Cui D, Kawano H, et al. Useful polyclonal antibodies against synthetic peptides corresponding to immunoglobulin light chain constant region for immunohistochemical detection of immunoglobulin light chain amyloidosis. Pathol Int. 2001;51:264–70.
12. Hoshii Y, Kiyama M, Cui D, Kawano H, Ishihara T. Immunohistochemical study of immunoglobulin light chain amyloidosis with antibodies to the immunoglobulin light chain variable region. Pathol Int. 2006;56:324–30.
13. Matsuo S, Imai E, Horio M, Yasuda Y, Tomita K, Nitta K, et al. Revised equations for estimated GFR from serum creatinine in Japan. Am J Kidney Dis. 2009;53:982–92.
14. Rajkumar SV, Dimopoulos MA, Palumbo A, Blade J, Merlini G, Mateos MV, et al. International Myeloma Working Group updated criteria for the diagnosis of multiple myeloma. Lancet Oncol. 2014;15:e538–48.
15. Komatsuda A, Maki N, Wakui H, Ohtani H, Hatakeyama T, Yasuda T, et al. Development of systemic λ-light chain amyloidosis in a patient with γ-heavy chain deposition disease during long-term follow-up. Nephrol Dial Transplant. 2005;20:434–7.
16. Komatsuda A, Kimura H, Ichikawa Y, Ohtani H, Wakui H, Imai H. Nephrotic syndrome in a patient with renal amyloidosis due to polyangiitis overlap syndrome. Nephrol Dial Transplant. 2002;17:669–71.
17. Komatsuda A, Wakui H, Ohtani H, Maki N, Nimura T, Takatsu H, et al. Amyloid A-type renal amyloidosis in a patient with sarcoidosis: report of a case and review of the literature. Clin Nephrol. 2003;60:284–8.
18. Hatakeyama T, Komatsuda A, Matsuda A, Togashi M, Maki N, Masai R, et al. Renal amyloidosis associated with extracapillary glomerulonephritis and vasculitis in a patient with inflammatory bowel disease treated with infliximab. Clin Nephrol. 2008;70:240–4.
19. Komatsuda A, Morita K, Ohtani H, Yamaguchi A, Miura AB. Remission of the nephrotic syndrome in a patient with renal amyloidosis due to rheumatoid arthritis treated with prednisolone and methotrexate. Am J Kidney Dis. 1998;32:E7.
20. Satoskar AA, Burdge K, Cowden DJ, Nadasdy GM, Herbert LA, Nadasdy T. Typing of amyloidosis in renal biopsies: diagnostic pitfalls. Arch Pathol Lab Med. 2007;131:917–22.
21. Joss N, McLaughlin K, Simpson K, Boulton-Jones JM. Presentation, survival and prognostic markers in AA amyloidosis. Q J Med. 2000;95:535–42.
22. Ueno T, Takeda K, Nagata M. Remission of proteinuria and prevention of renal function in patients with renal AA amyloidosis secondary to rheumatoid arthritis. Nephrol Dial Transplant. 2012;27:633–9.
23. Palladini G, Russo P, Bosoni T, Sarais G, Lavatelli F, Foli A, et al. AL amyloidosis associated with IgM monoclonal protein: a distinct clinical entity. Clin Lymphoma Myeloma. 2009;9:80–3.