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Abstract

Objective. Renal involvement is a rare event during primary SS (pSS). We aimed to describe the clinico-biological and histopathological characteristics of pSS-related nephropathy and its response to treatment.

Methods. We conducted a French nationwide, retrospective, multicentre study including pSS patients fulfilling American–European Consensus Group criteria or enlarged American–European Consensus Group criteria, and with biopsy-proven renal involvement.

Results. A total of 95 patients were included (median age 49 years). An estimated glomerular filtration rate (eGFR) of <60 ml/min was found in 82/95 patients (86.3%). Renal biopsy demonstrated tubulointerstitial nephritis (TIN) in 93 patients (97.9%), and frequent (75%) plasma cell infiltrates. Glomerular lesions were found in 22 patients (23.2%), mainly related to cryoglobulin. The presence of anti-SSA (76.8%) and anti-SSB (53.8%) antibodies was particularly frequent among patients with TIN and was associated with a worse renal prognosis. Eighty-one patients (85.3%) were treated, with CSs in 80 (98.8%) and immunosuppressive agents (mostly rituximab) in 21 cases (25.9%). Despite marked interstitial fibrosis at initial

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biopsy, kidney function improved significantly during the 12-month period following diagnosis (final eGFR 49.9 *vs* 39.8 ml/min/1.73 m^2 at baseline, $P < 0.001$). No proven benefit of immunosuppressive agents over steroid therapy alone was found in this study.

Conclusion. Renal involvement of pSS is mostly due to TIN with marked T, B and especially plasma cell infiltration. Renal dysfunction is usually isolated but can be severe. Use of CSs can improve the eGFR, but further studies are needed to define the best therapeutic strategy in this disease.

Key words: primary Sjögren's syndrome, tubulointerstitial nephritis, cryoglobulinaemia

- Renal involvement during primary SS is characterized by tubulointerstitial nephritis, with frequent plasma cell infiltrates.
- Anti-SSA/SSB antibodies are frequent in tubulointerstitial nephritis and associated with worse renal prognosis in primary SS.
- CSs can improve renal function in primary SS, and no proven benefit of immunosuppressants over steroids was found.

Introduction

Primary SS (pSS) is a chronic autoimmune disorder characterized by lymphocytic infiltration of exocrine glands, resulting in significant loss of secretory function, with oral and eye dryness. The course of the disease is also characterized by the occurrence of extraglandular features, with frequent multi-organ involvement, including the kidney. However, overt renal disease is a rare event during pSS, and its prevalence was found to be around 3–5% in most recent series [1–3]. The main types of pSS-associated nephropathies previously described are tubulointerstitial nephritis (TIN) and membrano-proliferative GN (MPGN) related to cryoglobulinaemia. In addition, electrolyte disturbances such as renal tubular acidosis are frequent when appropriately screened [4]. Therefore, as tubulointerstitial involvement is sometimes insidious and slowly progressive, renal disease is probably underdiagnosed, a fact that may account for the low reported prevalence in large pSS retrospective cohorts [1, 5]. Very few reports of biopsy-proven renal disease in pSS have been published so far: the largest recent studies having reported 41 patients in China [6], 33 in Europe [1] and 24 in the USA [7]. For this reason, treatment of pSS-related nephropathy is not codified, and data in the literature are lacking.

In the present study, we conducted a French nationwide analysis of 95 biopsy-proven pSS-related nephropathies. We describe here the clinical and biological presentation of pSS-associated renal disease, the renal pathology data, and the response to treatment.

Methods

Study population

This retrospective study, the REnal INvolvement in SS study, was conducted in French universities and general hospitals across departments of Internal Medicine, Nephrology and Rheumatology. The study was performed in accordance with the ethical standards of the Declaration of Helsinki and was approved by the institutional review board of Hôpital Européen Georges Pompidou. We used electronic newsletters and emails to contact practitioners registered on the Société National Française de Médecine Interne, the Société de Néphrologie-Société Francophone de Dialyse and the Club Rhumatismes et Inflammation (section of the French Society of Rheumatology) websites, and screened the French Assessment of Systemic Signs and Evolution in SS (ASSESS) database [3]. Inclusion criteria were: pSS diagnosis, based on the American–European Consensus Group (AECG) criteria [8] or the enlarged AECG criteria, defined by the presence of three or more of four AECG items among subjective oral dryness, subjective ocular dryness, anti-SSA/SSB positivity and postivie minor salivary gland biopsy results, as previously described [9]; and presence of nephropathy, based on renal biopsy findings. Renal biopsy was performed for clinically relevant renal symptoms according to local practice. We excluded secondary SS associated with RA, SLE, scleroderma or PM. Positivity of anti-DNA antibodies, chronic hepatitis C, chronic HIV infection, sarcoidosis and cervical radiotherapy were considered as exclusion criteria.

Patients' characteristics

Practitioners in charge of the patients collected clinical and biological data at diagnosis of renal involvement and during follow-up. EULAR SS Disease Activity Index (ESSDAI) was recorded at the time of nephropathy diagnosis [10]. Data were retrospectively collected and centrally reviewed, including the renal pathology findings. Kidney function was assessed by the estimated glomerular filtration rate (eGFR) using the Modification of Diet in Renal Disease Study equation [11], and renal dysfunction was defined as an eGFR of < 60 ml/min/1.73 m^2. Chronic kidney disease (CKD) was defined by the presence of a renal dysfunction for at least 3 months and acute kidney

injury (AKI) as an eGFR decrease of > 25%, known or presumed to have occurred within the prior 3-month period.

Response to therapy

The assessment of renal response to therapy was based on the evolution of eGFR during follow-up. A renal response was defined by an improvement of eGFR >20%, compared with baseline values.

Interstitial fibrosis quantification and characterization of renal cellular infiltrate

Kidney biopsy specimens were centrally collected when possible for additional analyses, including computer-assisted quantification of cortical interstitial fibrosis, using segmentation method and colour image analysis, as previously described [12]. Interstitial leucocyte infiltrate was quantified (in a double-blinded manner using semi-quantitative analysis on Masson's trichrome slides) for 25 patients. Characterization of the interstitial cellular infiltrate was also performed for 20 patients by imunohistochemistry (using anti-CD3, anti-CD19 and anti-CD138 primary antibodies for T cell, B cell and plasma cell detection, respectively). These patients were randomly selected and did not differ from the patients of the entire cohort (supplementary Table S5, available at *Rheumatology* Online). Cellular infiltration was characterized as predominant when it represented the majority of the cells infiltrating the kidney slide.

Statistical analyses

Data are expressed as mean (S.D.) or median (range) for continuous variables, and as number (percentage) for non-continuous variables, respectively. Paired *t*-tests were used to analyse the evolution of paired values, and Fischer tests to compare non-paired values. Baseline characteristics of patients according to the improvement of eGFR ⩾20% throughout follow-up were compared using the Chi-square test and the Wilcoxon rank-sum test for categorical and continuous variables, respectively. This 20% threshold corresponded to the median value of eGFR improvement in the cohort. We estimated the eGFR dynamics over time, accounting for the correlation among repeated measurements within each individual, through linear mixed models with random intercept and random slope**.** The best model (Akaike's criterion) was obtained with no slope change. We modelled eGFR dynamics for the first 60 months, during which the median number of eGFR measurements was 4 per subject (min = 2; max = 4). Slopes of eGFR were compared between the groups. Mean eGFR evolution was depicted by plotting the mixed model predictions. $P < 0.05$ was considered as statistically significant. Analyses were performed with STATA software (release 13; Stata Corp., College Station, TX, USA).

Table 1 Characteristics of patients with biopsy-proven pSS-related renal involvement

Number of patients	95
Demography	
Gender	86 F/9 M
Age, median (IQR)	49.0 (17.9–87.3)
pSS diagnosis, *n* (%)	
AECG criteria (*n* = 95)	82 (86.3)
Enlarged criteria only (*n* = 95)	13 (13.7)
Other autoimmune diseases associated with pSS (*n* = 95)	11 (11.6)
Dry eyes (*n* = 93)	91 (97.8)
Subjective (*n* = 93)	82[a] (88.2)
Objective (*n* = 65)	56 (86.2)
Dry mouth (*n* = 94)	89 (93.7)
Subjective (*n* = 94)	87[b] (92.6)
Objective (*n* = 42)	32 (76.2)
ESSDAI, median (IQR)	16.0 (9–41)
Comorbidities, *n* (%)	
Hypertension (*n* = 95)	27 (28.4)
Diabetes (*n* = 95)	7 (7.4)
Hypertension and diabetes (*n* = 95)	6 (6.3)
Immunological features, *n* (%)	
ANA (+) (*n* = 95)	85 (89.5)
Anti-SSA (+) (*n* = 95)	73 (76.8)
Anti-SSB (+) (*n* = 93)	50 (53.8)
Anti-SSA and SSB (−) (*n* = 95)	22 (23.2)
Salivary gland biopsy (+), Chisholm score ⩾3, (*n* = 85)	82 (96.5)
RF (+) (*n* = 82)	53 (64.6)
Cryoglobulinaemia (+) (*n* = 89)	26 (29.2)
Low C3 (<800 mg/l) (*n* = 75)	13 (17.3)
Low C4 (<150 mg/l) (*n* = 76)	27 (35.5)
Hypergammaglobulinaemia (⩾16 g/l) (*n* = 81)	55 (67.9)

IQR: interquartile range; AECG: American–European Consensus Group; ESSDAI: European SS Disease Activity Index. Missing data for [a]'two subjective ocular symptoms' and [b]'one subjective dry mouth'.

Results

Patient's characteristics

A total of 95 patients were included, including 86 women and 9 men, and the median agewas 49 years (range 17.9–87.3) at pSS diagnosis. pSS-related characteristics at diagnosis are summarized in Table 1. Eight-two patients (86.3%) met the AECG criteria, while 13 patients (13.7%) met the enlarged criteria. Another autoimmune disease was associated with pSS in 11 patients (11.6%): autoimmune hepatitis (*n* = 4), Hashimoto's thyroiditis (*n* = 3), primary biliary cirrhosis (*n* = 2), coeliac disease (*n* = 1) and myasthenia gravis (*n* = 1). All 22 patients (23.2%) with negative anti-SSA or SSB antibodies had positive minor salivary gland biopsy (focus score ⩾1). RF was present in 53/82 patients (64.6%) and cryoglobulinaemia in 26/89 (29.2%). Hypergammaglobulinaemia, defined as total gammaglobulin level above 16 g/l, was

Table 2 Characteristics of renal involvement at presentation

Number of patients	95
Age, median (IQR), years	50.9 (7–87.3)
Delay between renal symptoms and pSS, median (IQR), months	0 (−265.7, 352.1)
Delay between renal symptoms/pSS, *n* (%)	
A least 1 year before	10 (10.5)
Simultaneous (1 year)	55 (57.9)
1–5 years after	20 (21.1)
>5 years after	10 (10.5)
Renal dysfunction (eGFR <60), *n* (%)	82 (86.3)
Acute kidney injury	30 (31.6)
CKD	52 (54.7)
Proteinuria	
Presence, *n* (%)	25 (26.3)
Dosage, mean (S.D.), g/day	1.4 (0.3)
Isolated electrolyte disturbances, *n* (%)	17 (17.9)
Lithiasis, *n* (%)	9 (9.5)
Nephrocalcinosis, *n* (%)	5 (5.3)
CKD stages, *n* (%)	
II (⩾60 ml/min/1.73 m^2)	13 (13.7)
IIIa (45–59 ml/min/1.73 m^2)	16 (16.8)
IIIb (30–44 ml/min/1.73 m^2)	34 (35.8)
IV (15–29 ml/min/1.73 m^2)	23 (24.2)
V (<15 ml/min/1.73 m^2)	8 (8.4)
Unknown	1 (1.1)

IQR: interquartile range; CKD: chronic kidney disease.

found in 55/81 patients (67.9%). No measurement of serum IgG4 was available.

The median ESSDAI score at renal biopsy was 16 (9–41) (detailed in supplementary Table S1, available at *Rheumatology* Online). In association with renal involvement, the ESSDAI items reflecting 'biological activity' (78.9%) and 'articular involvement' (28.4%) were the most frequently present. Only 12 patients (12.6%) had isolated renal involvement.

Characteristics of initial renal presentation

The median age at diagnosis of renal involvement was 50.9 years (range 17–87). The characteristics of nephropathy are summarized in Table 2. Diagnosis of nephropathy was concomitant with pSS diagnosis in most cases (57.9%), and preceded pSS diagnosis in 10.5%. Renal test abnormalities included renal dysfunction in 86.3% of all included patients (CKD in 54.7%, AKI in 31.6%), followed by proteinuria (26.3%), isolated electrolyte disturbances (17.9%) (acidosis and hypokalaemia), nephrolithiasis (9.5%) and nephrocalcinosis (5.3%). At diagnosis, severe renal dysfunction was rare, with only 32.6% of patients displaying an eGFR below 30 ml/min/1.73 m^2 and only one patient requiring dialysis.

Analysis of kidney biopsy specimens

Kidney biopsies were performed between 1977 and 2013, including 81/95 (85%) biopsies after 2000. The mean number of glomeruli per specimen was 19 (range 2–63), with a median number of sclerotic glomeruli of 4 (range 0–32). Renal biopsy showed TIN in 93/95 (97.9%) cases, which was isolated in 73/95 (76.8%) patients. Glomerular lesions were found in 22/95 (23.2%) patients and were associated with interstitial cellular infiltration in 20/22 cases. Glomerular disease included polyclonal cryoglobulinaemia-related MPGN in 8 patients, and other glomerulopathies (such as membranous nephropathy or focal segmental glomerulosclerosis) in 14 cases, (Fig. 1). No feature suggestive of IgG4-related disease was noted.

Light microscopy examination of kidney biopsy among patients with TIN revealed that the cellular infiltrate was mainly composed of lymphocytes, but contained plasma cells in 45/66 (68%) cases. We were able to perform a centralized semi-quantitative analysis of the interstitial cellular infiltrate in 25 cases with TIN, and a qualitative immunohistochemistry analysis in 20 patients, whereas other specimens were lacking (Table 3). The degree of cellular interstitial infiltration varied from <25% of the total interstitial area to > 75% in some cases. Interstitial cellular infiltrates were positive for T cell markers in 20/20 (100%) patients, B cell markers in 19/20 (95%) and plasma-cell markers in 15/20 (75%) patients. T cell infiltration was predominant in 13/20 (65%), B cell infiltration in 2/20 (10%) and plasma-cell infiltration in 5/20 (25%) of kidney biopsies.

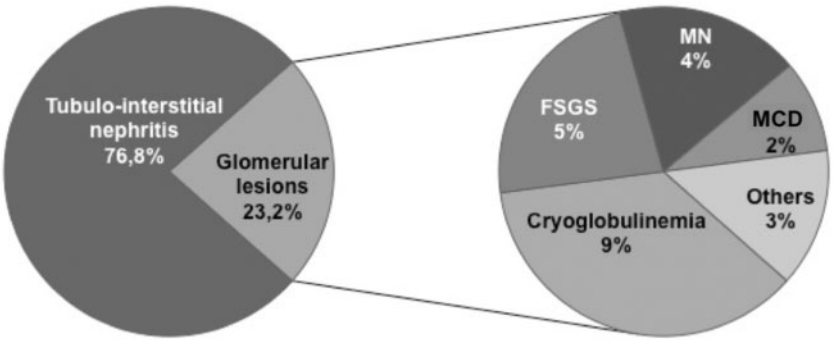
A computerized quantification of the cortical interstitial fibrosis was performed in 37 kidney biopsy specimens. The median fibrotic surface area was 27.1% (14.3–73), with interstitial fibrosis involving >25% of the total renal cortical area in 25 (67.6%) of all studied cases (Table 3).

Finally, correlations of immunological features with histological findings showed that patients with isolated anti-SSA antibodies (71.4 *vs* 28.6%, $P < 0.05$) and those with both anti-SSA/SSB antibodies (88 *vs* 12%, $P < 0.05$) more frequently had isolated TIN rather than pSS-associated glomerular disease (supplementary Table S2, available at *Rheumatology* Online).

Treatment

Treatment used for pSS-related nephropathy is summarized in supplementary Table S3, available at *Rheumatology* Online. Eighty-one patients (85.3%) received immunosuppressive treatment. Sixty patients received CSs alone, and 21 patients received CSs in combination with other immunosuppressive agents, mostly rituximab (RTX) ($n = 18$). The median initial daily dose of prednisone was 55 mg (range 5–80), and the median dose of prednisone was 10 mg/day (range 0–60) at 6 months, 8 mg/day (0–40) at 12 months and 0 mg/day (range 0–60) at the end of follow-up. Patients received RTX as first-line therapy in 12 cases and as second-line in 6. RTX was given at the dose of 1 g at days 1 and 15 for 10 patients, and at the dose of 375 mg/m^2/week during 4 weeks, for 8 patients. The mean ESSDAI was higher in patients treated

Fig. 1 Renal biopsy findings in the 95 patients with pSS



FSGS: focal and segmental glomerulosclerosis; MCD: minimal change disease; MN: membranous nephropathy; MPGN: membrano-proliferative glomerulonephritis (related to cryoglobulinaemia); TIN: tubulointerstitial nephritis.

Table 3 Histological characteristics in patients with pSS-related tubulointerstitial nephritis

Cortical area with significant interstitial cellular infiltration (n = 25) (%)	
<25%	5 (20)
25–50%	6 (24)
50–75%	7 (28)
>75%	7 (28)
Characterization of interstitial cellular infiltrate (n = 20)	
T-cell infiltration	20 (100)
T-cell predominant	13 (65)
B-cell infiltration	19 (95)
B-cell predominant	2 (10)
Plasma-cell infiltration	15 (75)
Plasma-cell predominant	5 (25)
Characterization of interstitial fibrosis (n = 37)	
Fibrosis, median (IQR), %	27.1 (14–73)
Fibrosis <25%	12 (32.4)
Fibrosis ⩾25%	25 (67.6)

All values listed as n (%) unless otherwise indicated.

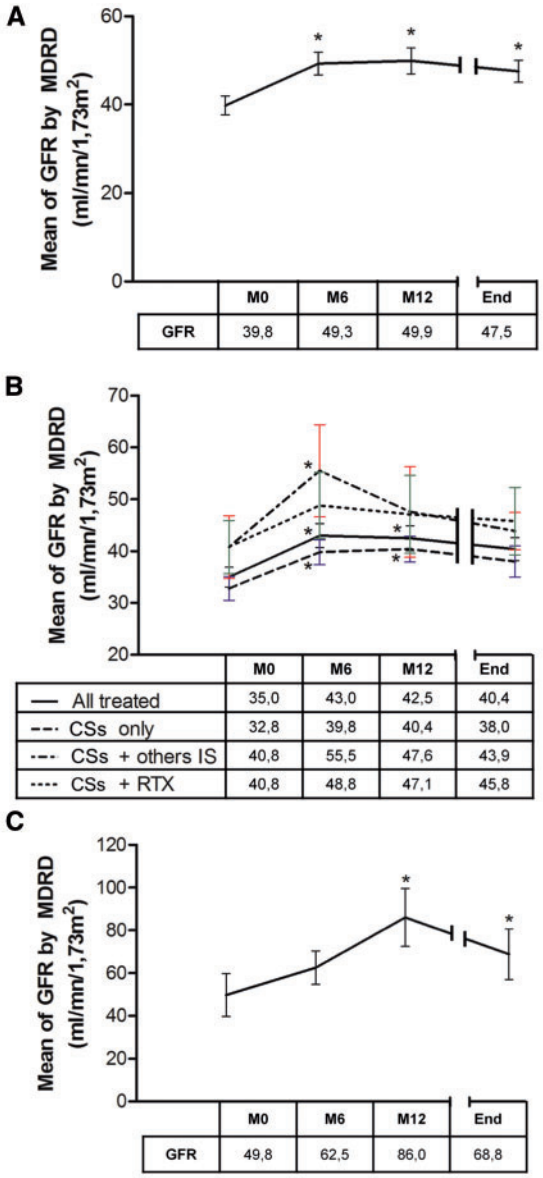
with RTX than in those not treated with RTX [22.6 (2.2) *vs* 17.3 (0.7); $P = 0.03$]. Fourteen patients (14.7%) received neither CSs nor immunosuppressive agents, mainly because of severe renal fibrosis considered to be irreversible in the majority of cases. Only one patient refused to receive CSs for the treatment of TIN.

Outcome

Renal function, as assessed by eGFR, was measured at baseline, 6 months (M6), 12 months (M12) and at the end of follow-up [median follow-up 60.4 months (range 12.1–174.7)]. In the entire population, renal function improved significantly, with mean eGFR increasing from 39.8 (2.1) ml/min/1.73 m^2 at presentation to 49.3 (2.6) ml/min/1.73 m^2 at M6 ($P = 0.0001$), 49.9 (3.0) ml/min/1.73 m^2 at M12 ($P = 0.00009$) and 47.5 (2.5) ml/min/1.73 m^2 ($P = 0.0003$) at the end of follow-up (Fig. 2A). Among the 64 patients with isolated TIN and at least two available eGFR values during follow-up, renal function significantly improved, with eGFR increasing from 35.0 (1.9) ml/min/1.73 m^2 to 43.0 (2.3) ml/min/1.73 m^2 at M6 and 42.5 (2.4) ml/min/1.73 m^2 at M12. Improvement of eGFR occurred regardless of the treatment used (Fig. 2B). In the eight patients with cryoglobulinaemia-related MPGN, mean eGFR also significantly improved at M12 and at last follow-up ($P = 0.03$) (Fig. 2C).

We next analysed the impact of the CSs/RTX combination, CSs/non-RTX combination and CSs alone in terms of achieving an eGFR gain of >20% compared with baseline, in the subgroup of patients with isolated TIN. In univariate analysis, renal improvement was more frequent among patients with severe initial renal dysfunction. Surprisingly, hypertension was associated with a good renal prognosis ($P = 0.02$). Positivity of anti-SSA or anti-SSB antibodies was associated with poor prognosis ($P = 0.01$ and 0.05, respectively). None of the pathology findings was associated with renal prognosis throughout follow-up (Table 4). There was no difference between the

FIG. 2 Evolution of renal function during follow-up



(**A**) Evolution of eGFR-MDRD in the whole population of patients. (**B**) Evolution of eGFR-MDRD in the 64 patients with TIN. (**C**) Evolution of eGFR-MDRD in the eight patients with cryoglobulinaemia-related MPGN. *$P < 0.05$ compared with baseline value (paired student *t*-test). eGFR : estimated glomerular filtration rate; IS: immunosuppressants; MDRD : Modification of Diet in Renal Disease Study; MPGN : membrano-proliferative glomerulonephritis; RTX: rituximab.

combinations of immunosuppressive agents plus CSs compared with CSs in univariate analysis, or in multivariate analysis, after adjustment for age at renal biopsy, hypertension and the presence of both anti-SSA or SSB antibodies (supplementary Table S4, available at *Rheumatology* Online). After modeling the eGFR dynamics over time, we found no slope difference between the CSs/RTX combination, CSs/non-RTX combination and CSs alone (supplementary Fig. S1, available at *Rheumatology* Online).

With a median follow-up of 60.4 months (range 12.1–174.7), only five patients (5%) reached end-stage renal disease, including four patients with isolated TIN and one with focal segmental glomerulosclerosis associated with TIN. During this period, two cases of lymphoma were reported: one breast cancer and one lung cancer. Both cases involved patients with cryoglobulinaemia. Five patients receiving immunosuppressive agents (including RTX in three cases) presented with severe infection. Five patients died: one from septic shock, one from lung cancer and three from unknown causes.

Discussion

Renal involvement related to pSS, although only slowly progressive, can lead to chronic renal dysfunction that can impact long-term prognosis. The present study, which is the largest series of biopsy-proven pSS-related nephropathies published so far, focused on the clinical, biological and pathological presentation of this complication.

In our study, pSS diagnosis was based on the AECG criteria, which were satisfied in the vast majority of cases. However, as objective measurement of sicca symptoms was missing at pSS diagnosis in a minority of patients, we choose to use the enlarged AECG criteria, based on the presence of $\geqslant 3$ out of the four items, including mandatory positive anti-SSA/SSB serology and/or positive minor salivary gland biopsy results [9]. In addition, to avoid analysis bias, we excluded patients with secondary SS, associated with other systemic rheumatic diseases. Our study population was homogeneous, with clear female predominance and a median age of 49 years, as expected in pSS patients without renal disease [13, 14]. Of note, a high proportion of patients presented with hypergammaglobulinaemia and/or cryoglobulinaemia, features consistent with an important B cell polyclonal activation.

This study provides important insights into the presentation of renal involvement during pSS. Among the most important findings, renal disease was diagnosed in the majority of cases either simultaneously or closely after pSS diagnosis, and presented as CKD, mild proteinuria and/or electrolyte disturbances. This presentation was in accordance with the tubulointerstitial lesions observed on kidney biopsies. Our results confirmed those of previous series, suggesting that the large majority of patients had TIN [1, 2, 7], a lesion that mimics minor salivary gland infiltration by both lymphocytes and plasma cells [15]. In contrast, glomerular disease was found in $< 25\%$ of cases, mainly related to cryoglobulinaemic MPGN. Although membranous nephropathy has previously been frequently associated with pSS [16], we observed only four such cases in our series.

Regarding correlations between immunological and histopathological features, we found that the presence

Table 4 Baseline characteristics according to the improvement or not of eGFR in patients with tubulointerstitial nephritis

Patient characteristics	No eGFR gain or a gain of $<20\%$ ($n = 32$)	eGFR gain of $\geqslant 20\%$ ($n = 32$)	P-value[a]
Demography and comorbidities			
Gender, female, *n* (%)	30 (94)	30 (94)	0.9
Age at diagnostic, median (IQR), years	46 (31–63)	54 (34–66)	0.4
Age at kidney biopsy, median (IQR), years	51 (35–65)	58 (33–66)	0.5
Hypertension, *n* (%)	3 (9)	11 (34)	0.02[b]
Diabetes, *n* (%)	1 (3)	3 (9)	0.5
pSS characteristics			
ESSDAI, median (IQR)	15 (12–18)	16 (12–20)	0.3
AECG criteria, *n* (%)	28 (88)	26 (81)	0.5
Enlarged criteria, *n* (%)	4 (13)	6 (19)	0.4
(i) Anti-SSA, *n* (%)	31 (97)	23 (72)	0.01
(i) Anti-SSA and -SSB, *n* (%)	25 (78)	17 (53)	0.05
Chisholm score $\geqslant 3$, *n* (%)	26 (82)	27 (84)	1
RF positivity, *n* (%)	21 (66)	15 (47)	0.3
Cryoglobulinaemia positivity, *n* (%)	9 (28)	7 (22)	0.5
Serum gammaglobulin, median (IQR), g/l	23 (17–32)	20 (16–38)	0.8
Hypergammaglobulinaemia (>16 *vs* <16 g/l), *n* (%)	26 (81)	25 (78)	0.8
Characteristics of renal involvement			
Delay between renal disease/symptoms of pSS, year, median (IQR)	1.5 (0.5–5.4)	1.0 (0.1–4.3)	0.3
(i) eGFR at inclusion, median (IQR), ml/min/1.73 m^2	40 (33–52)	30 (20–37)	0.002
Isolated electrolyte disturbances, *n* (%)	6 (19)	10 (31)	0.2
Lithiasis, *n* (%)	5 (16)	3 (9)	0.2[b]
Nephrocalcinosis, *n* (%)	4 (13)	1 (3)	0.2[b]
Proteinuria, median (IQR), g/24 h	0.5 (0.3–1.0)	0.5 (0.2–0.8)	0.5
P/C, median (IQR), g/mmol	0.07 (0.03–0.1)	0.05 (0.02–0.1)	0.4
Lymphocyte infiltration, *n* (%)	32 (100)	26 (81)	0.2
Plasmocyte infiltration, *n* (%)	23 (72)	16 (50)	0.5
Fibrosis $>25\%$ *vs* $\leqslant 25\%$, *n* (%)	8/13 (62)	13/18 (72)	0.9
Degree of cellular infiltration $<50\%$ *vs* $\geqslant 50\%$, *n* (%)	9/21 (43)	12/21 (57)	0.08[b]

[a]Baseline characteristics were compared using the Chi-square test and the Wilcoxon rank-sum test for dichotomous and continuous variables, respectively. [b]The Fisher exact test was used as appropriate. IQR: interquartile range; AECG: American–European Consensus Group; ESSDAI: European SS Disease Activity Index; eGFR: estimated glomerular filtration rate; P/C: protein/creatinine ratio; pSS: primary SS.

of anti-SSA and particularly anti-SSB antibodies was very frequent in pSS interstitial nephropathy, suggesting that these pSS patients warrant careful renal function work-up. Of note, anti-SSA and anti-SSB antibodies were, respectively, found in 76.8 and 53.8% of our patients, whereas their prevalence was usually roughly 55% for anti-SSA and 33% for anti-SSB, in previous large epidemiological studies [3]. Importantly, the presence of anti-SSA and/or anti-SSB is often considered as a marker of disease activity in pSS [17, 18].

Our study is one of the first to provide a detailed analysis of the cells infiltrating the kidney in pSS-related TIN. As reported in salivary glands [15], we found that T cell and B cell infiltration was a constant feature within the kidney. Evans *et al.* [19] recently showed in 12 patients with TIN that the interstitial infiltrate was predominantly a $CD4^+$ T cell infiltrate. In contrast to the case in other causes of TIN, such as immune-allergic, infectious or sarcoidosis TIN, plasma cells were found in 75% of pSS-related TIN cases and were even predominant in 25% of biopsies. Of note, none of our patients had features consistent with IgG4-related renal disease, that is, presence of storiform fibrosis or pseudotumoral infiltration of the kidneys [20]. Therefore, clinicians should be aware that the presence of plasma cells in the setting of TIN is very suggestive of pSS-related nephropathy. Finally, these findings suggest that specific therapies targeting B cells or plasma cells could be relevant, presuming that those cellular populations have a pathogenic role in this disease.

One of the main objectives of this study, despite its retrospective nature, was to evaluate the impact of therapeutic intervention on renal function. In contrast to previous findings by Goules *et al.* [5] that TIN was benign, four of our patients with isolated TIN reached end-stage renal disease. Our results are consistent with those of Maripuri *et al.* [7], who showed that treatment with glucocorticoids or other immunosuppressive agents can hamper the progression of renal disease over time. In our study, the mean

GFR improved from 39.8 at baseline to 49.9 ml/min/1.73 m^2 at 1 year. The improvement in eGFR after treatment may be considered as modest, but this limitation is probably explained by the high level of renal fibrosis observed at presentation, reflecting the silent and insidious progression of TIN. It is important to underline that more than two-thirds of the renal biopsies (analysed with computerized quantification of fibrosis) had >25% interstitial fibrosis, a threshold that is considered to be a strong prognostic marker, associated with poor renal prognosis in most nephropathies.

We observed that patients with more severe initial renal dysfunction were more prone to experience good renal response, especially patients with cryoglobulinaemia-related MPGN (Fig. 2C). This is most likely due to an earlier diagnosis for patients with proliferative glomerular disease, who will usually present with hypertension, high-range proteinuria and acute renal failure, in contrast with TIN patients who present with low-range proteinuria, no hypertension and slowly progressive renal failure. Along this line, the finding that hypertension was weakly associated with a good renal prognosis is probably related to associated confounders, such as better medical surveillance or the use of anti-hypertensive therapies. Of note, we were neither able to correlate the severity of interstitial fibrosis, nor the characteristics of the cellular infiltrate (i.e. B cell or plasma cells predominant in the infiltration) with the response to therapy, probably because of the small number of samples in which precise characterization was done. Interestingly, presence of anti-SSA or anti-SSA/SSB antibodies was associated with poor renal outcome.

A large majority of patients included in this study received CSs, and it is not possible to draw definitive conclusions concerning the efficacy of this treatment in pSS TIN when compared with supportive therapy, since the exact course of TIN patients without steroid treatment cannot be anticipated. Importantly, we found no additional benefit of immunosuppressive agents or RTX in comparison with CSs alone in patients with TIN. Among the limitations of our study, we must therefore underline the fact that immunosuppressive agents or RTX were administered according to the clinician's decision, in the absence of therapeutic guidelines and probably more frequently for patients with severe renal disease.

The efficacy of RTX in pSS is still controversial, as the TEARS trial failed to demonstrate any efficacy of this drug. However, patients included in this trial had low disease activity, and the primary outcome concerned fatigue, dryness and pain, but not the evolution of systemic involvement [21]. In contrast, a retrospective registry showed good efficacy and safety of RTX on pSS systemic manifestations, including kidney involvement [22].

Overall, these findings suggest that CSs alone seem to be an acceptable therapeutic option as first-line therapy in patients with TIN; given the prognosis, they are possibly better than supportive treatment alone. Immunosuppressive agents or RTX should probably be used in patients with refractory or relapsing disease, as well as in patients with cryoglobulinaemic GN [23]. Moreover, since B-cell or plasma-cell infiltration is frequently found in pSS TIN, B-cell targeting agents such as RTX could be evaluated in CS sparing strategies.

In conclusion, our cohort may represent a subgroup of pSS patients, since renal involvement was severe enough to prompt a kidney biopsy. However, our study demonstrates that TIN represents the more frequent renal manifestation of pSS. Presence of either anti-SSA or anti-SSA/SSB antibodies is predictive of both the risk of developing TIN, but also of experiencing poor renal outcome. Our data reveal that pSS-related TIN is characterized by frequent and sometimes predominant plasma-cell infiltration of the kidney, an observation that should be highly suggestive of pSS diagnosis. Finally, renal function significantly improved with treatment, although we found no clear benefit of immunosuppressive agents or RTX in comparison with CSs alone. Prospective and controlled trials are thus needed to define the place of immunosuppression and especially B-cell targeting agents in pSS-related TIN.

Acknowledgements

The authors thank all the clinicians and pathologists who participated in the REINSS study: C. Agard, A. Audemard, S. Bally, J.J. Boffa, D. Le Thi Huong-Boutin, P. Cacoub, P. Cathebras, G. Choukroun, E. Daugas, J.G. Fuzibet, C. Garrouste, M. Hamidou, P.Y. Hatron, D. Joly, J.E. Kahn, B. Knebelmann, T. Kofman, B. Laurent-Pilonchery, C. Lavigne, G. Le Guenno, T. Lobbedez, J. Schmidt, C. Sordet, A. Barbier-Dupas, A. Guillaudeau, A. Moreau, B. Ambrosetti, B. Mougenot, B. Mac-Gregor, C. Deminiere, C. Guilbeau-Frugier, C. Mussini, D. Droz, D. Desvaux-Belghiti, F. Comoz, H. Perrochia, H. Beaufils, J. Verine, L.H. Noel, J.P. Duong Van Huyen, A. Modesto, L. Marcellin, M. Mignon-Conte, D. Nochy, N. Patey-Mariaud De Serre, P. Rouvier, S. Lepreux, V. Algagnac-Oskman, M.-C. Copin, B. Gosselin, B. Delisle, F. Paraf, I. Brocheriou, J.-P. Saint-Are, J.-L. Kermeny, P. Callard. Author contributions: M.J., A.K., V.L.G., X.M., E.T., H.F. and B.T. designed the study. M.J., A.K., V.L.G., R.M., S.F., N.J.C., A.L.F., L.C., E.D., G.M., J.B.F., E.L., P.J., E.H., A.L.Q., P.R., A.H., N.C.C., P.R., P.V., V.M.Y., A.M.S., C.C., S.F., L.D., R.S., X.M., E.T., H.F. and B.T. collected data. M.J., E.K. and H.F. performed the statistical analysis. M.J., H.F., V.L.G., X.M., E.T., A.K. and B.T. interpreted the data. M.J., A.K., V.L.G., H.F. and B.T. wrote the manuscript.

Funding: No specific funding was received from any bodies in the public, commercial or not-for-profit sectors to carry out the work described in this manuscript.

Disclosure statement: R.S. has received consulting fees from Roche of <5000?. All other authors have declared no conflicts of interest.

Supplementary data

Supplementary data are available at *Rheumatology* Online.

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